Comparison of biomass and size spectra derived from optical plankton counter data and net samples: application to the assessment of mesoplankton distribution along the Northwest and North Iberian Shelf

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Data from vertical net hauls and from a submersible optical plankton counter (OPC) were compared in terms of biomass and slope of the normalized biomass–size spectra (NB–SS), a proxy for the size structure of the community. The relationship between the estimates of biomass in the overlapping range sampled by both methods (0.2–2 mm equivalent spherical diameter (ESD)) was linear and not significantly different to 1 ($B_{OPC}/B_{NET} = 1.1 \pm 0.1$, $r^2 = 0.84$). However, the relationship varied depending on the size fraction considered; the ratio $B_{OPC}/B_{NET}$ was 0.10 ± 0.04 ($r^2 = 0.30$), 0.8 ± 0.2 ($r^2 = 0.66$), and 2.4 ± 0.5 ($r^2 = 0.64$) for the size fractions 0.2–0.5, 0.5–1, and 1–2 mm ESD, respectively. The discrepancies between methods were presumably due to the combined effect of the limitations of the instrument in the lower detection limit and the errors in the volume of water sampled by the net for the smallest size fraction, and to net avoidance enhanced by clogging for the largest size fraction. The agreement between methods improved when the NB–SS of the different data sets were compared. The slope (b = −1.1) and the intercept (a = 14.6) of the NB–SS integrated across stations were not significantly different (Student’s t-test) for the linear model fitted to net samples, OPC data, or pooled data from both methods. Station by station, the slopes of the NB–SS from the net ($b_{NET}$) and the OPC ($b_{OPC}$) were not significantly different in 61% of the stations. This percentage increased to 78% when the comparison was limited to the mesoplankton size range. As an example of the applicability of the OPC, we showed the distribution of mesoplankton biomass and size structure along the NW and N Iberian Shelf during the winter–spring transition of 2002 and its relationship with the hydrographic scenario.

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Introduction

Concurrent measurements of physico-chemical variables and standing stocks of planktonic organisms undertaken over the appropriate spatio-temporal scales are critical to our understanding of marine ecosystem dynamics. Mesoplanktonic organisms (i.e. 0.2–2 mm) constitute an important component of the planktonic community, channelling matter and energy towards higher trophic levels and representing the main food source for many important fish stocks. Conventional methods for sampling plankton are based on net tows, which are often poorly suited to capture the spatio-temporal variability and patchy distributions of these organisms and are difficult to integrate with physical data acquired at nearly real-time. A variety of automatic plankton samplers based on optical principles have been...
developed in recent decades and are nowadays widely applied in resolving the spatio-temporal patterns of the different components of the planktonic community (Foote, 2000). The optical plankton counter (OPC) developed by Herman (1988, 1992) is a practical method for characterizing the abundance and size distribution of plankton in the size range of meso- and macroplankton (0.25–17 mm ESD). The OPC has been extensively used in the past decade to study zooplankton distributions in a variety of marine (references in Table 1) and freshwater (e.g. Sprules et al., 1998) ecosystems. More information about the OPC and its applications can be downloaded at: http://www.es.umb.edu/faculty/mzh/files/web-opc/web-opc.htm. However, the results from the OPC are sometimes difficult to reconcile with those from net sampling systems, with both OPC underestimates and overestimates relative to the estimations from a variety of net samplers (Table 1). These discrepancies stress the need for more comparison exercises among different sampling methodologies.

In this article we present the results of the comparison of the biomass estimates and size spectra obtained using a simple 20-μm plankton net and a subsmersible OPC during a cruise in the NW and N Iberian Shelf by means of a Sea Bird 25 CTD equipped with a SeaPoint fluorometer, and particle-size distributions in the 0.25–17 mm ESD size range obtained with a submersible OPC (Herman, 1988; Focal Technologies Ltd.), were acquired at night (between 21:00 and 05:00 GMT) during the winter–spring transition (from 14 to 30 March 2002) (Figure 1). A total of 110 stations distributed in sections perpendicular to the coastline were profiled. In 70 of these stations (Figure 1), plankton samples were collected after the CTD-OPC cast from the surface to 10-m depth above the bottom or to a maximum 100-m depth at deeper stations by means of vertical hauls of a conical 20-μm plankton net, 30-cm mouth diameter, and 165-cm length, carrying a flowmeter (General Oceanics model R2030). The choice of a net of these characteristics was guided by the intention to capture the widest range of size classes of planktonic organisms as possible in a single tow.

