

HARMFUL ALGAE NEWS

An IOC Newsletter on toxic algae and algal blooms

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No. 36

• Turkey

Mucilage event associated with diatoms and dinoflagellates in Sea of Marmara, Turkey

The massive presence of mucilaginous organic matter, resulting from planktonic and benthic algal blooms, has become more frequent in many coastal waters around Europe, especially in the Adriatic. The appearance of mucilage in the Adriatic Sea has been reported periodically since 1800, with major mucus blooms during the 1990s [1]. The mucilage phenomenon of the Adriatic Sea had usually been related to extracellular organic matter of phytoplanktonic origin.

An extensive mucilage event

consisting of white gelatinous material initially suspended at the surface and in the water column was noticed along the Turkish coast of the Marmara Sea (especially Izmit Bay). Marmara Sea has a rather complex hydrological system, in a zone of transition between dense (salinity 37- 38.5 ‰) and warmer waters originating in the Mediterranean Sea, and cold, lower-salinity water (20-22 ‰) coming from the Black Sea. The pycnocline lies at 10 to 30 m depth and varies seasonally [2].

The first mucilage was observed in

mid-autumn 2007 along the north-eastern part of Marmara Sea with temperatures $18.4 \pm 1.0^\circ\text{C}$. It extended from Izmit Bay to the Dardanelles during the calm weather period; it was denser and of longer duration in Izmit Bay, which is affected by intense industrial activity, and which has a weaker circulation compared to Marmara Sea. To identify phytoplankton species responsible for the mucilage, water samples were collected from surface and deep water at target sites (Fig 1)

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• Mexico

A bloom of *Ceratium balechii* in Michoacan, Pacific Mexico

From 5 to 12 June, 2007, the National Autonomous University of Mexico (UNAM) conducted the Talud XI cruise on the Pacific coast of Mexico. While at stations 15 and 16 off Michoacan State, a marked red discoloration was observed. Microscopical analyses revealed the presence of *Ceratium balechii* Meave, Okolodkov et Zamudio [1]. This species, a very frequent bloom forming organism [2](Fig.1A), has been commonly confused mainly with *Ceratium dens* Ostenfeld et Schmidt [3: Fig. 11, see the drawing of the anomalous chain of two individuals e)] and recently, with *Ceratium divaricatum* Kofoid [4]. Nevertheless, the cell morphology is quite distinct. The body is pentagonal, asymmetric, and very different from the above mentioned species. Also, the antapical horns have different size and direction. Meave *et al* [1] propose that according to the roughness and size of the antapical horns there may be two forms, hypothesizing that they result from an adaptation to different water densities. At the moment, we are not able to speculate on this last point, since our observations in a single place and population showed a wide size variability (Fig.1B) as has been shown in cultures [5]. We have observed minute specimens (Fig.1C) as well as some showing teratogenic changes (Fig.1D). This species, unlike *C. dens* and *C. divaricatum*, is

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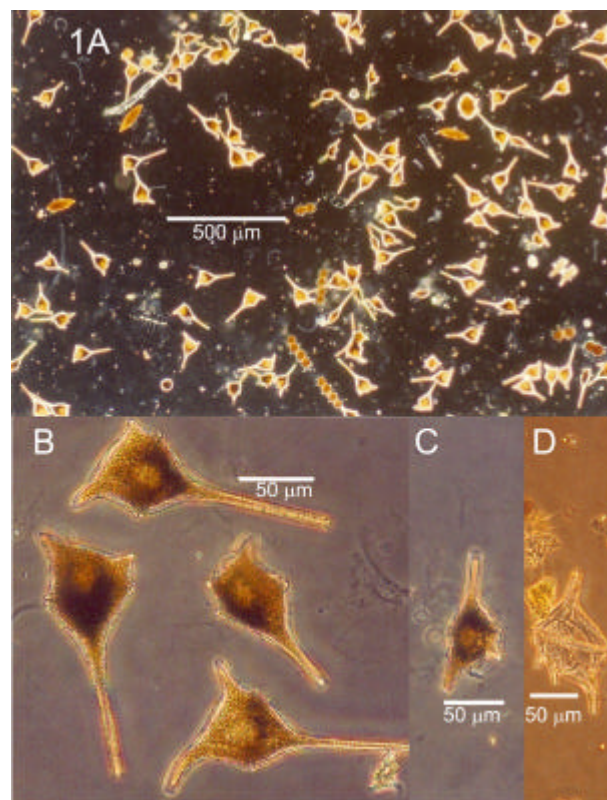


Fig. 1. A) *Ceratium balechii*, dark field at 40x, 1 ml sample. B) Four cells of different size. C) Small cell. D) Anomalous specimen with three antapical horns. B-D) Phase contrast, 200x.

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from Marmara Sea (nine sampling stations from Istanbul Strait on October and December 2007, February and March 2008; from Prince Islands on February 2008; and from Izmit Bay on October 2007) during the bloom period (September 2007-March 2008). Temperature, salinity, pH, conductivity and dissolved oxygen profiles were obtained with a SEABIRD CTD probe during the cruise in Istanbul Strait and Prince Island. Some nutrients and chlorophyll *a* analyses were performed according to standard methods. Phytoplankton samples were collected for qualitative and quantitative analyses by Nansen bottle and plankton nets (with mesh size 10 μ and 30 μ) respectively. Phytoplankton analysis was carried out by the Utermöhl method. During the mucilage event, aggregates were sampled in the water column by SCUBA divers using syringes, and also collected from sediments and mussels. Some environmental parameters during the different sampling periods are summarized in Table 1.

