

## Dissolved organic carbon production by microbial populations in the Atlantic Ocean

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### Abstract

Dissolved organic carbon (DOC) production by microbial populations was measured at 19 stations in the Atlantic Ocean to quantify the fraction of photoassimilated carbon that flows through the dissolved organic pool at basin scale and to assess the relationship between the percentage of DOC production, phytoplankton size structure, and rates of net community production. Experiments were conducted during four cruises carried out between May 1998 and October 1999, covering three upwelling regions: Benguela (SW Africa), Mauritania (NW Africa) and NW Spain, and the oligotrophic North Atlantic subtropical gyre between 30°N and 36°N. Photic zone integrated particulate organic carbon (POC) production rates ranged from 10 to 1,178 mg C m<sup>-2</sup> h<sup>-1</sup>, thus covering a wide productivity spectrum. The percentage of DOC production with respect to total integrated primary production ranged from 4 to 42%, being larger in oligotrophic, picoplankton-dominated waters, where a balanced metabolism of the microbial community was observed, than in productive, net autotrophic waters, where large-sized cells formed the bulk of the phytoplankton biomass. A highly significant relationship was calculated between DOC and POC production rates in upwelling conditions. By contrast, the relationship between these variables in oligotrophic environments was weak, which suggests that different processes could be controlling the release of dissolved organic matter in productive and unproductive waters.

Dissolved organic matter (DOM) is one of the least understood pools of marine matter and represents a major reservoir of organic carbon in the ocean. A large fraction of the dissolved organic carbon (DOC) present in the ocean ultimately derives from primary producers. However, a great deal of controversy still exists on the ecological significance and the ultimate control of DOC production in the ocean.

The magnitude of DOC-related fluxes remains still largely uncertain, especially in oligotrophic regions. The high rates of DOC uptake by heterotrophic bacteria in relation to primary production recently measured in unproductive waters (e.g., Hansell et al. 1995; del Giorgio et al. 1997), largely justifies the growing biogeochemical interest of measuring and modeling DOC production in planktonic ecosystems.

Initially, the radioactive carbon method for primary production estimation was modified for the measurement of direct excretion of dissolved organic compounds from algal cells. More recently, it has been recognized that several processes, besides direct excretion from intact algal cells, are

involved in the appearance of phytoplanktonic material in the dissolved organic compartment, such as cell lysis or microzooplankton grazing (Nagata 2000 and references therein). It is therefore fundamental to redefine the commonly employed term PER (percentage of extracellular release) as the fraction of recently photoassimilated carbon that flows through the DOC pool as a result of direct excretion from intact algal cells and natural processes ending in cell breakage. The technique generally employed for the measurement of DOC production rates by natural microbial populations is based on the release of labeled dissolved organic materials and does not allow distinguishing among the DOC produced by these processes.

According to Fogg (1983), DOC production typically represents from 5 to 40% of total primary production in nutrient-rich and oligotrophic waters, respectively, suggesting an inverse relationship between the fraction of dissolved organic carbon released and the magnitude of total photosynthetic carbon incorporation. Conceptually, higher losses of photosynthesized materials to the DOM pool would be expected in stratified oligotrophic environments as a consequence of the intrinsic functioning of microbial food webs (e.g., Kiørboe 1993; Legendre and Le Fèvre 1995; Azam 1998). By contrast, Baines and Pace (1991) concluded that the percentage of extracellular release (PER) was constant, averaging 13% of total carbon fixation, along the marine productivity gradient.

In this investigation, we have measured DOC production rates over a wide range of productivity levels (integrated particulate organic carbon [POC] production rates ranging from 10 to 1,178 mg C m<sup>-2</sup> h<sup>-1</sup>) and phytoplankton size structures (POC production by <2 μm phytoplankton ranging from 4 to 67%). Our aim was to elucidate the functional relationship between DOC production; planktonic community structure, as inferred from phytoplankton size; and phytoplankton-mediated carbon and oxygen fluxes (size-fractionated primary production and net community production).

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Table 1. Sampling regions. *n*, number of stations sampled at each sampling region.

Cruise	Research vessel	Date	Region	<i>n</i>
AMT-6	RRS <i>James Clark Ross</i>	22 May 1998	Benguela	1
AMT-6	RRS <i>James Clark Ross</i>	6 Jun 1998	Mauritania	1
Azores-1	<i>BIO Hespérides</i>	8–17 Aug 1998	North Atlantic subtropical gyre	8
OMEX-898	R/V <i>Professor Shtokman</i>	1–11 Aug 1998	NW Spain	5
OMEX-1099	R/V <i>Thalassa</i>	14–20 Oct 1999	NW Spain	4

## Methods

**Sampling regions**—The rate of DOC production by microbial populations was measured at 19 stations during four cruises carried out in the Atlantic Ocean between May 1998 and October 1999 covering three upwelling regions: Benguela (SW Africa), Mauritania (NW Africa) and NW Spain, and the oligotrophic North Atlantic (NA) subtropical gyre between 30°N and 36° N (Table 1; Fig. 1). Vertical profiles of temperature and salinity were conducted at each station with a Neil Brown Mark III conductivity-temperature-depth (CTD) profiler attached to a rosette sampler equipped with 10-liter Niskin bottles. The CTD conductivity sensor was calibrated using salinity determinations performed with an

Autosal salinometer on water samples drawn from selected depths.

