A SIMPLE VOLUMETRIC METHOD FOR PRIMARY PRODUCTION MEASUREMENTS IN SEDIMENTS

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1. Introduction

Microphytobenthos assemblages abundant in shallow sea water, often play an important role in organic matter production and regulation of fluxes of compound at the sediment-water interface. In investigations of photosynthetic activity of microbenthic algae the following methods are commonly applied: ^{14}C fixation, oxygen exchange e.g. incubation techniques with transparent and dark chambers connected with Clark's oxygen electrodes, and recently oxygen microprofile technique which requires the application of oxygen microelectrodes with a sensing tip of a few microns (1). Each method has its advantages and disadvantages (2, 3). In the present studies a simple microvolumetric method was applied. Since the number of studies concerning sublittoral microphytobenthic production remains more scarce than the intertidal, this work concerns a spatial study of the microphytobenthic production in the Gulf of Fos down to a depth of 12 m, which represents more than 50% of the Gulf area.

2. Material and Methods

Samples of sediments were collected by diving at eleven stations with sediments composed of fine sands (0.1; 0.5 and 1 m depth) and muddy sands (1.5; 2; 3; 4; 5; 6; 8 and 12 m depth) along a transect in the Carteau Bay, a shallow cove (average depth 4 m) located in the Gulf of Fos (43°25' N; 4°56' E, French Mediterranean Coast). The top layer of sediments (0-1 cm) was cut out from the core of 2.1 cm diameter and freeze-dried. Pigments (chlorophyll *a* and pheopigments) were extracted for 24 h with 90% acetone and measured according to (4). Organic carbon, total nitrogen and sulphur contents were determined on a CHN analyzer. The measurements of dark respiration and photosynthesis at saturated irradiance of PAR (650 μmol photons$\cdot m^{-2}\cdot s^{-1}$) were performed in September 1995. It was found that the optimum irradiance for the highest net and gross production in sediments form the Gulf of Fos is around 200 μmol photons$\cdot m^{-2}\cdot s^{-1}$ (5) and many literature data indicate that photosynthesis of microbenthic algae becomes light-saturated at irradiance above 600 μmol photons$\cdot m^{-2}\cdot s^{-1}$ (6). The gas exchange rates were determined by the microvolumetric method described in detail by

(7). This method enables evaluation of gas exchange (photosynthesis and respiration) in small biological objects, and allows the estimation of oxygen production or consumption at rates varying from 10^{-1} to 10^{-4} $\mu l\ O_2 \cdot h^{-1}$ with an accuracy of ±5% (7). The small subsamples taken from the top layer of sediment (0-2 mm) were placed on the bottom of measuring chambers whereas carbonate buffer was settled as a suspending drop (Fig. 1). All measurements were made at 18°C, near the same, as it was in situ at the time of collection and at constant and relatively high CO_2 level (0.03% in the gas phase of the measuring chambers maintained by Warburg no. 10 carbonate buffer). The measurements were made immediately after collecting sediments from the field, at least in 4 replicates.

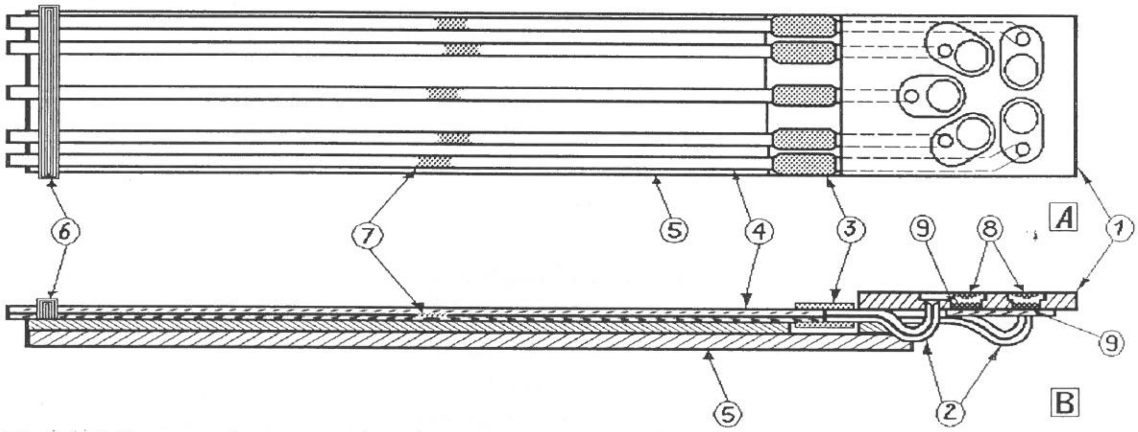

Fig. 1. Microrespirometer chamber systems: A - view from above; B - cross-section; 1 - brass plate with five microchambers; 2 - capillaries; 3 - plastic tubes; 4 - measurement capillaries; 5 - mounting base; 6 - comb; 7 - small column of coloured kerosene; 8 - carbonate buffer as a suspending drop; 9 - samples of sediment on the bottom of measuring microchambers.