Historically, diatoms have always been considered as the algal group mainly involved in this kind of event due to their abundance in mucilaginous aggregates [3] and for their known extracellular release of polysaccharides [1, 4]. In our observations, high diatom densities were recorded during this period (Fig. 2). During the first days of this bloom, the phytoplankton community was characterized by

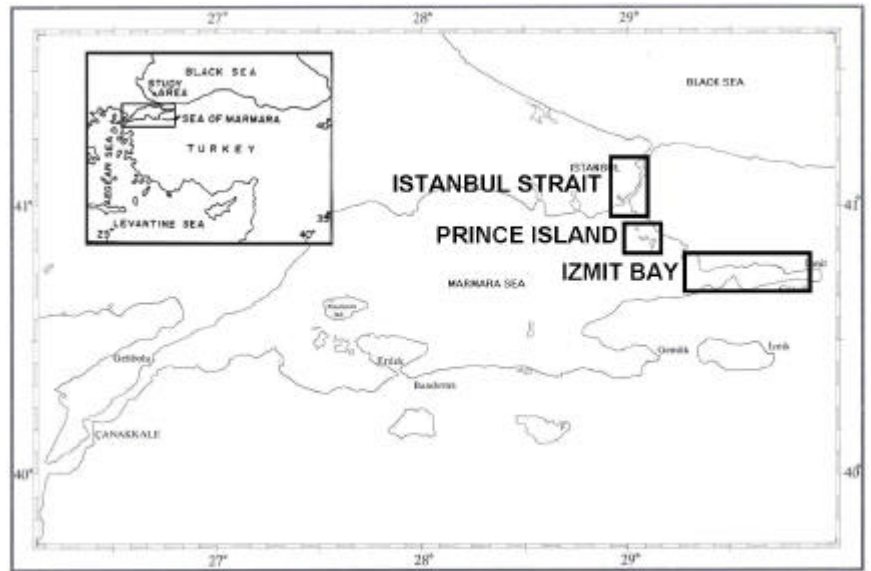


Fig 1. Sampling sites (Istanbul Strait, Prince Island, and Izmit Bay) from Marmara Sea during the mucilage events.

abundant diatom species (*Proboscia alata*, *Rhizosolenia* sp., *Pseudosolenia calcar-avis*). Total diatom density was more than 10⁷ cells L⁻¹. This phenomenon successively increased and spread on a large scale in the Marmara Sea. However, shifts were observed in dominant species during the rest of the bloom period (*Thalassiosira* sp., *Ditylum brightwellii*, *Coscinodiscus* spp., *Leptocylindrus minimus*, *Skeletonema costatum*, *Chaetoceros* spp., *Cerataulina pelagica*, *Cylindrotheca closterium*, *Pseudo-nitzschia* cf. *seriata*). In February 2008 simultaneously with the diatom bloom (max value 3.9 x 10⁶ cells L⁻¹), the dinoflagellate *Gonyaulax fragilis* became abundant in the mucilage. The

presence, in fairly high numbers, of *Gonyaulax fragilis* is reported in several papers [4, 5, 6] concerning the mucilage events in the Adriatic Sea. In our observations, *Gonyaulax fragilis* contributed to the bloom as biomass and possibly to mucilage production, but its density did not reach high numbers; its maximum concentration (36 x 10³ cells L⁻¹) was recorded in the pycnocline (15 m) off Prince Island. The dominant species of phytoplankton during the mucilage event were similar at all investigated sites from Marmara Sea. Furthermore, a large scale increase of coccolithophores (especially *Emiliana huxleyi*) was also observed during the mucilage event off Istanbul.

Mucilage can affect marine ecosystems by massive development of

Table 1. Comparison of minimum and maximum values of some parameters at the surface during the mucilage events in 2007-2008, and different periods of study in Marmara Sea [2,7].

Autumn-Winter period of different years	Izmit Bay		Istanbul Strait		Prince Island
	1999-2000 (from Aktan et al. 2005)	2007-2008 Mucilage Period	1997-1998 (from Aktan et al. 1999)	2007-2008 Mucilage Period	2007-2008 Mucilage Period
Water temperature (°C)	21.2-23.0	17.5	4.8-15.3	6.7-16	7.4-9.6
D. oxygen (mg L ⁻¹)	8.3-15.6	9.4	7.4-11.0	10.1-10.7	7.9-10.1
NO ₂ +NO ₃ -N (μg N L ⁻¹)	7.7-15.7		1.4-2.8	6.2-8.4	3.8-10.6
PO ₄ -P (μg P L ⁻¹)	2.3-14.5		0.0-0.2	0.1	0.0-0.1
T.Phosphorus (μg P L ⁻¹)	13.0-28.1		0.2-0.5	0.1	
SiO ₂ -Si (μg Si L ⁻¹)	57-257		3.0-7.1	0.01- 0.04	0.035-0.043
Conductivity (μS cm ⁻¹)			29	19.1-19.2	22.5-28.9
pH		8.5	8.32-8.48	9.4-9.6	7.5-9.6
Surface salinity (psu)	20-22	20		17.1-18.1	21.1-25.3
Secchi disc (m)	2.5-5.5	<1	6.0-7.0	5-5.5	3.7-5.5
Chlorophyll a (mg m ⁻³)	3.6-7.0	-	0.8-0.9	0.13-1.86	1.3 1-9
Dominant algae in density	dinoflagellates	Single and colonial diatoms	Dinoflagellates coccolithophores (<i>E. huxleyi</i>)	Single and colonial diatoms	Single and colonial diatoms
Dominant algae in frequency	dinoflagellates	Single and colonial diatoms	Dinoflagellates coccolithophores (<i>E. huxleyi</i>)	Diatoms and dinoflagellates (<i>G. fragilis</i>)	Diatoms and dinoflagellates (<i>G. fragilis</i>)

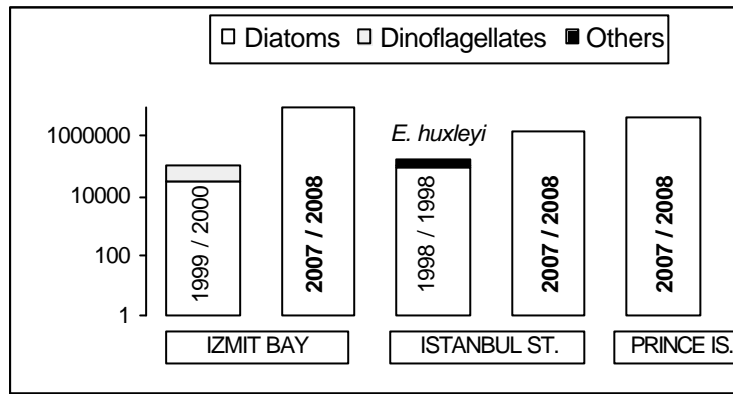


Fig. 2. Comparison of mean densities (cells L^{-1} , logarithmic) of phytoplankton groups during the mucilage events in 2007-2008 and the different periods of study at target sites (Izmit Bay, Istanbul Strait, Prince Islands) of Marmara Sea.