**Size-fractionated chlorophyll *a***—Seawater samples (150–250 ml) were filtered sequentially through 5- and 2- $\mu$ m polycarbonate filters and Millipore APFF glass fiber filters, which were frozen (–20°C) immediately. Fluorescence due to chlorophyll *a* (Chl *a*) was measured ashore with a Safas flx spectrofluorometer after extraction in 90% acetone at 4°C for 24 h. Concentrations were calculated after calibration with pure extracts obtained by high performance liquid chromatography (HPLC). During the Atlantic Meridional Transect (AMT) cruise (Aiken et al. 2000), 200–300-ml seawater samples were filtered sequentially through 20- and 2-, and 0.2- $\mu$ m polycarbonate filters, and the concentration of Chl *a* was measured on a Turner 10-AU fluorometer calibrated with pure Chl *a* after extraction in 90% acetone at –20°C for 24 h.

**DOC production rates**—At each station three to five 30-ml samples were collected at selected depths from the photic layer, inoculated with 925 to 1,480 KBq (25 to 40  $\mu$ Ci) of  $\text{NaH}^{14}\text{CO}_3$  and incubated in an on-deck incubator, which simulated the irradiance experienced by the cells at the original sampling depths, for 2 h in order to minimize concurrent bacterial consumption of recently released DOC during the incubation period (see review by Fogg 1983). The bottles were kept refrigerated by pumping surface seawater into the incubator. After the incubation period, two 7–10-ml subsamples were drawn from each bottle and filtered by gravity through Millipore APFF glass fiber filters (equivalent to GFF-type filters). These type of filters are known to retain picoeukaryotes (e.g., Gasol and Morán 1999). Filtrates were acidified with 40–75  $\mu$ l of 50% HCl (until a pH around 2 was reached) and bubbled with  $\text{CO}_2$  free air for 12 h. Filters were decontaminated by exposing them to concentrated HCl fumes for 12 h. Scintillation cocktail was then added to both filters and filtrates. Duplicate blank tests were run in parallel by inoculating and immediately processing 0.2- $\mu$ m filtered seawater in the same way as mentioned before. Radioactivity was measured with an LKB  $\beta$ -scintillation counter. Quenching corrections were made using an external standard. Blank values were  $187 \pm 22$  disintegrations per minute (dpm, mean  $\pm$  SE) for all the experiments. The variation coefficient (SD/mean) for the measurements of DOC production rates was  $28 \pm 2\%$  (mean  $\pm$  SE).

Two recent papers have explicitly questioned the adequacy of using glass fiber filters for the measurement of POC and DOC production (Karl et al. 1998; Morán et al. 1999) due to  $^{14}\text{C}$ -DOC adsorption onto this type of filters; conse-

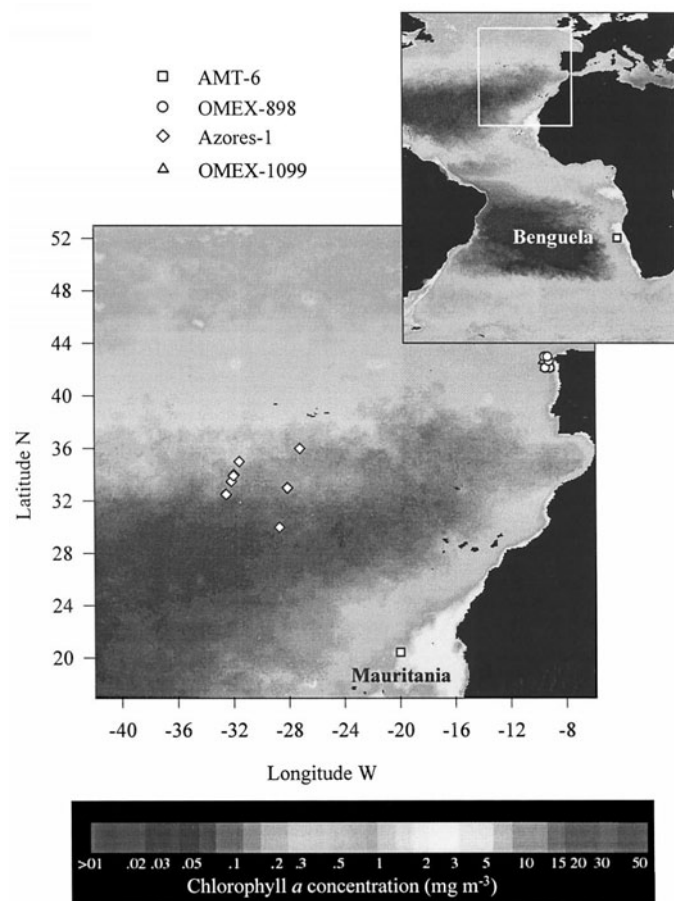


Fig. 1. Map of the stations sampled from May 1998 to October 1999 in the Atlantic Ocean (see also Table 1), superimposed on a SeaWiFS yearly composite image for 1998.

quently, DOC production rates reported here should be considered as conservative estimates.

*Size-fractionated POC production rates*—At each station four 75-ml acid-cleaned polypropylene bottles (three transparent bottles and one dark bottle) were filled with seawater from five or six depths corresponding to optical depths ranging from 100 to 1% of surface irradiance levels. Each bottle was inoculated with 185 to 555 KBq (5 to 15  $\mu\text{Ci}$ ) of  $\text{NaH}^{14}\text{CO}_3$  and then incubated for 6–7 h in the same on-deck incubator used for DOC production incubations. Previous studies did not show statistically significant differences between primary production estimates derived from on-deck and in situ incubation procedures (Joint et al. 1993). After the incubation period, samples were filtered at low vacuum pressure (<50 mm Hg) through the same type of filters described above for Chl *a* determinations. Filters were then decontaminated by exposing them to fumes of concentrated HCl for 12 h. Scintillation cocktail (3.5 ml) were then added to each filter, and the radioactivity was measured with an LKB  $\beta$ -scintillation counter. Quenching corrections were made using an external standard.