### Material and methods

### Sampling strategy

Sampling was carried out on board RV “Thalassa” during the PELACUS 0302 cruise. Simultaneous profiles of temperature, salinity, and fluorescence obtained along the NW and N Iberian Shelf by means of a Sea Bird 25 CTD...
Processing of net samples

A possible consequence of the use of a net with such a fine mesh size and a relatively small ratio of filtering area to mouth area (R = 5.7) to sample coastal-shelf waters is clogging of the net (Sameoto et al., 2000) (Figure 2, inset graph). The comparison of the volume of water sampled according to the flowmeter ($V_f$) and the theoretically estimated volume ($V_i = A \times z$, where $A$ is the mouth area and $z$ is the sampled depth), showed evidence of clogging at the majority of stations (Figure 2). To deal with this problem, we have assumed that net samples are representative of the upper portion of the water column that extends from the surface to the depth below which the net experienced severe clogging. We estimated this depth ($z^*$) considering the volume of water sampled according to the flowmeter record and the mouth area of the net:

$$z^* = \frac{V_f}{A}$$

Once on board, net samples were fractionated through a series of sieves of 20, 40, 80, 200, 500, 1000, and 2000-μm mesh size. Each fraction was washed in filtered seawater, transferred to pre-weighed glass-fibre filters, and stored frozen until further processing in the laboratory, where the dry weight per volume ($d_W, \text{g}\text{m}^{-3}$) was obtained. The volume of water sampled by the net was calculated from the flowmeter records. Biomass in each size fraction was expressed in carbon units ($B_{\text{NET}}$, mg C m$^{-3}$) using the empirical relationship of Bode et al. (1998):

$$\log B_{\text{NET}} = \frac{\log d_W - 0.626}{0.832}$$

OPC data processing

Series of mathematical transformations were applied to the raw OPC data to make them comparable with the estimations of biomass derived from the net samples. ESD size was calculated according to the algorithm proposed by Herman (1992), including a 15% increase to compensate for underestimation in size due to the random orientation of particles as they cross the OPC light beam (Herman, 1992; Beaulieu et al., 1999). ESD was converted to carbon biomass ($B_{\text{OPC}}, \text{mg C}$) according to the general allometric relationship proposed by Rodriguez and Mullin (1986):

$$\log B_{\text{OPC}} = 2.23 \log \text{ESD} - 5.58$$

and referred to the volume of water sampled by the OPC down to the estimated value of $z^*$ of each station.

Normalized biomass–size spectra (NB—SS)

The normalized biomass–size spectrum [nb(wi)] for each size class i with a nominal weight $w_i$ was estimated following Platt and Denman (1977):

$$\text{nb}(w_i) = \frac{B(w_i)}{\Delta w_i}$$

where $B(w_i)$ and $\Delta w_i$ are the biomass and the weight interval of each weight class i. The nominal weight of each weight class was fixed as its geometric mean. We considered the slope of the linear regression model fitted to the log$_2$-transformed data as the parameter that summarizes the size structure of the community. The slopes of the NB—SS were obtained from three different groups of data: OPC data in the size range 0.5—3.0 mm ESD (b$_{\text{OPC}}$), and data from net samples in the whole size range (20×10$^{-3}$—2 mm ESD; b$_{\text{NET}}$) and in the mesoplankton size range (0.2—2.0 mm ESD; b$_{\text{NET,M}}$). The slopes were compared using Student’s t-test (Zar, 1999).