aggregates that cover large areas, and it can also seriously affect fisheries and tourism [1]. During these observations, neither hypoxia nor anoxia nor fish kills were recorded, but the large quantity of mucilage aggregates affected fishing and sport diving activities (personal communications from fishermen and divers). Extensive benthic mucilage aggregates were also observed on the sediments and mussels; the main bloom forming species are planktonic and benthic colonial and single cells of diatoms within the mucilage aggregates

on the sediments. During our dive we noticed that dense sedimentation of these aggregates (on sediment, mussels and crabs, etc.) has a negative effect on the benthic ecosystem. Following this preliminary study, we have started a long term monitoring project of these mucilaginous phenomena to explore their distribution and seasonal dynamics, potential causes (nutrients, climatic, hydrographical and oceanographic conditions), consequences (benthic and pelagic ecosystem and fisheries) and the possible relationship with grazers.

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associated with upwelling. Even though *C. divaricatum* is very variable [6: Fig. 31], the antapical horns are never directed downwards.

Cortés and Nuñez pointed out two peaks of abundance in spring (March-April) and autumn (October) in the Gulf of California [2]. Our observations in early summer, far outside and south of the Gulf of California, provide a new record in a different climatological and hydrographical region (Fig 2). Surface samples (10 ml sedimented for 24 hours) from 30 stations were observed (400x) and quantified [7], but 1ml was used for stations 15 and 16 due to the higher abundance. The concentrations (Fig 2) are the highest recorded for this species in the Pacific: 1,806 cells·ml⁻¹ at station 16 (688 cells·ml⁻¹ at station 15). Concurrently, at station 16 were recorded the lowest temperature and salinity (24.52°C and 34.24 PSU) during the cruise, in agreement with the idea of a relationship of the species with upwelling. Even though *C. balechii* is a

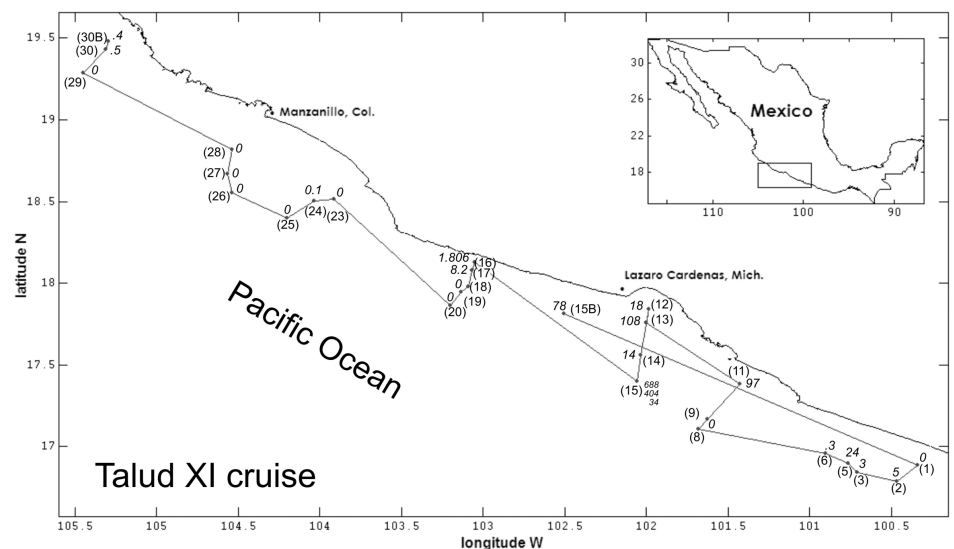


Fig. 2. Map showing the track of the Talud XI cruise. Station numbers in parenthesis and plain numbers are counts of *C. balechii* (cells·ml⁻¹).

non toxic organism, it has been related to fish mortality (together with *C. furca* and *C. falcatum*), mainly due to oxygen depletion.

Acknowledgements

To A. Nuñez-Pasten for his invaluable help in sampling.

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• Italy

What is *Chrysophaeum taylorii* Lewis & Bryan doing in Sardinia (Tyrrhenian Sea, Mediterranean)?

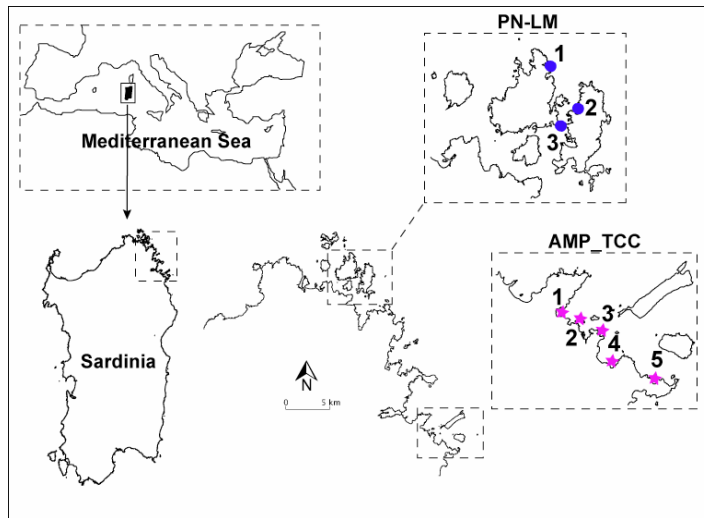


Fig. 1. Geographical localization of the sampled areas interested by the *C. taylorii* proliferation.

We report the first event of an intense growth of *Chrysophaeum taylorii* Lewis & Bryan (Pelagophyceae) [1] on a broad section of the coast (Fig. 1) of eastern Sardinia (Tyrrhenian Sea, Western Mediterranean) during summer 2007. This species was observed once before in a sample collected off Elba (Tuscany Archipelago) in summer 2005, when its growth was very low (personal communication Sartori & Boddi) and without any kind of “public

nuisance”. In contrast, this aspect was important in Sardinia where *C. taylorii* covered primarily hard benthic substrate (mainly granitic), and sandy and plant substrates (predominantly *Posidonia oceanica*) in marine areas of high naturalistic and tourist interests.