*O<sub>2</sub> production and respiration rates*—Oxygen production and consumption rates were determined by light- and dark-bottle incubations at four selected depths. During the OMEX and Azores-1 cruises, four light and eight dark, 125-cm<sup>3</sup> gravimetrically calibrated, borosilicate bottles were carefully filled from each depth, using silicone tubing, overflowing >250 cm<sup>3</sup>. An initial set of four dark bottles were fixed immediately for initial oxygen concentration, the remainder being kept under a 24-h light–dark diel cycle in the same on-deck incubator as for <sup>14</sup>C incubations. Fixing and storage procedures, reagents, and standardization followed the recommendations of Grasshoff et al. (1983). Dissolved oxygen concentration was measured through automated precision Winkler titration performed with a Metrohm DMS Titrino, using a potentiometric end-point detector as described in Serret et al. (1999). In the AMT-6 cruise, five replicates of 60-cm<sup>3</sup> borosilicate glass bottles were used, and measurements of dissolved oxygen were made with an automated photometric Winkler titration system based on that described in Williams and Jenkinson (1982), as described in Serret et al. (pers. comm.). Net community production (NCP) and dark community respiration (DCR) were estimated as the change in oxygen concentration in the light and dark bottles, respectively, after incubation. Gross oxygen production (GP) was calculated as the result of NCP minus DCR.

*Photic zone integration*—Photic zone integrated values of size-fractionated Chl *a*, POC production, DOC production, NCP, DCR, and GP were obtained by trapezoidal integration of the volumetric data down to the depth of 1% surface incident irradiance. Photic zone depth ranged from 30 m at upwelling stations to 120 m in oligotrophic subtropical waters.

## Results

*Oligotrophic versus upwelling conditions*—The stations visited in this study covered a wide and almost continuous

Table 2. Averaged values ( $\pm$  SE) for physical and biological variables during oligotrophic and upwelling conditions.

Variable	Oligotrophic ( <i>n</i> = 12)	Upwelling ( <i>n</i> = 7)
Surface temperature (°C)	23 $\pm$ 1	16 $\pm$ 1
Surface salinity	36.3 $\pm$ 0.2	35.7 $\pm$ 0.1
$\Delta\sigma_t$ (0–150 m)	1.8 $\pm$ 0.2	0.7 $\pm$ 0.1
Chl <i>a</i> (mg m <sup>-2</sup> )	18 $\pm$ 6	91 $\pm$ 15
% Chl <i>a</i> <2 $\mu\text{m}$	45 $\pm$ 7	11 $\pm$ 2

range of productivity levels. Nevertheless, they were grouped into oligotrophic or upwelling conditions for the sake of comparison (Table 2). The stations located in the NA subtropical gyre were considered oligotrophic and the stations located at the two African upwelling systems were considered nutrient-enriched. The stations located off the NW Spanish coast, sampled during the summer upwelling event (OMEX-898), were also considered nutrient-enriched, whereas those visited during autumn were considered oligotrophic (Teira et al. 2001). Oligotrophic stations showed higher surface temperature and salinity values than upwelling stations, which were characterized by advection of cold, nutrient-rich waters up to the surface. This translated into a relatively reduced gradient of  $\sigma_t$  measured between the surface and 150 m.

Photic zone integrated Chl *a* concentration was four- to fivefold higher at upwelling than at oligotrophic stations, and large cells (>2  $\mu\text{m}$ ) formed the bulk of phytoplankton (~90%) at these highly productive regions. Picoplankton cells (<2  $\mu\text{m}$ ) dominated under oligotrophic conditions (~50%), indicating that contrasting planktonic communities inhabited the environments we studied.

*POC versus DOC production rates*—POC production rates ranged from 0.04 mg C m<sup>-3</sup> h<sup>-1</sup> under oligotrophic conditions to 47 mg C m<sup>-3</sup> h<sup>-1</sup> in upwelling regions. Photic zone integrated POC production rates varied between 10 mg C m<sup>-2</sup> h<sup>-1</sup> at the subtropical gyre and 1,178 mg C m<sup>-2</sup> h<sup>-1</sup> during upwelling conditions, thus covering a wide range of productivity levels. DOC production rates ranged from 0.01 mg C m<sup>-3</sup> h<sup>-1</sup> in oligotrophic conditions to 2.81 mg C m<sup>-3</sup> h<sup>-1</sup> in the most productive environments, with photic zone integrated values ranging from 2 to 72 mg C m<sup>-2</sup> h<sup>-1</sup>. Thus, POC production rates were two orders of magnitude higher in upwelling than in oligotrophic conditions, whereas DOC production rates only varied by an order of magnitude, which suggests that DOC production was not a constant fraction of total organic carbon (TOC) production.