Results

Comparison of biomass estimates

The relationship between the biomass estimated from OPC data ($B_{\text{OPC}}$) and net samples ($B_{\text{NET}}$) in the overlapping ESD size range sampled by both methods (0.2—2 mm) is shown in Figure 3a. $B_{\text{OPC}}$ and $B_{\text{NET}}$ ranged approximately from 20 to 180 and from 20 to 800 mg C m$^{-3}$, respectively. In 19 out of the 70 stations sampled (Figure 1), $B_{\text{NET}}$ was up to four times higher than the upper limit of the estimated range of $B_{\text{OPC}}$ and of the maximum values of mesoplankton biomass estimated by other authors in the Bay of Biscay (Poulet et al., 1996). We found that at these stations the volume of water sampled according to the flowmeter record, and thus the estimated depth below which severe net clogging was assumed, was very low ($V_f < 0.5 \text{ m}^3$, $z^* < 4 \text{ m}$). This problem seriously flawed the comparison of methods, so we decided to disregard these stations for
further analysis. Once these stations have been removed from the comparative analysis, the relationship between $B_{OPC}$ and $B_{NET}$ in the mesoplankton size range was linear and not significantly different from 1 ($B_{OPC} : B_{NET} = 1.1 \pm 0.1, r^2 = 0.84$; Figure 3b). However, the relationship varied depending on the size fraction considered, showing a large discrepancy between methods in the 0.2–0.5 mm ESD size range (Figure 3c; $B_{OPC} : B_{NET} = 0.8 \pm 0.2$; $r^2 = 0.66$).
Comparison of normalized biomass–size spectra (NB–SS)

The linear models fitted to the NB–SS derived from net samples \(20 \times 10^{-3}–2\ \text{mm ESD size range}\) and OPC data (0.5–3 mm ESD) integrated across stations were not significantly different (Student’s t-test, \(p > 0.05\)) (Figure 4). The slope and intercept of the linear model fitted to the general NB–SS (pooled data) were \(b = -1.11 \pm 0.05\) and \(a = 14.6 \pm 0.2\) (\(r^2 = 0.85\)), respectively.

Station by station, the NB–SS were linear in 39 out of the 51 stations selected for the analysis (e.g. Figure 5a). The remaining stations showed a clear departure from linearity (e.g. Figure 5b), in which case the results of Student’s t-test for differences between the slopes of the linear models fitted to the different groups of data (i.e. OPC, and net samples in the mesoplankton and in the whole size range sampled) (Table 2) must be taken only as indicative of the trends of the NB–SS. The slopes of the NB–SS derived from net samples (b\(_{\text{SS}}\)) and OPC data (b\(_{\text{OPC}}\)) were not significantly different in 61% of the stations (Table 2; e.g. Figure 5a). This rose to 78% when the comparison was limited to the mesoplankton size range (H\(_b\); b\(_{\text{NET,SM}}\) = b\(_{\text{OPC}}\)). An example of this type of NB–SS is shown in Figure 5a. The slopes were significantly different in only 11 stations, 8 of which showed steeper slopes in the NB–SS derived from OPC data than from net samples (i.e. b\(_{\text{OPC}}\) < b\(_{\text{NET,SM}}\); e.g. Figure 5c) and the rest showed the opposite (i.e. b\(_{\text{OPC}}\) > b\(_{\text{NET,SM}}\); e.g. Figure 5d).

Mesoplankton biomass and size structure in the NW and N Iberian Shelf during the winter–spring transition. Relationships with hydrographic variability

The spatial distribution of salinity and temperature at 50-m depth (Figure 6a, b) revealed the hydrographic signature of two regional-scale features: the Portugal Coastal Counter Current (PCCC) in the NW Iberian Shelf (Bode et al., 2002; Álvarez-Salgado et al., 2003), identified by its characteristic high salinity (35.7–35.8) and temperature (\(> 13.4^\circ\text{C}\)), and the general anticyclonic circulation (AC) on the easternmost part of the southern Bay of Biscay, characterized by lower salinity and temperature values (ca. 35.55 and \(< 12.8^\circ\text{C}\)). These features converged in the central Cantabrian Sea (around 4°W), as indicated by the relatively higher salinity and temperature variability in this area (35.5–35.7 and 12.4–12.8°C, respectively). Vertical mixing was observed in the PCCC domain, while an incipient thermal stratification was observed in the area occupied by the AC. Local processes, such as freshwater inputs in the vicinity of river discharges and estuaries, induced important thermohaline variability in the surface layers (data not shown).