3. Results and Discussion

The most important features of the superficial (first cm) sediments at the stations studied in the Gulf of Fos and microphytobenthic rates of gas exchange determined in surface layer (0-2 mm) were presented in the Table.

Depth [m]	NPP [dw]	NPP [chl*a*]	R [dw]	R [chl*a*]	GPP/R [dw]	Chl*a*	Pheop	Chl*a*/ Pheop	430/ 665	Corg	S
0.1	0.0164	2.509	-0.0094	-1.637	2.74	5.76	0.85	6.79	2.26	0.248	0.349
0.5	0.0171	2.829	-0.0032	-0.274	6.34	6.04	1.73	3.48	2.32		
1.0	0.0057	1.468	-0.0019	-0.654	4.11	3.87	1.14	2.89	2.54	0.092	0.098
1.5	-0.0240	-5.317	-0.0287	-7.473	0.16	3.84	6.44	0.54	3.65	0.674	0.339
2.0	-0.0646	-18.436	-0.0763	-21.750	0.15	3.51	9.71	0.36	4.19	0.655	0.827
3.0	-0.0421	-2.590	-0.0690	-4.240	0.39	16.28	35.70	0.46	3.43	1.587	1.058
4.0	-0.0263	-9.980	-0.0334	-12.680	0.21	2.63	12.70	0.21	3.45	0.737	0.464
5.0	-0.0044	-0.058	-0.0134	-0.325	0.67	41.24	91.05	0.46	3.18	1.098	0.468
6.0	-0.0123	-0.480	-0.0285	-2.103	0.57	13.54	28.05	0.48	2.95	0.756	0.346
8.0	-0.0083	-1.081	-0.0078	-1.033	0	7.63	28.90	0.26	3.56	1.773	0.782
12.0	0	0	-0.0054	-8.205		0.66	4.04	0.16	4.61	0.887	0.293

NPP[dw] - rates of net primary production expressed in $\mu l\ O_2 \cdot mg^{-1} dw \cdot h^{-1}$; NPP[chl*a*] - rates of net primary production normalized to chlorophyll *a* [$\mu l\ O_2 \cdot \mu g^{-1} chl a \cdot h^{-1}$]; R[dw] - dark respiration rates expressed in $\mu l\ O_2 \cdot mg^{-1}dw \cdot h^{-1}$; R[chl*a*] - dark respiration rates normalized to chlorophyll *a* [$\mu l\ O_2 \cdot \mu g^{-1}chl a \cdot h^{-1}$]; GPP/R[dw] - ratio of gross primary production to dark respiration [$\mu l\ O_2 \cdot mg^{-1}dw \cdot h^{-1}$]; Chl*a* - chlorophyll *a* contents in the top layer (0-1 cm) of sediments [$mg\ chl a \cdot g^{-1}dw$]; Pheop - concentration of pheopigments in the first cm of sediments [$mg\ pheop \cdot g^{-1}dw$]; Chl*a*/Pheop - ratio of chlorophyll *a* to pheopigments [$mg \cdot g^{-1}dw$]; 430/665 - ratio of OD at 430 nm to OD at 665 nm; C_{org} - organic carbon expressed in % sediment dry weight; S - total sulphur contents in the top layer (0-1 cm) of sediments [$mg\ S \cdot g^{-1}dw$]

At the stations located along the transect in Carteau Bay with an increase in depth characteristic changes in the gas exchange rate in the top layer of sediments could be observed. Only the most shallow 3 stations (0.1; 0.5 and 1 m) show positive values of net primary production and relatively low dark respiration rate. Consequently, the ratio of gross primary production to dark respiration is above 2. It was noted (8) that in the shallowest part of the Gulf of Fos (<1 m) the oxygen production of microphytobenthos exceeds that of phytoplankton. In sediments chlorophyll *a* prevails pheopigments and their ratio attains values of about 3 or more and organic carbon content is below 3 mg $C_{org} \cdot g^{-1}dw$. The stations of intermediate depths (1.5-4 m) show in the top layer negative, high values of net primary production and the highest dark respiration values. It could indicate intensive development of microorganisms. In sediments of these station organic carbon content is from above 6 to a dozen or so mg $C_{org} \cdot g^{-1}dw$. The above features observed at the stations of intermediate depths could be explained by intensive farming in the Carteau Bay covering an area of 0.053 km^2 at the depth 4-5 m. Mussel cultures on suspended ropes were shown to have a strong impact on benthic community by increasing organic matter and pigment contents in the sediments. Cumulative effects of biodeposition induce higher respiration rates (9). At deeper stations (5-8 m) in spite of the highest chlorophyll *a* and pheopigment concentrations, dark respiration rate and negative values of net primary production are low. At the deepest station (12 m) there was no net primary production. It is in accordance with the results of (8). Dark respiration (-0.0054 $\mu l\ O_2 \cdot mg^{-1}dw \cdot h^{-1}$) and pigment content in the sediment top layer were relatively low.