In order to validate this observation, we first investigated the relationship between DOC and POC production rates by using the whole data set. A statistically significant log-log linear relationship was found between the rates of DOC and POC production (Fig. 2) for both volumetric ( $P < 0.0001$ ) and depth-integrated ( $P < 0.0001$ ) data (Table 3). Model II slopes were also calculated—because error is inherent to both *x* and *y* variables—and are shown in Table 3. DOC production increased as a function of particulate production, but in the case of both volumetric and integrated data, the

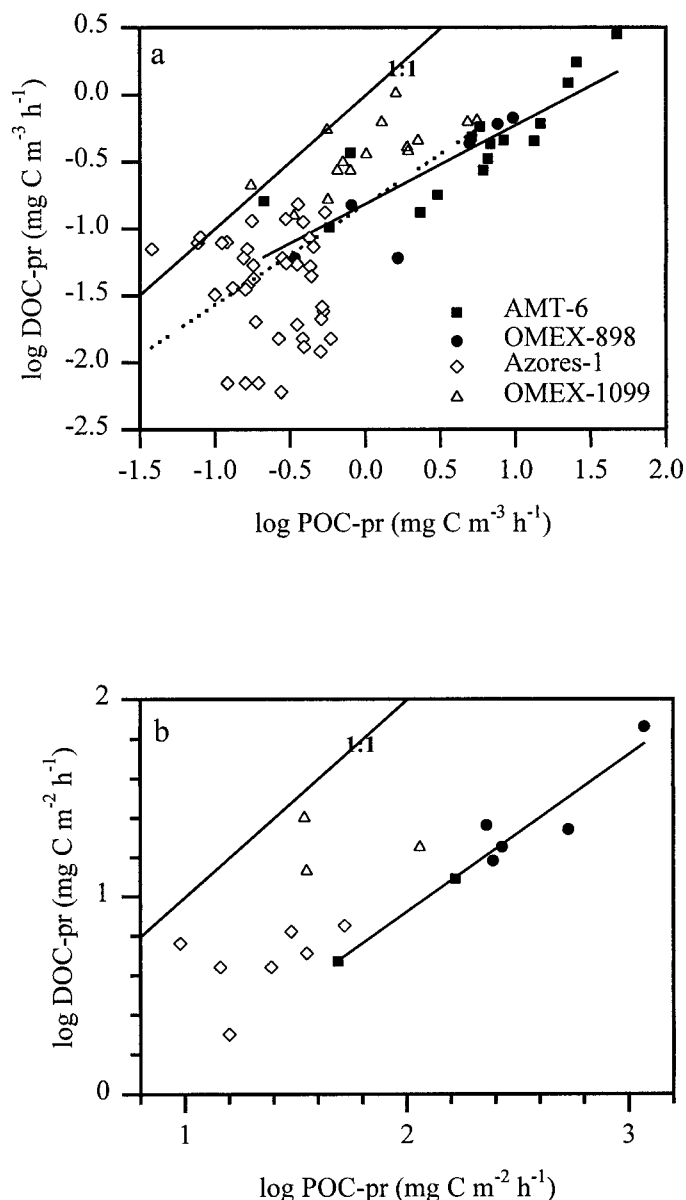


Fig. 2. Log-log relationship between DOC and POC production rates obtained using (a) volumetric and (b) photic zone integrated data. Open and solid symbols, as well as dotted and solid lines, correspond to oligotrophic and upwelling conditions, respectively. Regression parameters are given in Table 3.

slope of the regression line (0.67 and 0.53, respectively) was significantly  $<1$  ( $P < 0.00001$  and  $P < 0.0001$ , respectively). The deviation of the observed relationship from the line of slope 1 indicates that DOC production rate was not a constant fraction of the total amount of carbon incorporated by primary producers (*see* Baines and Pace 1991). However, model II slope estimates do not confirm such deviation from the line of slope 1 in the case of volumetric data, which complicates the interpretation of regression slopes.

When data deriving from oligotrophic and upwelling conditions were analyzed separately (Fig. 2), a highly significant

linear relationship was obtained between  $\log[\text{POC-pr}]$  and  $\log[\text{DOC-pr}]$  for upwelling regions, which explained more than 70 and 90% of the variability in volumetric and integrated DOC production, respectively (Table 3). By contrast, weak (for volumetric data) or not significant (for depth-integrated data) relationships were found when only the oligotrophic data set was considered. POC production only explained 33% of the variability in volumetric DOC production. It is worth noting that the equation relating integrated rates of DOC and POC production in the upwelling regions has a slope of 0.80 (model II slope 0.83), which is considerably higher than the slope obtained using the complete data set (0.53) and not significantly  $<1$  ( $P = 0.052$ ). The two regression models displayed similar results when the upwelling data set was used. However, they highly disagree for the oligotrophic data set, probably because of the weak relationship found between DOC and POC production rates in unproductive environments.

Our results show an apparent inverse relationship between DOC and POC production rates, probably arising from the high variation in PER measured at oligotrophic environments.

*PER, phytoplankton size structure, and rates of net community production*—Phytoplankton size structure, expressed in this work as the relative contribution of picoplankton cells ( $<2\text{-}\mu\text{m}$  phytoplankton) to POC production, usually has been referred to as a variable summarizing food web structure (Legendre and Le Fèvre 1991; Kiørboe 1993), whereas NCP could be considered a variable summarizing community energetics (Smith and Hollibaugh 1997 and references therein). When these two variables were represented against PER, a close association was found between high PER values and oligotrophic picoplankton-dominated environments where the trophic status of the planktonic system was heterotrophic (Fig. 3). A statistically significant exponential relationship was found between PER and either the percentage of POC-pr  $< 2\ \mu\text{m}$  and NCP. The arcsine transformation was performed in order to normalize percentages. The corresponding regression equations obtained were:  $\text{PER} = 7.9e^{0.03(\% \text{POC-pr} < 2)}$ ;  $r^2 = 0.83$ ,  $P < 0.001$ ,  $n = 10$ ;  $\text{PER} = 28.7e^{-0.004(\text{NCP})}$ ;  $r^2 = 0.69$ ,  $P = 0.02$ ,  $n = 7$ . PER and percentage of POC-pr  $< 2$  are in arcsine units. Phytoplankton size explained  $>80\%$  of the variability in the PER derived from the integrated data set, which might suggest the influence of trophic processes in the release of dissolved organic substances, mainly in oligotrophic conditions. In upwelling stations, where the contribution of picoplankton to total POC production ranged from 4 to 34%, PER was a rather constant value ( $\sim 7\%$ ).