Surface Chl \(a\) concentrations were low (<1 mg Chl \(a\) m\(^{-3}\)) in the PCCC domain (Figure 6c) and increased eastwards from the central Cantabrian Sea (>2 mg Chl \(a\) m\(^{-3}\)) due to the consolidation of the seasonal thermocline in this part of the shelf.

Mesoplankton biomass derived from the OPC output, integrated to 40-m depth (i.e. maximum depth of the thermocline in stratified conditions), is shown in Figure 6d. The highest values (>3 g C m\(^{-2}\)) were found in localized coastal areas in the W and NW sector of the Shelf, such as the river discharge from the river Douro (41°N), the mouth of the Rías Baixas (between 42°N and 43°N)

\(r^2 = 0.30\), a good match in the 0.5–1 mm ESD (Figure 3d; \(b_{\text{OPC}}\) : \(b_{\text{NET}} = 0.8 \pm 0.2\); \(r^2 = 0.66\)) and a slight overestimation by the OPC relative to net samples in the 1–2 mm ESD size range (Figure 3c; \(b_{\text{OPC}}\) : \(b_{\text{NET}} = 2.4 \pm 0.5\); \(r^2 = 0.64\)).

Figure 4. NB–SS integrated across stations showing the parameters of the linear regression models fitted to OPC data, net samples in the mesoplankton size range, and pooled data.
and the vicinity of Cape Estaca de Bares (around 8°W), and in a band that spreads all over the shelf in the easternmost part of the Cantabrian Sea (from 5°W to 2°W).

The spatial distribution of the slope of the NB-SS derived from OPC data exhibited clear differences in the study area (Figure 6e). The higher slopes (b > -0.5), corresponding to flatter spectra and indicating a predominance of the larger size fractions within the mesoplankton size range, were found in coastal locations where continental inputs were relatively important, such as the Rías Baixas or the northernmost Galician rias (around 43.45°N, 7.5°W). Conversely, the steeper slopes (b < -1.1), which indicate a predominance of the smaller size fractions, were found in the PCCC domain. The easternmost part of the Shelf showed slope values slightly higher than the slope of the NB-SS integrated across habitat types (b > -1.1). In summary, a clear coastal-offshore gradient from flatter to steeper NB-SS, along with a weaker west-to-east trend from steeper to flatter NB-SS, was observed.

Discussion

Comparison of biomass estimates

Field comparisons between biomass estimations derived from OPC data and from various types of sampling nets have been conducted in a range of marine ecosystems (Table 1). In some of these studies, the OPC overestimated biomass relative to net samples (e.g. Grant et al., 2000), while in other studies the OPC underestimated biomass (e.g. Herman,
the net samples, a good agreement between methods for the resulting in very low estimations from the OPC relative to the problem of net clogging may be enhanced due to the presumably concentration estimated by the OPC (ca. 180 mg C m$^{-1}$). The error in the estimation of V$_f$ was too low in comparison with the theoretically assumed to be severe ($z^*$), so we decided to remove these stations for the comparative analysis. These stations (Figure 1) were located in the vicinity of river discharges and estuaries and in areas of relatively high (i.e. $>3$ mg Chl a m$^{-3}$) Chl a concentrations (Figure 6c), where the problem of net clogging may be enhanced due to the presumably high concentrations of suspended matter.

The comparison of biomass for different size fractions revealed a huge discrepancy for the smallest size fraction, resulting in very low estimations from the OPC relative to the net samples, a good agreement between methods for the intermediate size fraction and a slight overestimation by the OPC relative to net samples for the largest size fraction. The discrepancy observed for the smallest size fraction may be due to problems related to the lower detection limit of the OPC, nominally fixed at 0.25 mm ESD size (Herman, 2002). A more realistic lower detection limit for the OPC could be set around 0.35 mm in order to avoid these methodological constraints. On the other hand, the differences between methods for the largest size fraction could be attributed to the escape response by large copepods, likely enhanced by net clogging (Sameoto et al., 2000). Unfortunately, we lack estimations of the number of particles per volume sampled by the net which would have been useful in support of our explanations concerning the discrepancies between methods.