C. taylorii has never been reported as a possible agent of benthic Mediterranean mucilage. The hyperproduction of benthic mucilaginous material has been documented since the early 1990s along both Italian [2] and

French coasts [3]. In the former, the phenomenon has been very intense, especially in the area between Sicily and the Tuscan Archipelago and in the latter along eastern Corsica. In these cases the main producer species were *Chrysonephos lewisii* (Taylor) Taylor and *Nematochryopsis marina* (Feldman) Billard (Chrysophyceae) and *Acinetospora crinita* (Carmichael) Kornmann (Phaeophyceae) [4-5].

Our studies were performed at the request of the Marine National Park of La Maddalena (NP-LM; www.lamaddalenapark.it) and Marine Protected Area of Tavolara-Capo Codacavallo (MPA-TCC; www.amptavolara.it), whose watch-monitoring had highlighted the anomalous presence of the benthic mucilage in their areas of interest since the beginning of the summer, but with the maximum extension in August. Furthermore, this exceptional and “strange” situation had generated alarm among local people and tourists, whose reports were numerous.

Our activities were also carried out as a member of the BENTOX-NET (www.bentoxnet.it), the network of

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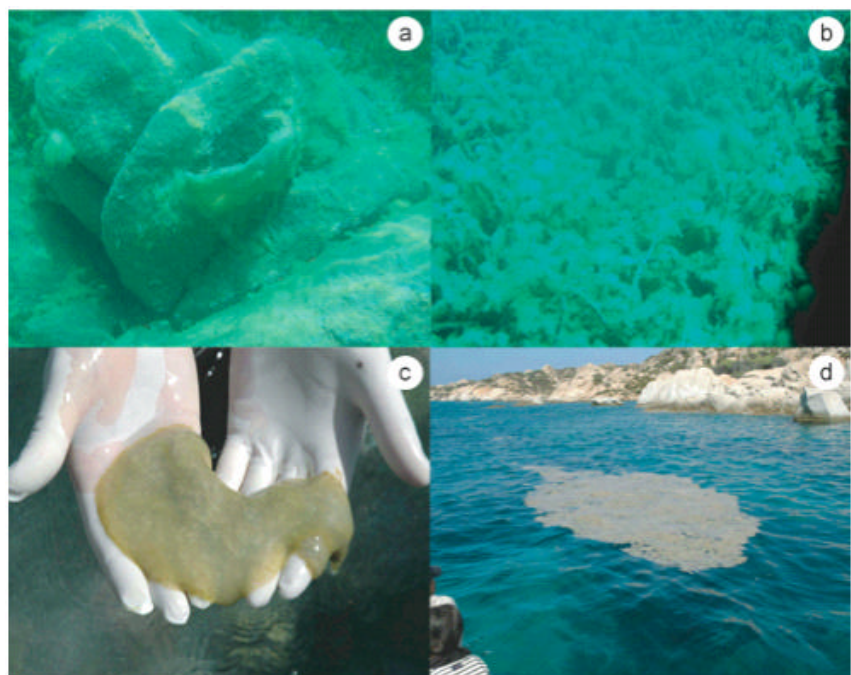


Photo 1. NP-LM. Mucilaginous cover associated to *C. taylorii* growth on different substrates: on granitic rock (a) and on *P. oceanica* (b); c) piece of mucilage and presence of gas bubbles; d) mucilaginous patch on the surface.

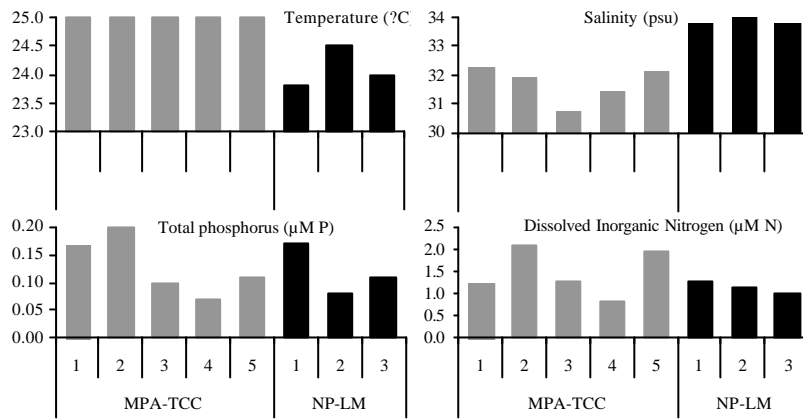


Fig. 2. Environmental data of the two areas sampled.

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Samplings were carried out at 5 stations in the PMA-TCC on the 8th of August and at 3 stations in NP-LM on the 22nd of August (Fig. 1). Water samples were collected for routine analyses (pH, salinity and chlorophyll *a* [6]; nutrients [7] and for phytoplankton and benthic microalgae studies. Floating and substrate samples of mucilage were also collected and fixed with both modified Lugol's solution and neutralized formaline (4%).

The field observations revealed a vast covering of yellow-green-brown mucilage (Photo 1a-b). The mucilaginous layer was 5-8 cm thick. Large portions came off the substrates due to hydrodynamic stress and/or gas bubbles (evident macroscopically; Photo 1c). Currents in the water column transported the smallest portions; the largest accumulated, forming patches on the surface, also transported by wind and currents (Photo 1d). Temperatures were rather uniform (Fig. 2), but higher in PMA-TCC (up to 25.0 °C) than in NP-LM (between 23.8 °C and 24.5 °C). Salinity ranged from 30.72 to 32.28 psu in PMA-TCC and from 33.78 to 34.01 psu in NP-LM. Nutrient concentrations were fairly low (maxima of 0.20 µM P for total phosphorus and 2.09 µM N for DIN in NP-LM and of 0.17 µM P and 1.26 µM N in PMA-TCC); chlorophyll *a* values were up to 0.96 mg m⁻³ in PMA-TCC and 1.11 mg m⁻³ in NP-LM (Fig. 2).