We represented the rates of NCP and PER over a production/biomass plot (Tremblay and Legendre 1994) for all stations where these four variables were concurrently measured (Fig. 4). A continuous gradient was observed from highly productive, large phytoplankton-dominated environments where the production/respiration (P/R) balance was autotrophic and low PER values were measured to oligotrophic, picoplankton-dominated communities showing a relatively high PER and negative P/R balances.

Table 3. Summary of regressions fit to the model  $\log(\text{DOC}) = b \log(\text{POC}) + a$ . Model II slopes are  $b/r$ , where  $r$  is the square root of  $r^2$ .  $n$ , the number of observations; SE, standard error; CL 95%, 95% confidence limits.

Data set	$n$	Intercept $a$ (SE)	Slope $b$ (SE)	$r^2$	$P$	Model II slope (CL 95%)
Volumetric data						
All data	76	-0.86 (0.05)	0.67 (0.07)	0.55	<0.0001	0.90 (0.77–1.05)
Oligotrophic	54	-0.82 (0.09)	0.75 (0.15)	0.33	<0.0001	1.30 (1.04–1.63)
Upwelling	22	-0.82 (0.07)	0.59 (0.08)	0.72	<0.0001	0.69 (0.54–0.88)
Integrated data						
All data	17	0.03 (0.19)	0.53 (0.10)	0.67	<0.001	0.65 (0.48–0.89)
Oligotrophic	10	-0.12 (0.44)	0.66 (0.30)	0.39	0.055	1.05 (0.58–1.91)
Upwelling	7	-0.68 (0.26)	0.80 (0.11)	0.92	0.001	0.83 (0.60–1.14)

## Discussion

To our knowledge, this is the first empirical evidence that relates DOC production to the size structure of phytoplankton populations, although previous studies have focussed on the link between PER and phytoplankton size from a physiological perspective (e.g., Bjørnsen 1988; Malinsky-Rushansky and Legrand 1996). The significant relationships observed between PER and both phytoplankton size structure and rates of NCP might be interpreted as indicative of the importance of trophic-related processes on the production of DOC in the upper ocean.

*The DOC–POC production relationship*—The percentage of carbon flowing to the DOC pool measured in this study represented between 4 and 9% of total integrated primary production under upwelling conditions, characterized by high rates of integrated POC-pr, whereas in oligotrophic conditions, where lower integrated POC-pr values were measured, DOC production accounted for 11 to 42% of total carbon incorporation. The low and almost constant PER that we measured in upwelling conditions (Figs. 3, 4) has been consistently reported in the literature for productive oceanic environments, as well as for phytoplankton cultures (*see review in Nagata 2000*).

Since the recognition of the release of dissolved organic substances by phytoplankton as a considerable loss of photosynthate (Fogg et al. 1965), the phenomenon of extracellular release has been widely investigated, generating an active debate on the adequacy of the method and, more specifically, on the potential relationship between total primary production and PER. This latter point still remains a matter of controversy. As already mentioned above, most of the earlier investigations concluded that there was an inverse relationship between the amount of DOC released by microbial populations and the total amount of carbon incorporated photosynthetically (e.g., Berman and Holm-Hansen 1974; Mague et al. 1980; Morán 1999). However, different results emerged from the analysis carried out by Baines and Pace (1991), who concluded that PER was constant (on average 13%) along the productivity gradient characteristic of marine systems. Our results clearly support the hypothesis of an apparent inverse relationship between both variables as a consequence of the high variation in PER at low productive environments. It is important to stress the adequacy of our

data set for the analysis of the POC-pr vs. PER relationship, due to the large productivity gradient covered. Especially valuable are the measurements conducted at the oligotrophic subtropical gyre. The data set analyzed by Baines and Pace (1991), although covering a wide productivity gradient, was largely restricted to coastal marine environments. In this context, Nagata (2000) pointed out that the paucity of PER estimations in open ocean regions makes premature the use of a PER value of 13%, as estimated by Baines and Pace (1991), as a “global average” of PER for the marine environment.

Typically, phytoplankton exudation of organic matter was interpreted as an overflow mechanism of recently photosynthesized compounds that occurred during nutrient limitation (Fogg 1983) or as a mechanism of passive diffusion of organic compounds across the cell membrane (Bjørnsen 1988). Although it is not clear yet which is the actual mechanism of phytoplankton exudation, a highly significant positive relationship between DOC production rates and photosynthetic rates would be predictable. The linear regression between POC and DOC production obtained from our integrated data (Fig. 2) presented a slope significantly <1 (both for model I and model II slope estimates), which supports the inverse relationship between PER and total primary production, so that, as productivity increases, the production of DOC decreases in relative terms. A similar analysis was carried out by Morán (1999) who also found a statistically significant log-log linear relationship between POC and DOC production volumetric rates, obtaining a slope of 0.65, which is similar to that found in this study using the volumetric data set (0.67). On the other hand, the relatively low percentage of DOC-pr variability explained by volumetric and integrated POC-pr rates (55 and 67%, respectively), suggests that other processes apart from phytoplankton exudation are likely to be involved in the release of dissolved compounds by microbial communities in the natural environment. The results obtained when upwelling and oligotrophic data were compared clearly support the idea of different processes controlling extracellular release in these contrasting environments. The weak relationship observed between DOC and POC production in unproductive regions (volumetric POC-pr explained only 33% of DOC-pr variability) can be interpreted as indicative of the predominance of trophic (grazing, lysis) over physiological (exudation) processes in the release of dissolved organic materials, whereas in more productive

