HIGH-RESOLUTION SPATIO-TEMPORAL DISTRIBUTION OF A COASTAL PHYTOPLANKTON BLOOM USING LASER IN SITU SCATTERING AND TRANSMISSOMETRY (LISST)

Sílvia Anglès[1], Antoni Jordi[2], Esther Garcés[1], Mercedes Masó[1], Gotzon Basterretxea[3]

[1] Institut de Ciències del Mar (CMIMA-CSIC), Pg. Marítim de la Barceloneta, 37-49, 08003 Barcelona, Spain.

[2] Marine Sciences Research Center, Stony Brook University, 101 Endeavour Hall, Stony Brook, NY 11794-5000

[3] IMEDEA (UIB-CSIC), Miquel Marqués 21, 07190 Esporles, Baleares, Spain.

ABSTRACT

Development of optical observation technologies provides new insights into harmful algal bloom (HAB) detection and assessment of HAB species dynamics. Basing on preliminary laboratory tests, a laser in situ scattering and transmissometry instrument (LISST-100X) was used to monitor a high-biomass phytoplankton proliferation in the field. Short-term spatial and temporal changes in particle size distribution were measured during a recurrent *Alexandrium taylori* outbreak. Since the bloom was not monospecific, a size-fraction method to discriminate particular species from LISST-100X measurements was proposed. Results were validated to simultaneous microscopic counts of phytoplankton, and the significantly positive correlation obtained between the two methodologies confirmed the instrument's ability to discriminate phytoplankton at the group and species level. The LISST-100X obtains high-resolution in situ data, and is therefore a better alternative than the traditional microscope for assessing temporal and spatial evolution of HABs. Field observations showed high variability over a short time scale associated with diel vertical migration of *A. taylori* and the whole phytoplankton population (nanoplankton and microplankton). A numerical circulation model was used to investigate the influence of beach hydrodynamics in the observed horizontal variability. Simulations of the model suggested an important role of daily coastal circulation in determining the distribution of *A. taylori* in coastal environments.

RESUM

El desenvolupament de tecnologies d'observació òptica proporciona nous coneixements sobre la detecció de proliferacions d'algues nocives (PAN) i l'avaluació de la dinàmica d'espècies d'algues nocives. Basant-se en proves preliminars de laboratori, es va utilitzar un instrument *Laser in situ scattering and transmissometry* (LISST-100X) al camp per quantificar una proliferació de fitoplàncton d'alta biomassa. Es van mesurar els canvis espacials i temporals a curt termini en la distribució de mida de partícules durant una proliferació recurrent d'*Alexandrium taylori*. Atès que la proliferació no va ser monoespecífica, es va proposar un mètode de fraccions per mides per discriminar espècies concretes a partir de les mesures del LISST-100X. Els resultats es varen validar amb recomptes simultanis de fitoplàncton mitjançant microscòpia, i la correlació significativament positiva obtinguda entre les dues metodologies va confirmar la capacitat de l'instrument per discriminar el fitoplàncton a nivell de grup i d'espècie. El LISST-100X obté dades in situ d'alta resolució temporal, i per tant és una alternativa millor que el microscopi tradicional a l'hora de mesurar l'evolució temporal i espacial de les proliferacions d'algues nocives. Les observacions de camp van mostrar una alta variabilitat a curta escala temporal associada amb la migració vertical d'*A. taylori* i de la població total de fitoplàncton (nanoplàncton i microplàncton). Es va utilitzar un model numèric de circulació marina per investigar la influència de la hidrodinàmica en la variabilitat horitzontal observada. Les simulacions del model suggereixen un paper important de la circulació diària en la determinació de la distribució d'*A. taylori* en la zona costanera.

INTRODUCTION

Research into the population dynamics and ecology of microalgae that produce harmful algal blooms (HABs) is an essential issue for a better understanding of such events. The first step is to characterize the HAB species involved and the accompanying phytoplankton community, along with the biological, physical and chemical processes that interact with them in their natural environment. The variable nature of HABs, which tend to be episodic and patchy, requires a high resolution measurement of all these parameters. In certain high-biomass HAB events, when discoloration is not readily observed in the early stages, adequate evaluation of the bloom requires high-resolution data. Furthermore, precise estimation of cell abundance at this stage of bloom development is essential for forecasting purposes. Monitoring programs for HABs, aimed at detecting and characterizing distributions of HAB species, involve determining the target organisms at the species level and other components of the community as taxonomic or functional groups (GEOHAB, 2001). This is traditionally accomplished by laborious microscopic examination of discrete water samples collected along the coast at intermittent intervals of time, but these direct microscope counts are inadequate for rapid evaluation of the large quantity of samples required to describe the space and time variations of bloom-forming species.

To overcome this problem, a new generation of bio-optical instruments capable of rapidly assessing different aspects of phytoplankton dynamics is emerging. Technologies such as the FlowCAM, based on the combination of flow cytometry and image analysis (Buskey and Hyatt, 2006) and the CytoBuoy, a moorable cytometer (Dubelaar and Gerritzen, 2000), have successfully confirmed their ability to detect phytoplankton at the group and species level and have provided information on algal biomass. Recently, laser in situ scattering and transmissometry (LISST-100X) became available for automated detection of suspended particle size distribution (Agrawal and Pottsmith, 2000). Gentien et al. (1995) used the same optical principles to design a particle size analyzer that demonstrated its reliability in describing the distribution of particles along vertical profiles. Although the LISST-100X was originally designed for sediment analysis, previous studies have demonstrated its utility for determining phytoplankton and bacteria distributions (Serra et al. 2001, 2002). However, its application to detection at the species level in mixed phytoplankton communities has not been tested until now.

Frequent high-biomass blooms that occur along the Mediterranean coasts provide a good opportunity to test the ability of LISST-100X for detection at the species level in phytoplankton communities. Among the high-biomass bloom-producing species in the Mediterranean Sea, the noxious *Alexandrium taylori* Balech is responsible for most of the near-beach outbreaks. Appearance of these deleterious blooms leads to loss of aesthetic value caused by water discoloration, with considerable economic impacts on the local tourist industry (Masó and Garcés, 2006). These proliferations, often exceeding 10^6 cells l^{-1}, occur as green-brown patches that are usually noticeable for two months during the

summer. The exceptional duration of these blooms is due to relatively high growth rates and low loss of cells (Basterretxea et al., 2007; Garcés et al., 1999).