Microscopic analyses of unfixed samples of mucilage showed a layered aspect, as a whole of small layers or

tubes (Photo 2a). Within the mucilage there were many microscopic organisms but the most abundant was *C. taylorii* (Photo 2b). The cells of this species are peculiar with a really "unusual" aspect, as already pointed out [8-9]. The identification was not easy because this species is typical of coral reefs and its presence in the Mediterranean Sea was not expected. In fact, its distribution is reported in the tropical and subtropical Atlantic, Western Pacific, Micronesia and Australian [9-11]. In these areas it is often considered a nuisance species due to the copious production of mucilage. Sparrow and Heimann [12] indicated *C. taylorii* as species in "identity crisis", because of the increase, in the last 10 years, of strong growth in Australian waters.

The event in Tyrrhenian coastal areas of Sardinia during summer 2007 seems to be an example of this kind of exceptional event, but in very distinct biogeographical and environmental conditions, which raises a long list of questions.

Will this exceptional event turn out to be unique for the Mediterranean, or will *C. taylorii* blooms occur in future years? Will this species enlarge its presence? Will it be a nuisance again? Will it affect the benthic habitat and impact local biodiversity? Is *C. taylorii* an allochthonous species introduced into the Mediterranean by human activities? If yes, what was the vector? When did it happen? From which geographical area does it come from?...and many others!

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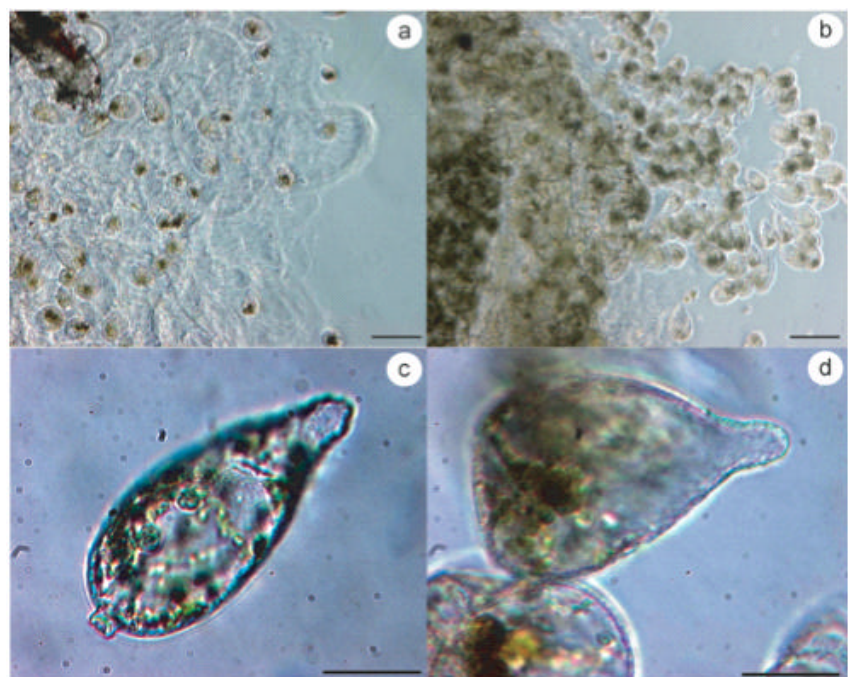


Photo 2. Unfixed samples collected at NP-LM: layered aspect of the mucilage with intertwined cohered stalks having the cells at their end (a and b; bars=100 µm); characteristic shape of cells with their peculiar tubular invagination (c and d; bars=20 µm).

• Mediterranean and Eastern Atlantic

Is *Gambierdiscus* expanding to new areas?

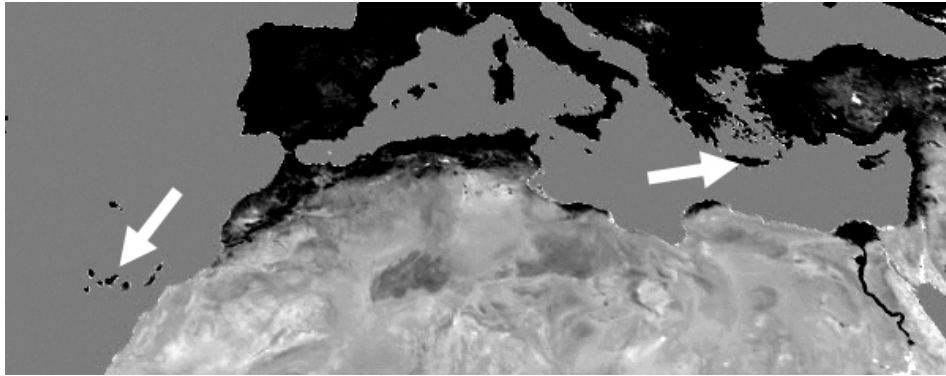


Fig. 1. Map indicating (arrows) the locations of *Gambierdiscus* sp. records (Canary Islands, Spain and Crete, Greece).

Recent findings of *Gambierdiscus* sp. in the Canary Islands, Spain and in Crete, Greece may indicate spreading of this genus into new areas.

The biogeographical distribution of *Gambierdiscus* species was until recently restricted to tropical and subtropical areas, specifically in discrete regions of the Pacific and West Indian Oceans and the Caribbean Sea [1]. The dinoflagellate *Gambierdiscus toxicus* is considered the primary causative agent of ciguatera fish poisoning [2], which is estimated to affect more than 25,000 persons per year [3].

Recently, Pérez-Arellano *et al.* [4] reported ciguatera incidents in the Canary Islands (Fig. 1), while Fraga *et al.* [5] documented the presence of *Gambierdiscus* sp. (Fig. 2) in the same

area, expanding the distribution of this “tropical” genus. In a search for toxic benthic dinoflagellates in Tenerife waters on March 2004, *Gambierdiscus* sp. was found epiphytically in tide pools. In a later survey, it was also found on drifting seaweeds in La Gomera waters. Although the relation between the ciguatera case and the presence of *Gambierdiscus* sp. is not proved, it is striking that both events happened the same year, and were reported by two unrelated teams, a medical group and a marine biology group.