In the earlier investigations carried out in the Gulf of Fos no significant correlation was found between benthic primary production and chlorophyll *a* contents at spatial scale in all sublittoral sediment samples (5, 8, 9). Other authors have found significant correlations between chlorophyll *a* and gross primary production in sublittoral sediments. In the present studies such a correlation was found in the case of changes in gross primary production at different depths, normalized to chlorophyll *a* [$\mu l\ O_2 \cdot \mu g^{-1} Chl a \cdot h^{-1}$]. The linear model of gross primary production was: depth = 6.54 - 1.63·GPP and R = -0.815. Also the relationship between net primary production and 430/665 ratio (linear model: NPP = 0.10237 - 0.037 · ratio 430/665) and between gross primary production normalized to organic carbon [$\mu l\ O_2 \cdot \mu g^{-1} C_{org} \cdot h^{-1}$] and the ratio of chlorophyll *a* to chlorophyll *a* + peopigments (linear model: GPP = -0.0029 + 0.01469 · ratio chl*a*/pigments) in sediments are well correlated R = -0.901 and 0.9775, respectively. Gross primary production normalized to dry weight of sediments or chlorophyll *a* does not show a correlation at $p<0.05$ with the pigment content in sediment. Moreover, it was found a relationship between net primary production and dark respiration normalized to dry weight (linear model: NPP = 0.01061 + 0.91664 · Respiration) or to chlorophyll *a* (linear model: NPP = 1.7255 + 0.98329 · Respiration). These parameters showed a strong correlation, R = 0.939 and 0.983, respectively. Linear relationship between respiration and gross photosynthesis in the cyanobacterial mat was described by (10), R value was above 0.96.

Until now microvolumetric methods were not commonly applied in the investigations of primary production in benthic microalgae assemblages. The employed microvolumetric method, till now generally used in the measurements of gas exchange rate in small biological objects, enabled successfully to determine precisely photosynthetic and respiration rates in benthic microalgae. The measured values of primary production and respiration rates are in accordance with the literature data, also with the values noted in the Fos Bay. The measurements performed in that Bay by incubation techniques with transparent and dark chambers and oxymeter application (5, 8) showed that in autumn the net primary production rate at the station of 0.5 m depth is ca. 1.3 $\mu l\ O_2 \cdot \mu g^{-1} chl a \cdot h^{-1}$. Similar values were found in spring (1.29) and winter (1.1), and only in summer net primary production was lower, about 0.4 $\mu l\ O_2 \cdot \mu g^{-1} chl a \cdot h^{-1}$. In my experiments the data from sediment samples collected in autumn at the same depth (0.5 m) and place were twice as large, i.e. about 2.8 $\mu l\ O_2 \cdot \mu g^{-1} chl a \cdot h^{-1}$. However, it should be stressed that microrespirometer measurements enable saturation by PAR, whereas the production in situ at 0.5 m depth could not be measured in saturated conditions. The applied microvolumetric method appeared to be very effective. It is relatively simple, cheap, sensitive and makes it possible to test the same sediment sample at different temperatures or irradiances in the short time. Owing to the data obtained photosynthetic light curves commonly used in primary production modelling could be determined.

4. References

1 Revsbech, N.P. (1983) in Polarographic Oxygen Sensors (Gnaiger/Forstner ed.) pp. 265-273, Springer-Verlag, Berlin Heidelberg

2 Revsbech, N.P. Jørgensen, B.B. & Brix, O. (1981) Limnol. Oceanogr. 26, 717-730

3 Jönsson, B (1991) Limnol. Oceanogr. 36, 1485-1492

4 Lorenzen, C. (1967) Limnol. Oceanogr. 12, 343-346

5 Barranguet, C Cervetto, G & Fontaine M.-F. (1996) C. R. Acad. Sci. Paris, Sciences de la vie 319, 51-56

6 Blanchard, G.F. Guarini, J.-M. Gros, P. & Richard, P. (1997) J. Phycol. 33, 723-728

7 Zurzycki, J. & Starzecki, W. (1971) in Plant Photosynthetic Production. Manual of Methods (Šesták Z. Čatský J. & Jarvis P.G., eds) pp.257-275, Dr W. Junk N.V.Pub., The Hague

8 Barranguet, C. Plante-Cuny, M.R. & Alivon, E. (1996) Hydrobiologia 333, 181-193

9 Barranguet, C. Alliot, E. Plante-Cuny M.-R. (1994) Oceanol. Acta 17, 211-218

10 Epping, E.H. & Jørgensen, B.B. (1996) Mar. Ecol. Prog. Ser. 139, 193-203

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