The objective of the present study is to assess the utility of the LISST-100X instrument short-term spatial and temporal changes of a noxious *A. taylori* bloom on a Mediterranean beach. Since the bloom was not monospecific, we propose a new method for calibrating the instrument measurements to determine the phytoplankton concentration for each species involved in the bloom. To explore the ability of the LISST-100X to detect and count phytoplankton at the group and species level, automated field cell abundance estimations for each species obtained by the instrument are compared with simultaneous samples counted by traditional microscopy.

In addition, the influence of coastal circulation on the observed cell abundance distribution is investigated using numerical simulations of a hydrodynamic model. Several studies have demonstrated that large-scale seasonal bloom dynamics are associated with major oceanographic processes such as shelf currents and tidal fronts (e.g., (Anderson et al., 2005; McGillicuddy et al., 2005). For smaller scales, Basterretxea et al. (2005) have demonstrated by numerical simulations that wind-induced currents can accumulate phytoplankton cells in near-coastal areas, thus favoring the occurrence and persistence of blooms. However, the interconnections between small-scale coastal blooms and local circulation in areas of restricted dynamism such as harbors, bays, lagoons and enclosed beaches remain largely unknown.

MATERIALS AND METHODS

LISST-100X description

A laser in situ scattering and transmissometry probe (LISST-100X Type-C, Sequoia Scientific, Inc.) was used to measure in situ the particle size distribution and concentration. Detailed information of the LISST-100X operation is given in Agrawal and Pottsmith (2000). Basically, the LISST-100X instrument obtains the particle volume concentration by size ranges using a technique based on the laser diffraction theory. A collimated laser beam (wavelength 670 nm) illuminates particles and the light scattered is sensed by a 32-ring detector. Each ring measures the scattering intensity over a range of small forward angles for 32 different size classes logarithmically spaced from 2.5 to 500 μm. At these small angles, laser diffraction is unaffected by composition of particles because light scattering is determined almost entirely by light diffracted by the particle. With the software provided by the manufacturers, the scattering intensities measured by the detector are mathematically inverted to obtain the particle volume concentration assuming that particles are spheres.

LISST-100X calibration

LISST-100X measures the particle volume concentration (PVC_i) in the range 2.5-500 μm (d_i). Considering that LISST-100X assumes that particles are spherical, the particle number concentration (PNC_i) can be calculated from

$$PNC_i = \frac{PVC_i}{\frac{4}{3}\pi\left(\frac{d_i}{2}\right)^3} \tag{1}$$

And the total particle number concentration (PNC_T) is obtained by integration over all the size classes:

$$PNC_T = \sum_{i=1}^{32} PNC_i \tag{2}$$

These calculations can be applied to estimate the concentration for one single species (cultures or monospecific natural assemblages) or for phytoplankton groups. However, proliferations are often multi-specific, as was the case in the bloom monitored in this study. For this reason, we applied a size-fraction method (SFM) in order to consider the particle number concentration for each species involved (PNC_s) in the bloom rather than the total particle number concentration. Calculations of the PNC_T were modified as follows:

$$PNC_{S,j} = \sum_{i=1}^{32} f_{ij} PNC_i \quad \text{for} \quad j = 1...N \tag{3}$$

where N is the number of species involved in the bloom and f_{ij} the size fraction of species j in the size class i, which is subject to

$$0 \leq \sum_{j=1}^{N} f_{ij} \leq 1 \quad \text{for} \quad i = 1...32 \tag{4}$$

This condition implies that f_{ij} is 1 for species j sizes ranging over a whole size class i and 0 for species j sizes ranging outside a size class i. The size fraction f_{ij} was determined by linear regression between total particle number concentration for each species (PNC_s) measured by microscopic phytoplankton analysis and particle number concentration for each size class (PNC_i) measured by the LISST-100X. The type of linear regression used in this work was a least squares fit constrained to the condition (4).

To evaluate the instrument performance, laboratory measurements of two different strains of *A. taylori* (EMBL AJ251654, ICMB219) were made with the LISST-100X. Strains were cultured in 50 ml polycarbonate flasks using F2 media (Guillard, 1973) and incubated in a growth chamber at 20ºC and 100 µmol m² s¹ irradiance on a 12:12 h light:dark cycle. In order to test the instrument sensitivity at different cell densities, a dilution series of the cultures (10^2 to 10^6 cells l^{-1}) was measured with the instrument's mixing chamber. After processing of the cultures, a subsample was collected and immediately fixed with Lugol's iodine solution for microscopic phytoplankton counting. The general procedure for phytoplankton enumeration involved sedimentation (24 h) of a subsample in a 10 to 50 ml settling chamber and subsequent counting of cells in an appropriate area (Andersen and Throndsen, 2003) using a Leica-Leitz DM-IL inverted microscope.

Field work

From 31 July to 2 August 2006 an in situ experiment was performed during the maintenance phase of an *A. taylori* bloom in La Fosca, a small (~300 m long) sandy beach located in the NW Mediterranean Sea (Fig. 1), where recurrent blooms of this species are observed during the summer (Garcés et al., 1999, 2005). The shorefront has a fairly uniform bathymetry sloping gently between 2 and 7 m. Typical summer meteorological conditions at the sampling site are dominated by a breeze regime, which may be disrupted by traveling frontal systems for periods of a few days (Soriano et al., 2006). The tidal regime is microtidal with a spring range of less than 0.25 m (Tsimplis et al., 1995). Calm conditions were experienced during the sampling and no significant sediment resuspension that could interfere in the particle analysis was observed.