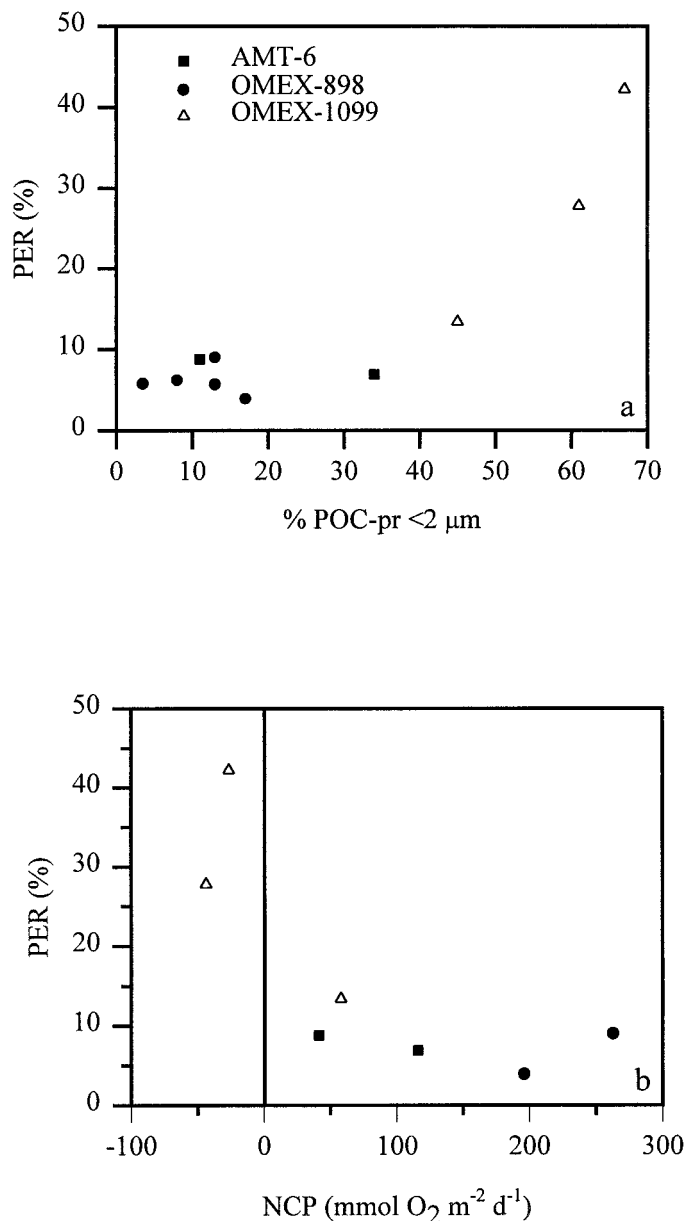


Fig. 3. Relationship between PER and (a) the relative contribution of cells  $<2 \mu\text{m}$  to POC production (% POC-pr  $< 2 \mu\text{m}$ ) and (b) rates of net community production (NCP). Symbols as in Fig. 2. Regression equations obtained after arcsine transformation are given in the text (lines not shown in the graphs).

regions, exudation appears to be the more relevant process (volumetric and integrated POC-pr explained 72 and 92% of DOC-pr variability, respectively); therefore, DOC production rates tend to be a constant and low fraction of the total amount of primary production (about 7%).

The hypothesis of different processes controlling DOC production at productive and unproductive environments is in good agreement with the conclusions reported by Sharp (1977), who first proposed that DOC release during periods of active phytoplankton growth was not a major source of DOC in the sea. This observation is consistent with the low averaged PER value measured during the upwelling condi-

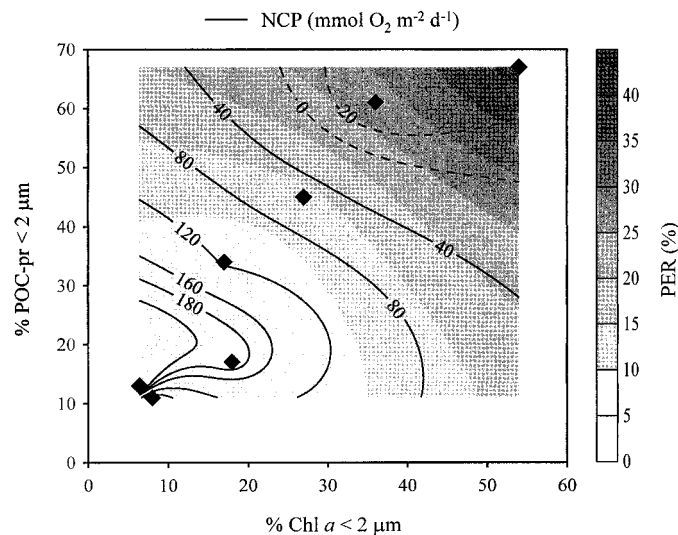


Fig. 4. Production/biomass plot of stations where size-fractionated Chl *a* and POC production, DOC production, and NCP rates were concurrently measured ( $n = 7$ ); *x* and *y* axes represent, respectively, the contribution of primary producers  $<2 \mu\text{m}$  to total phytoplankton biomass (expressed as Chl *a* concentration) and to POC production. Symbols represent combinations of these two variables at single stations. Superimposed shaded contours represent the relative contribution of photic zone integrated DOC production to total primary production (PER). Isolines represent photic zone integrated net community production rates ( $\text{mmol O}_2 \text{m}^{-2} \text{d}^{-1}$ ).

tions in our study. He also postulated that the degradation of old, dying phytoplankton as well as losses during food web transfer were the main internal sources of dissolved organic matter in the sea.