The detection of *Gambierdiscus* sp. cells on the west coasts of Crete (Fig. 1) in September and October 2007 is the first record of the causative agent of ciguatera in the Mediterranean Sea. *Gambierdiscus* sp. cells were found epiphytically on *Padina pavonica*, *Cystoseira* sp., *Corallina* sp. and *Jania* sp. in abundance levels up to 9.01 cells/gr fresh weight. The water temperature during that time ranged between 23.8 and 26.1 °C, while salinity was about 38.5 psu.

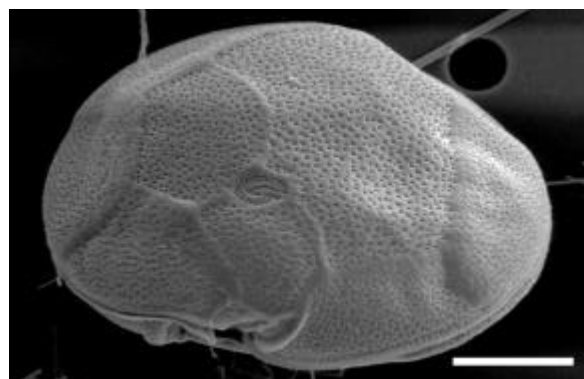


Fig. 2. SEM picture of *Gambierdiscus* sp. from the Canary Islands (scale bar: 20 µm).

Gambierdiscus sp. cells are round to ellipsoidal (Fig. 3), anterior-posteriorly compressed (Fig. 4), with the dorsoventral diameter (DV) and transdiameter (width, W) ranging from 57.12 to 76.16 µm and 52.36 to 78.54 µm, respectively; the DV/W ratio is about 1.02 (±0.07). The apical pore plate (Po) is situated near the center of the epitheca (Fig. 5), but in some cells seems to be displaced towards the ventral area (Fig. 6). Since the initial description of *G. toxicus* [6], five more *Gambierdiscus* species have been described: *G. belizeanus* [7], *G. yasumotoi* [8], *G. pacificus*, *G. australes* and *G. polynesiensis* [9].

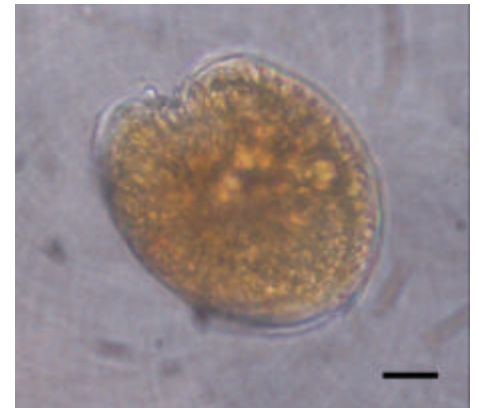


Fig. 3. Live *Gambierdiscus* sp. cell from Crete in apical view (scale bar: 15 µm).

However, Tester *et al* [10] suggested the existence of more species, and asked “if the type *G. toxicus* description included multiple species”; this situation makes the systematics of *Gambierdiscus* fluid. The taxonomy of *Gambierdiscus* is far from clear as new results of molecular and morphological

studies contradict in part those of Chinain *et al* [9, 11].

In both cases, Canary Islands and Crete, *Gambierdiscus* co-occurred with other epiphytic dinoflagellates of the genera *Ostreopsis*, *Prorocentrum*, *Coolia* and *Amphidinium*, as is usual in ciguatera related assemblages.

The occurrence of representatives of what was

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once considered a tropical or subtropical genus, like *Gambierdiscus*, accords with suggestions of several researchers regarding climate change impact on the geographical expansion of tropical microalgae [12], and the “tropicalization” of the Mediterranean Sea in recent years [13]. This fact, as well as the *Ostreopsis* species range expansion in the Mediterranean during the last 5-7 years, and their toxicity [14-18], constitute a serious threat to human health by ciguatera and palytoxin intoxications.

These two new records of *Gambierdiscus* sp. raise some

dinoflagellate communities on either the Canaries or Crete. *Gambierdiscus* sp. was reported in the 1950's in the Cabo Verde Islands by Silva [19] under the name of *Goniodoma* sp.. Her description is so detailed that there are no doubts that it is *Gambierdiscus*. As the Canaries are not so far from the Cabo Verde Islands they could belong to the same population. 2) Are the Greek and Canarian *Gambierdiscus* related? 3) Could *Gambierdiscus* in the East Mediterranean be the result of an invasion via the Suez Canal (a lessepsian immigrant), or through the Strait of Gibraltar?



Fig. 4. Live *Gambierdiscus* sp. cell from Crete showing anterior-posterior compression (scale bar: 10 μ m).

questions. 1) Are they really spreading to new areas, or were they found because they were searched for? To the best of our knowledge there are no published studies of benthic

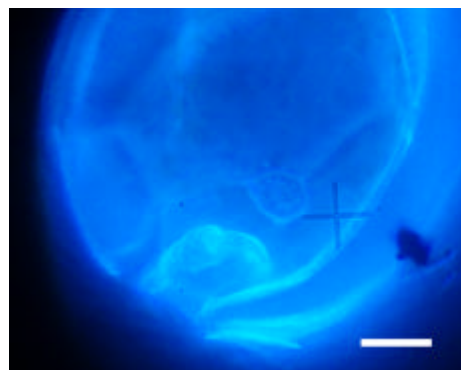


Fig. 6. Calcofluor-stained *Gambierdiscus* sp. cell from Crete showing Po and I' plate (scale bar: 10 μ m).

If the presence of *Gambierdiscus* in the Mediterranean is the result of a recent introduction, it could be a consequence of global warming, expressed in the Mediterranean as an increase in sea surface temperatures, especially in the Levantine Basin. The possible introduction of *Gambierdiscus* to the East Mediterranean as a likely consequence of climate change was discussed a few months before its finding in Crete, at the 4th European Phycological Congress [12].

To provide answers to these questions, morphological, genetic sequences and toxicological analyses of cultured strains of these *Gambierdiscus* species are being carried out.