To determine the temporal variations of the phytoplankton population, the first part of the experiment consisted in a continuous in situ record of the particle volume concentration during a diel cycle (from 14:00 GMT on 31 July until 19:00 GMT on 1 August) obtained with the LISST-100X deployed at the northern part of the beach at a depth of 0.5 m (Fig. 1). Simultaneous measurements of temperature and salinity were determined with a SBE 19plus CTD probe deployed at the same place. Water samples for microscopic phytoplankton analysis and chlorophyll *a* (Chl *a*) quantification were collected every hour during daytime. In order to estimate the spatial distribution of the phytoplankton, the second part of the experiment consisted in along- and cross-shore transects with a total number of 23 sampling points carried out from 12:00 to 13:20 GMT on 2 August (Fig. 1). Water surface measurements of the particle volume concentration were obtained in situ with the LISST-100X during 30 s at a depth of 0.5 m (the same like for the moored case). In order to remove short time scale variability, the particle volume concentration was time-averaged at each sampling point.

Subsamples (150 ml) of the water collected from La Fosca beach were immediately preserved with Lugol's iodine solution in the field. Posterior microscopic phytoplankton analysis in the laboratory involved the general procedure described above. For the identification of *Alexandrium* species, fixed specimens were stained with Calcofluor White M2R (Fritz and Triemer, 1985) and examined in an epifluorescence microscope under UV excitation (Axioplan, filter set Zeiss 487902, 1000x magnification). Tabular formula and morphological features of the thecal plates were studied following the criteria of Balech (1995). Subsamples (60 ml) for the quantification of total Chl *a* were filtered on 25 mm Whatman GF/F glass fiber filters and frozen. In the laboratory, filters were extracted in 8 ml 90% acetone and concentrations of Chl *a* were measured with a Turner Designs fluorometer.
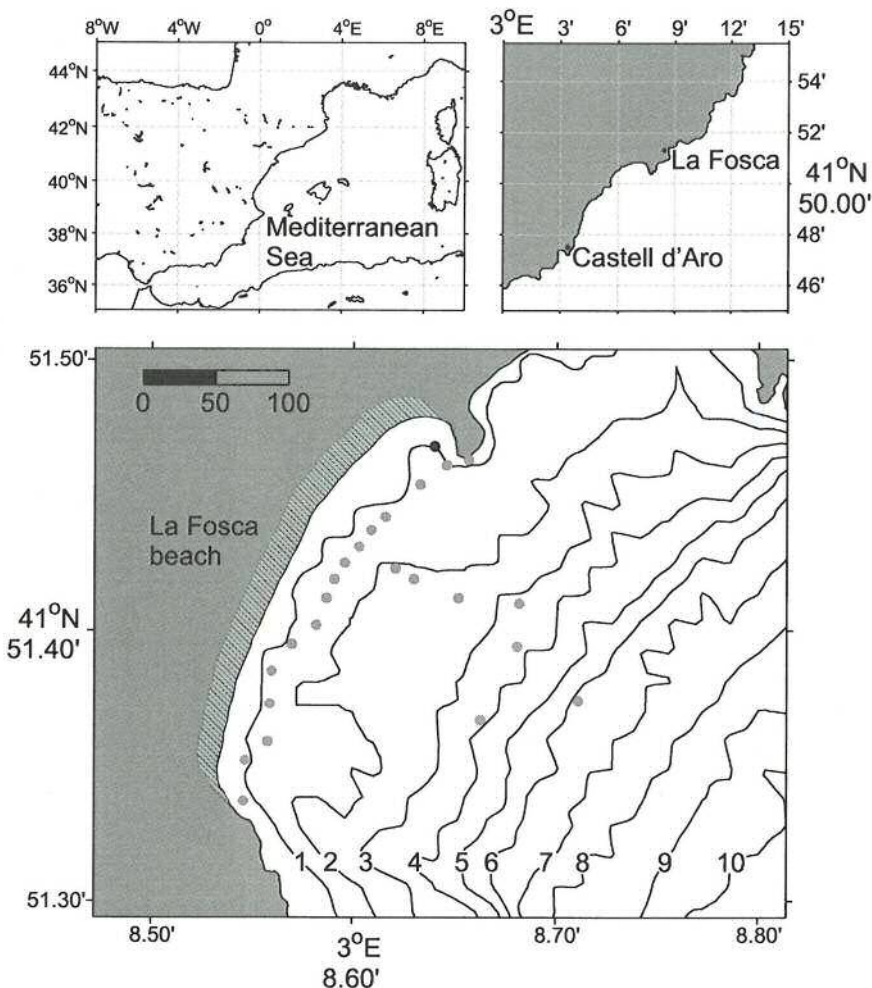

Fig. 1. Map of the study area displaying the coastal bathymetry and the location of the station for the diel cycle (black dot) and for the spatial distribution (gray dots).

Meteorological data of air temperature (accuracy ± 0.1°C), wind velocity (accuracy ± 0.1 m s¹), wind direction (accuracy ± 1°) and radiation (accuracy ± 1 W m⁻²) were provided from a nearby permanent station located in Castell d'Aro (Fig. 1) by the Catalan Meteorological Service (MeteoCat).