*DOC production, phytoplankton size, and community metabolism*—We calculated average carbon fluxes through the microbial community for the two contrasting scenarios studied: oligotrophic and upwelling regions (Fig. 5). Rates of carbon fixation were, on average, fivefold higher in upwelling regions, where the phytoplankton was dominated by large cells, than at the oligotrophic ocean, where picoplankton formed the bulk of the autotrophic standing stock. Absolute DOC production rates by microbial populations did not significantly differ between oligotrophic and upwelling conditions ( $P = 0.16$ ). However, the relative contribution of DOC released with respect to total primary production was, on average, more than threefold higher in oligotrophic ( $23 \pm 4\%$ ) than in upwelling ( $7 \pm 1\%$ ) conditions. The relatively higher contribution of DOC production to total photosynthesis measured in oligotrophic conditions was related to high relative rates of community respiration, leading to a nearly balanced system where most of the autotrophic production would be respired within the photic layer. On the contrary, in highly productive regions, the small fraction of recently photosynthesized carbon flowing to the DOC pool was associated with net autotrophic microbial metabolism.

Our results show that in oligotrophic environments for similar POC-pr values, DOC-pr is highly variable (Fig. 2), whereas 83% of PER variability across habitats was explained by changes in phytoplankton size structure (Fig. 3).

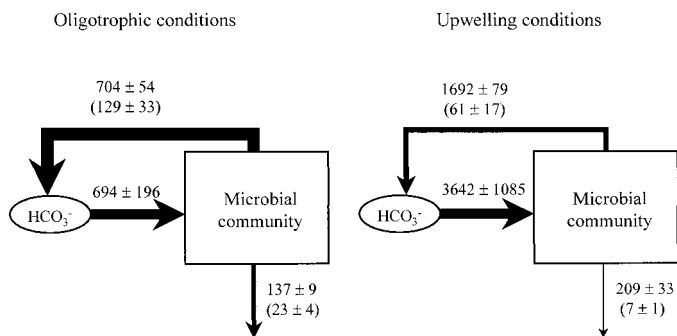


Fig. 5. Carbon flow through the microbial community obtained for the two contrasting environments studied in the present work: oligotrophic and upwelling conditions. Photic depth integrated fluxes are expressed in  $\text{mg C m}^{-2} \text{d}^{-1}$  (mean  $\pm$  SE). Daily rates were calculated from hourly rates utilizing the equation by Straskraba and Gnauck (1985). Oxygen fluxes were expressed in carbon units by using a respiration quotient  $\text{RQ} = 1$  (del Giorgio and Peters 1993). Only stations where all the fluxes were concurrently measured were used ( $n = 3$  for both oligotrophic and upwelling conditions). Numbers in parentheses and arrow thickness represent the relative contribution of any given flux with respect to TOC-pr rate.

Hence, not only the magnitude of carbon incorporation rates but also phytoplankton size structure should be considered in order to predict the magnitude of DOC production by microbial populations.

The close relationship found between PER, the rates of NCP, and phytoplankton size structure (Figs. 3–5), suggests that the observed balanced metabolism of the microbial compartment in the oligotrophic ocean could be a consequence of the intrinsic functioning of the planktonic food web. The higher losses of photosynthesized materials to the dissolved organic matter (DOM) pool as a result of processes such as phytoplankton cell lysis (Brussaard et al. 1995; Gobler et al. 1997; Agustí et al. 1998) or grazing by protozoa (Nagata 2000 and references therein), and the tight and complex trophic interactions characteristic of these systems imply that microbial food webs would constitute a nearly closed system (Legendre and Rassoulzadegan 1995 and references therein) where most of the autotrophic production would be respired in the photic layer (Legendre and Le Fèvre 1995 and references therein). By contrast, in highly productive systems, where the classical food web dominates, trophic processes implied in DOC production would be comparatively less important and the main source of DOC would be direct excretion from healthy cells, thus accounting for a constant and small (<10%) fraction of total primary production.

The results presented herein stress the need for concurrent measurements of DOC production, phytoplankton size structure, community respiration, cell lysis, and microzooplankton grazing under contrasting hydrographic conditions if we are to understand the relative importance of trophic vs. physiological processes on the production of dissolved organic matter in the oceans.

## References

AGUSTÍ, S., M. P. SATTÀ, M. P. MURA, AND E. BENAVENT. 1998. Dissolved esterase activity as a tracer of phytoplankton lysis:

Evidence of high phytoplankton lysis rates in the NW Mediterranean. *Limnol. Oceanogr.* **43**: 1836–1849.

AIKEN, J., AND OTHERS. 2000. The Atlantic Meridional Transect: Overview and synthesis of data. *Prog. Oceanogr.* **45**: 257–312.

AZAM, F. 1998. Microbial control of oceanic carbon flux: The plot thickens. *Science* **280**: 694–696.

BAINES, S. B., AND M. L. PACE. 1991. The production of dissolved organic matter by phytoplankton and its importance to bacteria: Patterns across marine and freshwater systems. *Limnol. Oceanogr.* **36**: 1078–1090.

BERMAN, T., AND O. HOLM-HANSEN. 1974. Release of photoassimilated carbon as dissolved organic matter by marine phytoplankton. *Mar. Biol.* **28**: 305–310.

BJØRNSSEN, P. K. 1988. Phytoplankton exudation of organic matter: Why do healthy cells do it?. *Limnol. Oceanogr.* **33**: 151–154.