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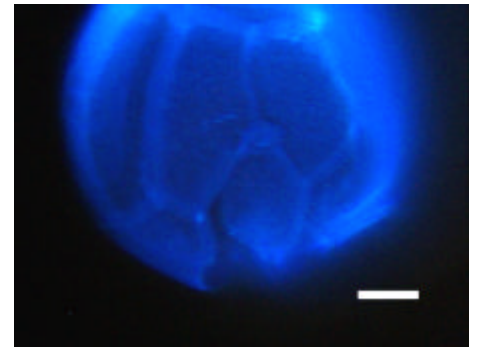


Fig. 5. Calcofluor-stained *Gambierdiscus* sp. cell from Crete showing Po and I' plate (scale bar: 10 μ m).

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Many will remember the original *Poseidon Adventure*, a 1972 disaster film (literally and critically!) about a capsized passenger ship on her way to the scrap yard, which was pushed to her limits by the new owners to save on dismantling fees. The more recent “Poseidon adventure” reported here as the NORCOHAB P-352 cruise aboard the *R.V. Poseidon* (Kiel) (Fig.1) highlights a much more successful (and better directed) effort with significant scientific discoveries to her credit. This cruise, endorsed by GEOHAB and under the auspices of the Core Research Project on *HABS in Fjords and Coastal Embayments*, was undertaken to study the coastal oceanographic processes and mechanisms underlying the dynamics of key toxic bloom species and the biogeographical distribution of their toxins in the water column. Following the prescribed format of GEOHAB research the studies were *international, multidisciplinary* and *comparative* with the ultimate aim of *modeling* dynamics and behaviour. A critical sub-set of target taxa, primarily *Alexandrium* and other toxic gonyaulacoid dinoflagellates, but also *Pseudo-nitzschia* and *Dinophysis* species, were selected for special attention on the basis of their important role in bloom ecology and human health, their likely occurrence in high abundance during the cruise programme, the availability of molecular probes and approaches for their detection and identification and their production of known toxins for on board chemical analysis. Of course we also kept our eyes and minds open to new information, such as an opportunistic search for the source of azaspiracid toxins, of which more later.

The cruise transects were from Bremerhaven, Germany across Dogger Bank with detailed sampling initiated along the Scottish east coast from the Firth of Forth to the Shetland Islands (Fig. 2). For comparison, sampling was

The Poseidon Adventure Redux – NORCOHAB Cruise in North Sea

also conducted within the Norwegian Current on the south coast of Norway and then along the north coast of Denmark. In addition to primary and secondary transects, drift stations were sampled over several days on the Scottish and Danish coasts to obtain time series data within a given water mass. Standard physical oceanographic

historical bloom period of these species (especially *Alexandrium*) based on information kindly provided by the IMR, Flødivigen, Norway (Einar Dahl) and the FRS Marine Laboratory, Aberdeen, Scotland (Eileen Bresnan). Alas cruel Nature saw fit to initiate and then terminate any such blooms prior to our arrival on the scene. An unusually warm



Fig. 1. The NORCOHAB P-352 research cruise vessel at port-of-call in Arendal, Norway.

parameters (temperature: ° C, salinity: psu, s_t) plus current velocity were supplemented with biooptical measurements with multiple profiling fluorosensors and various passive optical profilers (for turbidity and diffuse attenuation), including a hyperspectral radiometer. The use of multiple fluorosensors of differing wavelengths enabled the identification of sub-surface patches attributable to cyanobacteria. Although concentrations of the key toxic microalgae were below detection by passive optical sensors, the optical data provided a more accurate mapping of the higher turbidity and particle loads (including phytoplankton) along the Scottish coast than on the Norwegian coast, presumably due to higher riverine run-off and tidal flux.

The timing of the cruise was carefully planned to coincide with the

winter-spring transition in 2007 appears to have accelerated the typical bloom succession and we were left with mainly grazers and detritivores accompanied by relatively few pigmented dinoflagellates and some rather unhealthy diatom chains at most stations around the North Sea perimeter from early June to early July.

Our working hypothesis was that bloom dynamics of key toxic species in the North Sea are regulated at least as much by “top down” factors such as grazing by copepods and protists as by “bottom up” factors, including light, temperature and nutrients. Since such “top down” field studies are grossly under-represented in modelling simulations of toxic bloom dynamics, we conducted on board grazing experiments to quantify grazing losses of *Alexandrium tamarense*, caused by

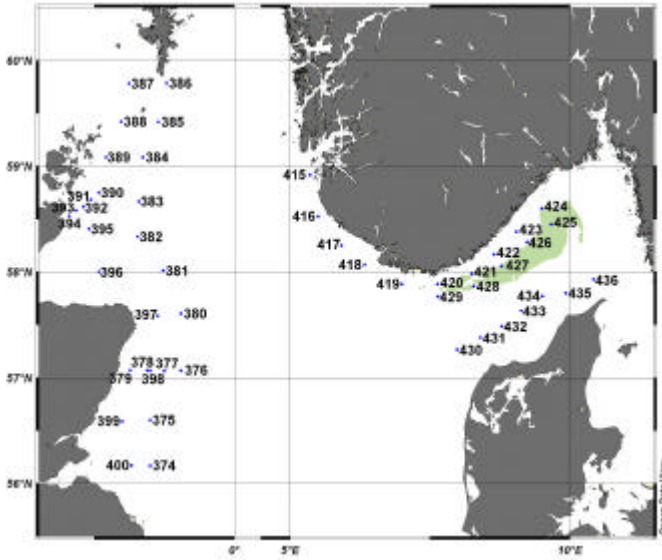


Fig. 2. Map of the primary and secondary transect stations sampled for HAB organisms and their toxins in association with physical and bioptical parameters in the coastal North Sea.

dominant metazoan and protistan grazers. In the absence of natural *Alexandrium* blooms at high cell concentrations, we added fluorescently labelled live *A. tamarens* cultured cells of known toxin content to different size fractions of natural grazer populations. A strong reduction in abundance of labelled cells at stations with high micrograzer abundance showed that *A. tamarens*-specific grazing coefficients may be as high as 2 d^{-1} with almost 80% of total grazing caused by micrograzers in the $<200 \mu\text{m}$ fraction (Fig. 3). The hypothesis about the importance of micrograzers appears to be sustained.