Hydrodynamic model

A numerical circulation model (FUNDY) was implemented for La Fosca beach. The FUNDY is a linear, shallow-water, sigma-coordinate, three-dimensional finite element model with spherical-polar extensions formulated in the frequency domain (Greenberg et al., 1998). This model has previously been used, among other applications, to study the influence of coastal circulation in *A. taylori* bloom dynamics in a pocket beach of the Mediterranean Sea (Basterretxea et al., 2005). The model solves the shallow water equations in linear
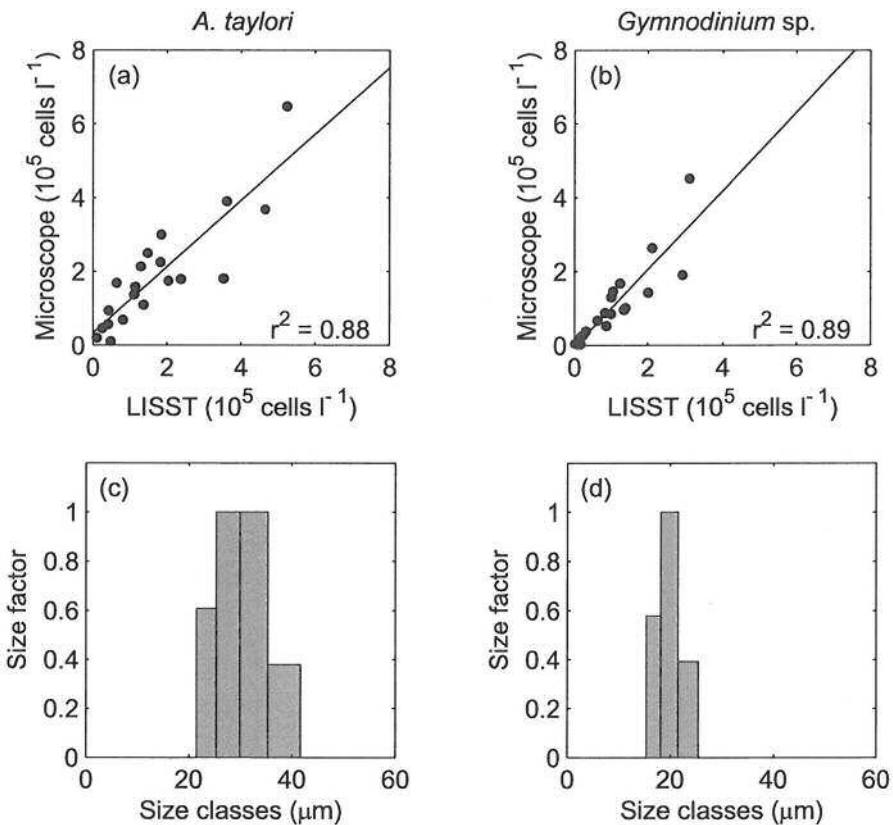

Fig. 2. Linear regression between counts of (a) *Alexandrium taylori* and (b) *Gymnodinium* sp. cells made using an inverted microscope and automated counts made using the LISST-100X. Correlation coefficient is indicated in the lower right corner. Size factor distribution of (c) *A. taylori* and (d) *Gymnodinium* sp. cells made using the LISST-100X and applying the SFM.

form with the Boussinesq and hydrostatic approximations. The vertical eddy viscosity is parameterized according to the Mellor and Yamada level 2.5 turbulence closure scheme, which has successfully been used to simulate the temporal variability of mixing on the continental shelf and the coastal sea (e.g., Wijesekera et al., 2003, Jordi et al., 2008). The computational domain extends from the beach to the inner shelf. Ocean boundaries lie far from the beach and hence are unlikely to influence the circulation there. Bathymetric data in the area were obtained with a shipmounted Biosonics DE-4000 echosounder equipped with a 200 kHz transducer. The mesh contained 6627 elements and 3504 nodes in the horizontal. Under each horizontal node, 10 one-dimensional linear elements were connected following a σ coordinate system.
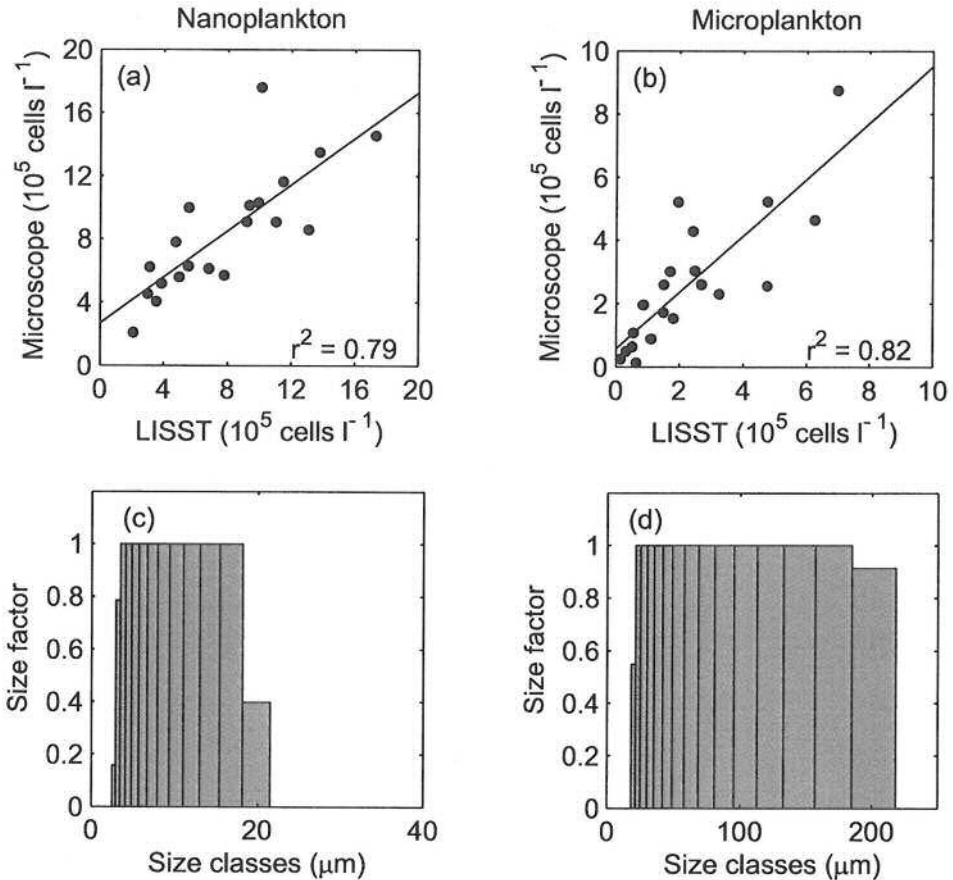

Fig. 3. Linear regression between counts of (a) nanoplankton and (b) microplankton cells made using an inverted microscope and automated counts made using the LISST-100X. Correlation coefficient is indicated in the lower right corner. Size factor distribution of (c) nanoplankton and (d) microplankton cells made using the LISST-100X and applying the SFM.