Apart from the problems associated with the definition of the lower detection limit of the OPC and the errors in the calculation of the water volume sampled due to net clogging, other important issues that preclude a direct comparison between methods were the uncertainties associated with conversion factors and the non-simultaneous sampling. These factors must be considered and controlled in future, specifically designed, intercomparison studies.

Comparison of NB–SS
The slope of the general NB–SS, i.e. integrated across stations (i.e. habitat types) and across sampled sizes (b = −1.11 ± 0.05) was close to the expected value of −1.2 that characterizes oceanic pelagic systems when biomass is expressed in carbon units (Quiñones, 1994). This value indicates a roughly uniform biomass distribution over logarithmic size classes (Sheldon et al., 1972). Such a distribution usually appears when the spectrum is obtained from a wide range of habitat types and sizes of organisms (Rodríguez, 1994). According to Boudreau and Dickie (1992) the slope of the integrated NB–SS across trophic levels, thus covering a wide range of sizes, reflects the physiological or primary scaling in the size distribution of ecological properties, where the size dependence of metabolism exhibits its ecological significance. The primary scaling of the NB–SS integrated across habitat types estimated from both OPC data and net samples provided similar results. Therefore, the primary or physiological structure of the plankton community could be estimated from the size spectrum of the mesoplankton community sampled by the OPC, provided that the spectrum is integrated across habitat types (i.e. filtering out the spatio-temporal variability).

The slopes of the size spectra estimated for each station (i.e. each particular habitat type) with different sampling methodologies that covered distinct size ranges (i.e. from $20\times10^{-3}$ to 2 mm ESD size for the net and from 0.5 to 3 mm ESD size for the OPC) were not significantly different in most of the sampled stations (61%). In these cases, the structure of the planktonic community, from microplankton to mesoplankton, could have been reliably inferred from the OPC data alone. There was also a good
match between the slopes of the spectra obtained from net samples and OPC data in the mesoplankton size range.

In 23% of the stations, the NBSS showed a strong non-linearity (e.g. station 113, Figure 5b), indicating a relative predominance of certain sizes. At 22% of the stations the spectrum derived from OPC data was linear, but its shape tended to be steeper (b < −1.1, Figure 5c) or flatter (b > −1.1, Figure 5d) than the general NBSS integrated across functional groups and habitats, reflecting a relative predominance of small or large mesoplankton, respectively.

In cases when a single habitat type was considered, the ecological or secondary scaling, which operates at the level of functional groups with a similar and size-independent growth efficiencies, emerged (Boudreau and Dickie, 1992), and may be more important than the primary, size-dependent scaling. The departure from the size spectrum integrated across functional groups and habitat types can provide us with information on the diverse externally induced (e.g. horizontal transport) and internally driven processes, both at the individual level (e.g. growth efficiency) and at the community level (e.g. predation), which are in the last instance responsible for the observed community structure patterns (Rodríguez, 1994).

Mesoplankton distribution in the NW and N Iberian Shelf

At the regional scale, a clear zonation defined by the contrasting influences of the PCCC and the southern Bay of Biscay anticyclonic circulation was reflected in the distribution of biomass and size structure of the mesoplankton community characterized by means of the slope of the NBSS derived from the OPC output. There was a significant positive relationship between these two variables.
(B_{OPC} = 55.7b_{OPC} + 104.3, r^2 = 0.54), indicating that high mesoplankton biomass corresponded with low values of the slope of the NB—SS and vice versa. A conspicuous coastal-offshore gradient was observed: large mesoplankton, and thus relatively high biomass values, were found in the coastal zone and in the vicinity of river flows, while small mesoplankton, and thus relatively low biomass values, were found offshore. A less marked trend was also observed from west to east. In the domain of the PCCC, small mesoplankton prevailed, while in the easternmost part, where thermal stratification was well underway, large mesoplankton were more important. Such patterns are consistent with earlier observations of microplankton in the NW Iberian Shelf (Bode et al., 2002) showing the relevant influence of hydrographic features on the distribution of plankton. Our results expand the influence of the hydrographic scenario on the distribution and size structure of the mesoplankton community, mainly integrated by zooplankton.

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