BRUSSAARD, C. P. D., AND OTHERS. 1995. Effects of grazing, sedimentation and phytoplankton cell lysis on the structure of a coastal pelagic food web. *Mar. Ecol. Prog. Ser.* **123**: 259–271.

DEL GIORGIO, P. A., AND R. H. PETERS. 1993. Balance between phytoplankton production and plankton respiration in lakes. *Can. J. Fish. Aquat. Sci.* **50**: 282–289.

———, J. J. COLE, AND A. CIMBLERIS. 1997. Respiration rates in bacteria exceed phytoplankton production in unproductive aquatic systems. *Nature* **385**: 148–151.

FOGG, G. E. 1983. The ecological significance of extracellular products of phytoplankton photosynthesis. *Bot. Mar.* **26**: 3–14.

———, C. NALEWAJKO, AND W. D. WATT. 1965. Extracellular products of phytoplankton photosynthesis. *Proc. R. Soc. Lond., B* **162**: 517–534.

GASOL, J. M., AND X. A. G. MORÁN. 1999. Effects of filtration on bacterial activity and picoplankton community structure as assessed by flow cytometry. *Aquat. Microb. Ecol.* **16**: 251–264.

GOBLER, C. J., D. A. HUTCHINS, N. S. FISHER, E. M. COSPER, AND S. A. SAÑUDO-WILHELMY. 1997. Release and bioavailability of C, N, P, Se and Fe following viral lysis of a marine chrysophyte. *Limnol. Oceanogr.* **42**: 1492–1504.

GRASSHOFF, K., M. EHRHARDT, AND M. KREMLING. 1983. Methods of seawater analysis, 2nd ed. Verlag Chemie.

HANSELL, D. A., N. R. BATES, AND K. GUNDERSEN. 1995. Mineralization of dissolved organic carbon in the Sargasso Sea. *Mar. Chem.* **51**: 201–212.

JOINT, I., A. POMROY, G. SAVIDGE, AND P. BOYD. 1993. Size-fractionated primary productivity in the northeast Atlantic in May–July 1989. *Deep-Sea Res.* **40**: 423–440.

KARL, D. M., D. V. HEBEL, AND K. BJÖRKMANN. 1998. The role of dissolved organic matter release in the productivity of the oligotrophic North Pacific ocean. *Limnol. Oceanogr.* **43**: 1270–1286.

KIØRBOE, T. 1993. Turbulence, phytoplankton cell size, and the structure of pelagic food webs. *Adv. Mar. Biol.* **29**: 1–72.

LEGENDRE, L., AND J. LE FÈVRE. 1991. From individual planktonic cells to pelagic marine ecosystems and to global biogeochemical cycles, p. 261–300. *In* S. Demers [ed.], Particle analysis in oceanography. Springer.

———, AND ———. 1995. Microbial food webs and the export of biogenic carbon in oceans. *Aquat. Microb. Ecol.* **9**: 69–77.

———, AND F. RASSOULZADEGAN. 1995. Plankton and nutrient dynamics in marine waters. *Ophelia* **41**: 153–172.

MAGUE, T. H., E. FRIBERG, D. J. HUGHES, AND I. MORRIS. 1980. Extracellular release of carbon by marine phytoplankton; a physiological approach. *Limnol. Oceanogr.* **25**: 262–279.

MALINSKY-RUSHANSKY, N. Z., AND C. LEGRAND. 1996. Excretion of dissolved organic carbon by phytoplankton of different sizes and subsequent bacterial uptake. *Mar. Ecol. Prog. Ser.* **132**: 249–255.

MORÁN, X. A. G. 1999. Producción primaria particulada y disuelta

- en sistemas planctónicos marinos: variabilidad de mesoescala y acoplamiento con la producción bacteriana heterotrófica. Ph.D. thesis, Univ. of Oviedo.
- , J. M. GASOL, L. ARIN, AND M. ESTRADA. 1999. A comparison between glass fiber filters and membrane filters for the estimation of phytoplankton POC and DOC production. *Mar. Ecol. Prog. Ser.* **187**: 31–41.
- NAGATA, T. 2000. Production mechanisms of dissolved organic matter, p. 121–152. *In* D. L. Kirchman [ed.], *Microbial ecology of the oceans*. Wiley-Liss.
- SERRET, P., E. FERNÁNDEZ, J. A. SOSTRES, AND R. ANADÓN. 1999. Seasonal compensation of microbial production and respiration in a temperate sea. *Mar. Ecol. Prog. Ser.* **187**: 43–57.
- SHARP, J. H. 1977. Excretion of organic matter by marine phytoplankton: Do healthy cells do it?. *Limnol. Oceanogr.* **22**: 381–399.
- SMITH, S. V., AND J. T. HOLLIBAUGH. 1997. Annual cycle and interannual variability of ecosystem in a temperate climate embayment. *Ecol. Monogr.* **67**: 509–533.
- STRASKRABA, M., AND A. H. GNAUCK. 1985. *Freshwater ecosystems: Modelling and simulation*. Elsevier.
- TEIRA, E., P. SERRET, AND E. FERNÁNDEZ. 2001. Phytoplankton size-structure, particulate and dissolved organic carbon production and oxygen fluxes through microbial communities in the NW Iberian coastal transition zone. *Mar. Ecol. Prog. Ser.* (in press).
- TREMBLAY, J.-É., AND L. LEGENDRE. 1994. A model for the size-fractionated biomass and production of marine phytoplankton. *Limnol. Oceanogr.* **39**: 2004–2014.
- WILLIAMS, P. J. LE B., AND N. W. JENKINSON. 1982. A transportable microprocessor-controlled precise Winkler titration suitable for field station and shipboard use. *Limnol. Oceanogr.* **27**: 576–584.

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