RESULTS

LISST calibration

In order to determine the potential of the LISST-100X for the in situ detection of A. *taylori* and co-blooming species, we first performed laboratory tests in which measurements of cultured strains of A. *taylori* using LISST-100X and microscopy cell counts were compared. The high correlation obtained between the two techniques ($r^2 = 0.95$) provided background for the in situ application of laser transmissometry in a natural environment.

Microscopic analysis of the water samples collected at La Fosca beach revealed that A. *taylori* was the most abundant species. The other co-blooming species was *Gymnodinium*
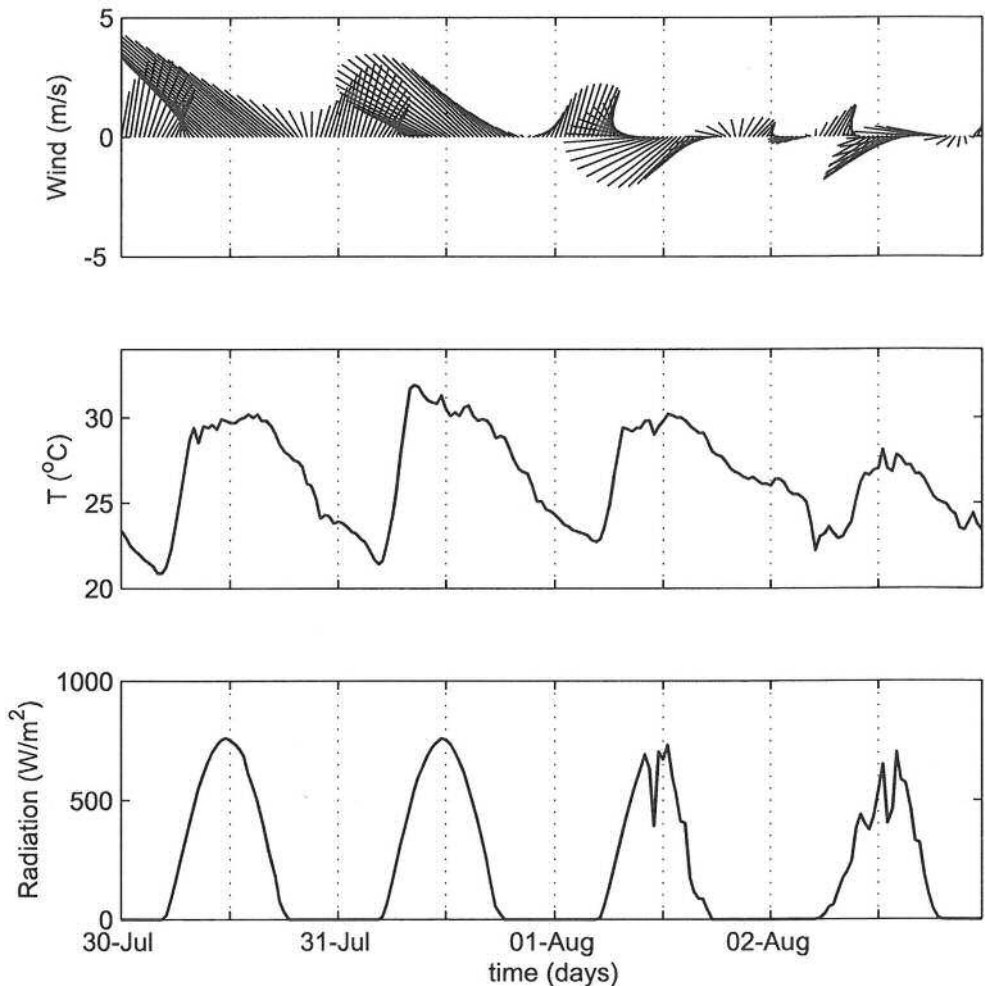

Fig. 4. Meteorological records from Castell d'Aro station: low-passed wind, air temperature and solar radiation. Wind vectors have been rotated 45° clockwise for convenience.

sp., a common accompanying species during *A. taylori* outbreaks. Therefore, we first focused our analysis on these two species and their particle number concentrations were estimated using the SFM following equation (3) and condition (4). Comparisons between microscopic cell counts and LISST-100X particle number concentrations of *A. taylori* (Fig. 2a) and *Gymnodinium* sp. (Fig. 2b) showed good agreement, with similar correlations of 0.88 and 0.89, respectively. Detected size fractions for both species varied from 23 to 45 µm for *A. taylori* and from 17 to 28 µm for *Gymnodinium* sp., which were also in accordance with the cell sizes observed under the microscope (Figs. 2c and 2d). Other phytoplankton species such as *Ostreopsis* sp, *Prorocentrum rathymum, P. micans,* and *Scrippsiella* spp. were detected, but in very low concentrations. Thus, in order to characterize the entire
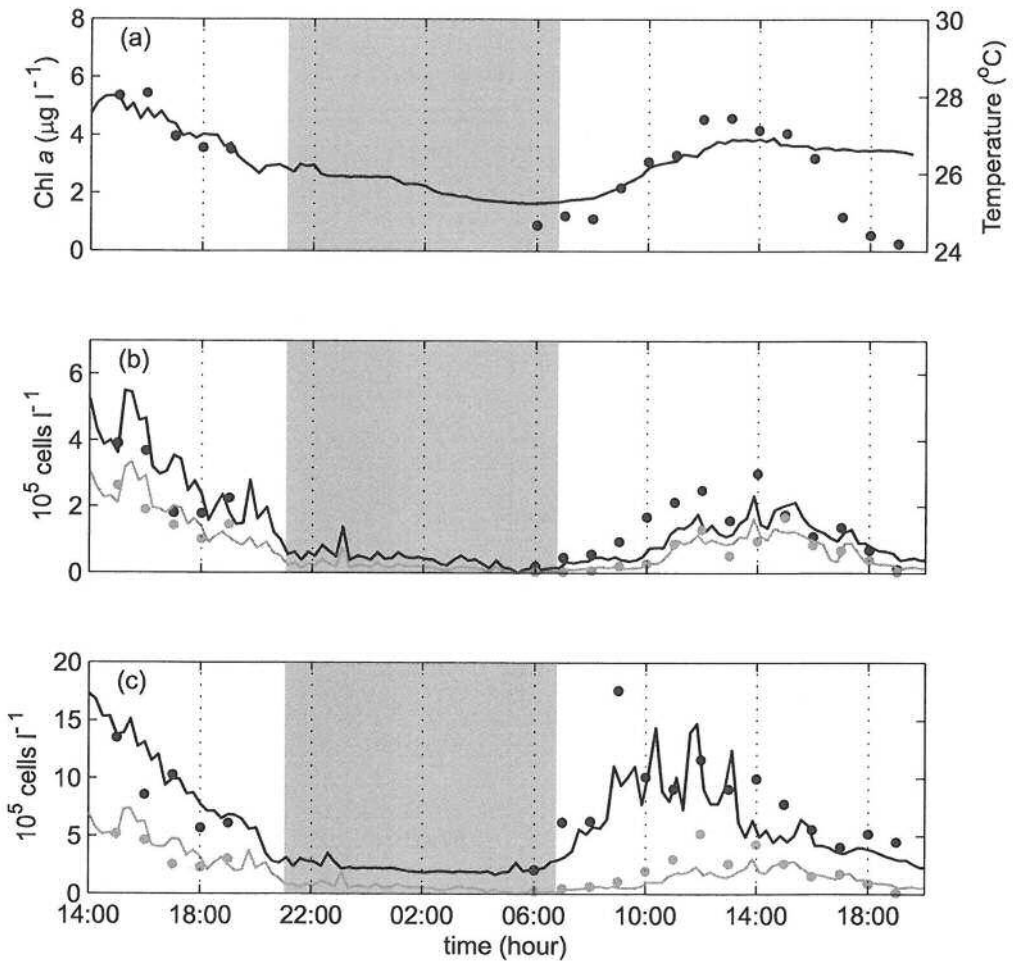

Fig. 5. Diel cycle from 31 July until 1 August for (a) chlorophyll a concentration (dots) and sea temperature (line), (b) *Alexandrium taylori* (black) and *Gymnodinium* sp. (gray) counts made using an inverted microscope (dots) and automated counts made using the LISST-100X (line), and (c) microplankton (black) and nanoplankton (gray) counts made using an inverted microscope (dots) and automated counts made using the LISST-100X (line).

phytoplankton population, all microscopic cell counts were grouped into nanoplankton (2-20 µm) and microplankton (20-200 µm), and LISST-100X concentrations were estimated for these groups. Correlations obtained by comparisons between the two methods were also significant (Fig. 3). Furthermore, LISST-100X size fractions for both groups fitted into the corresponding microscopy size estimations. It is important to note that occurrence of mesoplankton (>200 µm) was not detected by either the LISST-100X or the microscope, which confirms that planktonic macrograzers are scarce in this coastal environment (Calbet et al., 2003).

Temporal variations

Meteorological conditions experienced in the area during the previous day and the days of the experiment are shown in Figure 4. A breeze regime, resulting from the contrasting thermal response of land and sea, was the dominant pattern during the first two days
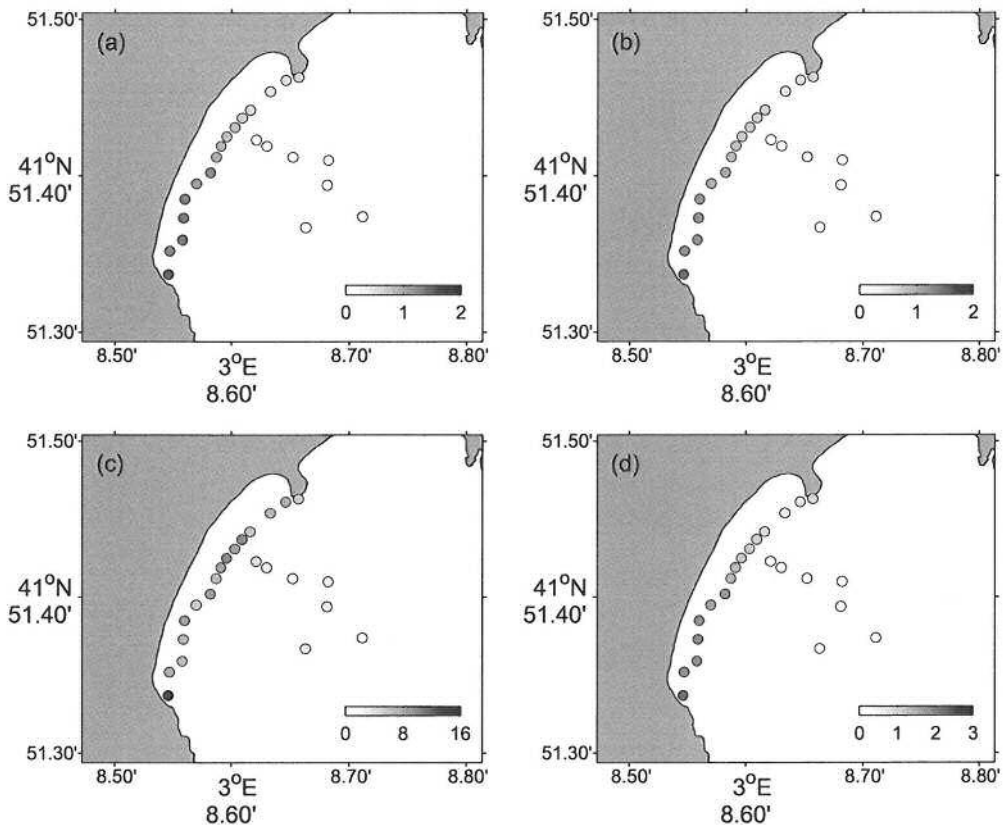

Fig 6. Spatial distribution on 2 August of (a) *Alexandrium taylori*, (b) *Gymnodinium* sp., (c) nanoplankton and (d) microplankton cell counts made using the LISST-100X. Units are 10^5 cells l^{-1}.

(30 and 31 July). A sea breeze blew from the southeast (onshore) during the day and there was a weaker land breeze circulation at night. Solar radiation and diurnal temperatures were lower during the experiment (1 and 2 August) than on previous days, which could have been responsible for the weak northeasterly flow that developed at midday on these two days.

Water temperature (Fig. 5a) reached maximum values of 28°C on 31 July. On 1 August temperature values were lower, and always below 27°C. No significant salinity variations were observed during the experiment (38.1 - 38.4). Chl *a* concentrations ranged from 5.5 µg l⁻¹ at midday on 31 July to 0.25 at night on 1 August (Fig. 5a) and water discoloration was clearly perceivable along the shoreline in the northern part of the beach. Generally, Chl *a* showed good agreement with phytoplankton cell abundances measured by the two methodologies, with a better correlation coefficient for the LISST-100X ($r^2 = 0.91$) than for the microscope ($r^2 = 0.81$).

Figure 5b shows the diel variation of the particles as depicted from in situ LISST-100X measurements of this part of the beach. Surface cell concentrations of *A. taylori* reached values of 10^5 cells l⁻¹ from 14:00 to 20:00 GMT, with a substantial cell decrease at night from 21:00 to 08:00 GMT. Maximum cell densities were attained on 31 July, whereas cell concentrations were lower on 1 August. Cell abundances showed a diel cycle typical of vertically migrating organisms, even though a stock of 10^4 cells l⁻¹ remained in surface waters at all times. *Gymnodinium* sp. followed a similar pattern to *A. taylori*, with lower concentrations in both methodologies (microscope counts and LISST-100X estimations). Nanoplankton and microplankton also followed this daily pattern (Fig. 5c).
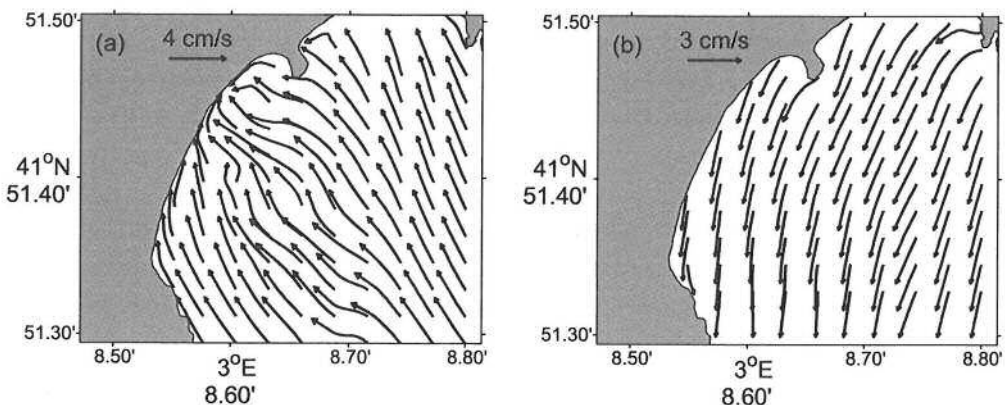

Fig 7. Wind-induced surface circulation patterns in La Fosca corresponding to the (a) averaged breeze regime at midday for 30-31 July, and (b) the averaged northeasterly wind at midday for 1-2 August.

Spatial distributions

Cell distributions along and across the beach measured in situ with the LISST-100X on 2 August under northeasterly winds are displayed in Figure 6. Inshore stations revealed a north-south gradient with *A. taylori* cell densities increasing to the south, whereas abundances rapidly decreased in the offshore direction. A similar spatial pattern was observed in *Gymnodinium* sp. and microplankton although abundances were higher in the latter. Nanoplankton cells showed a slightly different pattern, with enhancement in the nearshore but a less clear alongshore distribution and abundances reaching 10^4 cells l^{-1} offshore.

The numerical simulation of the FUNDY model was forced by the observed meteorological conditions (wind and heat flux) from 30 July to 2 August. Modeled surface averaged circulation patterns at midday for the first two days (breeze regime) and for the last two days (northeasterly winds) are displayed in Figure 7. Prior to compare coastal currents and phytoplankton distribution, it is convenient to check if the measurement of phytoplankton can be considered representative and therefore gives an overall picture of the horizontal distribution. If the time of measurement would be larger than the time taken by the current to move from the farthest station to the closest station, then the spatial distribution presented would be not representative. The distance between the farthest station and the closest was less than 300 m and the coastal currents are of the order of 3 cm s^{-1} (for northeasterly winds), therefore the fluid would take more than 165 min to move from the first to the last station. This time doubles the time in the measurement of the spatial distribution (about 80 min) and therefore we can consider our measurements as representative.

The response of surface currents to breeze regime generates a northwesterly flow (4-5 cm s^{-1}) and a progressive northward deflection of the currents near La Fosca beach (Fig. 7a). These conditions favor particle accumulation in the vicinity of the northern edge of the beach, where currents are weakened by the presence of a headland (note that this was the location of the LISST-100X during the first part of the experiment). Conversely, northeasterly winds produce currents flowing cyclonically along the coast (Fig. 7b). This flow tends to advect surface particles released near the beach southward, promoting cell accumulation at the southern boundary of La Fosca beach. Differences in *A. taylori*, *Gymnodinium* sp., nanoplankton and microplankton cell abundance between the first part (31 July with breeze regime) and the second part (2 August with northeasterly winds) of the experiment could be at least partially explained by the above mentioned patterns of surface flow.

Discussion

Advances in optical technologies provide new perspectives for the analysis of HAB-producing species and their dynamics. Herein we present a case study in which laser diffractometry has been successfully employed for rapid in situ assessment of a bloom-producing community in a coastal marine environment. The significantly positive correlation obtained between microscopy cell counts and LISST-100X measurements for the different species and phytoplankton groups demonstrates the viability of rapid cell abundance assessment using laser scattering transmissometry in both laboratory and field conditions. These results are in agreement with the values obtained by Serra et al. (2001) for purple sulphur bacteria, although better correlations were obtained in our case, which can be attributed to the use of the SFM. The fact that better correlations are attained for comparisons at the species level than for phytoplankton groups suggests that the SFM is suitable and improves LISST-100X estimations at the species level. The disagreements between the two techniques can be attributed to the intrinsic error they involve. Inverted microscope counting sensitivity is 20-500 cells l^{-1}, depending on the sample volume and on the number of fields counted (Andersen and Throndsen, 2003), and an important limitation of the LISST-100X is that particles outside the range of measurement are included in the nearest size, producing an overestimation of the smallest and largest particle sizes (Agrawal and Pottsmith, 2000). In our study, while particles larger than 200 µm were not detected, a low abundance of particles smaller than 2 µm was observed. Although the concentration was not significant and the SFM might have almost discriminated them, it is a possible source of error that could explain the lowest correlation obtained for nanoplankton.

Another interesting feature which should be considered when the LISST-100X is used in field studies is the assumption of a spherical shape when the measurements are converted into particle size distributions. In non-spherical particles, LISST-100X detects an altered diffraction pattern since the method provides the concentration of equivalent spheres. This may result in a deviated estimation of the particle number concentration, depending on the particle aspect ratio (lower over higher diameter) (Pedocchi and Garcia, 2006). Considering that phytoplankton displays a wide range of cell shapes, this should be taken into account in measuring natural phytoplankton assemblages. In our study, the two main species found in the field (*A. taylori* and *Gymnodinium* sp.) were nearly spherical cells, and their size ranges were thus clearly distinguishable. Another source of potential counting error is size overlapping of species. Whereas in monospecific blooms cell densities can be easily obtained from the particle number concentration for the size range of the species measured by the LISST-100X, in multispecific blooms discriminating phytoplankton species may be not as simple. As shown in this study, this can be solved by an appropriate calibration such as the SFM. Hence, as also recommended by Serra et al. (2001), the previous analysis of a sample under the microscope is required in order to identify all the species involved and to evaluate possible sources of errors. Afterwards, the LISST-100X can be deployed to acquire field measurements and subsequently apply the proper data

calibration to differentiate species.

The main advantage of LISST-100X over traditional microscope counting is the high-resolution data of phytoplankton that can be attained. Counting under the microscope is a time-consuming technique and the number of samples that can be handled is therefore lower. Other instruments (CytoBuoy, FlowCAM) have the ability to detect phytoplankton at the species level, but they need to be complemented by continuous in situ deployments of other sensors capable of highly resolved sampling on the HAB scales (Babin et al., 2005). While high-resolution time series of physical parameters such as temperature, salinity, turbulence and irradiance can be accomplished by a wide range of devices, high-resolution data of phytoplankton can only be acquired by means of bio-optical devices based mainly on fluorescence (Doubell et al., 2006) or apparent and inherent optical properties (Robbins et al., 2006). All these bio-optical instruments reach high-resolved sampling that can be easily related to phytoplankton and other water components. Here we suggest that the LISST-100X is a valuable tool for rapidly assessing HAB species in field environments and providing high-resolution data at an appropriate scale autonomously.

Phytoplankton variability observed over a short time scale indicates the profound importance of microscale and small-scale processes in the ecology of communities of marine microorganisms. In our first field experiment, surface densities of A. taylori showed a diurnal vertical migration pattern. The increase in cells in the water column in the morning through midday, with concentrations peaking in the afternoon, was followed by lower levels at night. Since no bottom densities were measured during the experiment, this diurnal pattern could be the result of an accumulation of cells on the surface during the day and dispersion at night caused by land-breeze. Nevertheless, vertical migration for the dinoflagellate has been previously described (Garcés et al., 1999), and therefore this biological process may influence its horizontal distribution over a time scale of hours. Moreover, the vertical migration was observed for Gymnodinium sp., nanoplankton and microplankton. This vertical migration of the whole phytoplankton community has also been hypothesized for bacteria in an A. taylori bloom, where strong daily changes in bacterial abundance occurred in the presence of large dinoflagellate populations performing daily vertical migrations (Gasol et al., 2005). In particular, A. taylori and Gymnodinium sp. populations seem to have a very similar behavior in vertical migration.

In the second part of the experiment, our results suggest that the observed horizontal distribution is linked to the coastal circulation. A typical summer sea breeze provokes the accumulation of the organisms at the northern part of the beach where discoloration is most often observed, and changes in the prevailing wind regime modify the horizontal distribution in agreement with the north-south distribution and accumulation in the southern edge of the beach measured in the second part of the experiment. However, this north-south gradient in the nearshore appears slightly modified by small-scale variability, which is more significant for the nanoplankton distribution. This could be explained by

the presence of thin layers for different phytoplankton populations. Unfortunately, we are unable to resolve the presence of thin layers with our sampling strategy. A detailed study of the role played by thin layers in phytoplankton distribution should be addressed. The possible inaccuracy of the LISST-100X for measuring particles near the smallest size range of the instrument detection could also contribute to the higher small-scale variability for nanoplankton. Furthermore, it is known that wind may prevent bloom maintenance by dispersing organisms offshore and/or through turbulent mixing (Yamamoto et al., 2002). However, we observed that wind-induced currents during the day accumulated the organism near the beach. The potential dispersion effect of currents at night, induced by land breeze and directed offshore, was limited by the vertical migration of *A. taylori,* with settling during the night mitigating cell dispersal (Basterretxea et al., 2005). Moreover, during the experiment, wind intensity was weak and consequently turbulence mixing did not overcome the thresholds for the appearance of negative effects on the bloom maintenance.

CONCLUSIONS

The LISST-100X provides rapid and consistent particle counts, and is therefore a valuable tool for assessing HAB species dynamics in field environments. The high-resolution in situ data that can be attained by this instrument allows the temporal and spatial evolution of HABs to be described. During the phytoplankton bloom investigated, temporal variability was due to diel vertical migration of the phytoplankton population, whereas the horizontal distribution seemed to be linked to the coastal circulation resulting from the prevailing wind regime.

ACKNOWLEDGEMENTS

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