Other on board grazing experiments were conducted to test reciprocal

different copepod species, and was highest for the largest suspension feeder *Calanus helgolandicus*. However, *C. helgolandicus* also seemed best adapted for feeding on *A. tamarens* cells and showed the best fitness among the copepods at the end of the incubation period.

One of the major objectives was to apply liquid chromatography coupled to a highly sensitive triple-quadrupole mass spectrometer (LC-MS/MS) to identify and quantify phycotoxins at trace levels throughout the cruise. With this equipment on board we were able to respond directly to relevant findings on the composition and concentration of toxin in various size-fractions and

effects of grazing on *Alexandrium tamarens* when exposed to three different copepod species selected from natural communities. Specifically, we examined the changes in cellular PSP toxin content and transcriptomic response of *A. tamarens* via microarrays. An apparent toxin defense response from *A. tamarens* against copepod grazing varied among

plankton assemblages from various depths in near-real time (i.e. between stations). The analytical instrumentation provided a gold-mine of information, allowing us to characterize many toxin derivatives (of spirolides, yessotoxins, saxitoxin/gonyautoxins, domoic acid, okadaic acid/dinophysistoxins, azaspiracids) from particulate fractions from the water column at sub-picomolar concentrations. Amazingly, in spite of the rather low concentrations of putative causative organisms, phycotoxins were found at most stations around the perimeter of the North Sea from plankton samples. Such sensitive analytical methods proved crucial to the dynamics determination of the occurrence and fate of toxins in various components of the planktonic food web.



Fig. 3. The predatory tintinnid *Favella ehrenbergii* with an ingested fluorescently labelled *Alexandrium tamarens* cell.

One highlight of the *Poseidon* cruise was the on board determination of azaspiracids in plankton samples, and the subsequent isolation and culture of the causative organism of azaspiracid poisoning (AZP) (no it is *not* *Protoperdinium!*). Of course we were lucky, but following Pasteur's dictum that: "in the field of observation, chance favors only the prepared mind", this was still a worthy accomplishment. The proximal source of azaspiracid (AZA) is a small photosynthetic dinoflagellate of approximately 12 -15 μm long-axis diameter, which was found at stations with high AZA-1 concentrations. The AZA-producing dinoflagellate has now been described morphologically and with reference to molecular phylogenetic analysis for several genes from single cell isolates collected off the Scottish east coast. The major azaspiracid component (AZA-1) was almost ubiquitous in the North Sea, but its abundance varied significantly (Fig. 4). In the western North Sea, AZA-1 was

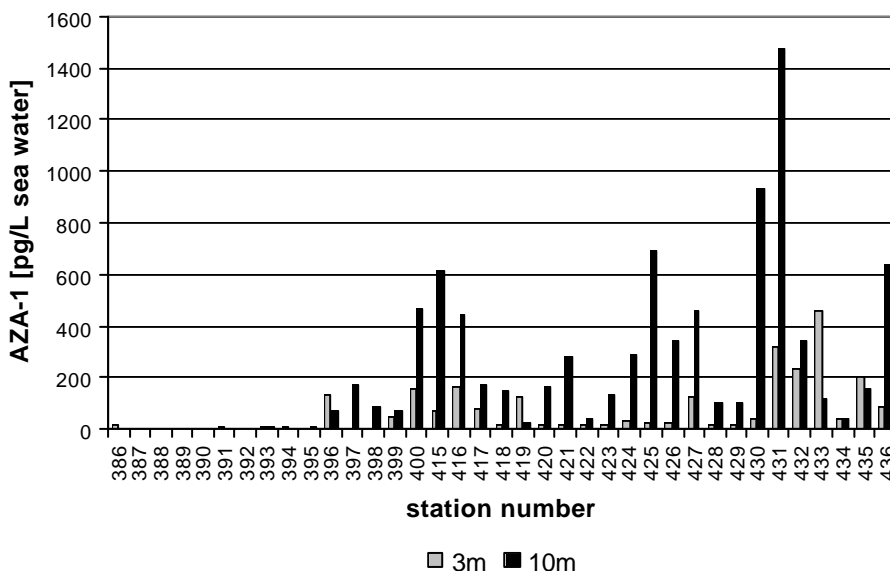


Fig. 4. The concentration of azaspiracid-1 (AZA-1) in the particulate size-fraction 3 – 20 μm collected from two water column depths (3 and 10 m) from stations in the coastal North Sea.

• USA

First toxic *Dinophysis* bloom observed in the Gulf of Mexico, USA

Dinophysis acuminata (Dinophyceae, Dinophysiales) is a thecate dinoflagellate that produces okadaic acid, the toxin responsible for diarrhetic shellfish poisoning (DSP). Although toxic blooms of *D. acuminata* have been a serious problem in Europe [1], relatively few reports of human illness have been linked to DSP in the US [2]. Blooms of *D. acuminata* have been reported on the US east coast [3, 4], but cell concentrations were not high

most abundant along the southern Scottish coast, but further north around the Orkney and Shetland Islands, AZA-1 was only detected at trace levels. In general, AZA-1 concentrations were higher in the eastern North Sea with highest levels found in the southern Skagerrak.

Further NORCOHAB expeditions in the North Sea are planned under the comparative GEOHAB framework. Following the proof of concept for integrated operation of chemical analytical instrumentation and biooptical profilers for detection of HABs and their toxins in near-real time, we will continue work on the biogeographical distribution of toxigenic organisms and their toxins in relation to food web transfer and chemical ecology of key species in coastal waters.

Reported by A. Cembella with contributions from B. Krock, U. Tillmann and U. John, AWI students and technicians. Thanks to B. Cembella (Optimare AG) and O. Zelinski (Bremehaven Hochschule) and co-workers.

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We thank Captain H. Hansen and the crew of the R.V. Poseidon.

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enough and/or populations did not persist long enough to result in significant shellfish intoxication. Consequently, no routine monitoring of *Dinophysis* or

dinoflagellate that blooms periodically in the Gulf and that can cause neurotoxic shellfish poisoning. No *K. brevis* blooms were observed in the region this year,



Fig 1. Port Aransas, Texas, on Mustang Island, a barrier island along the coast of central Texas, Gulf of Mexico, USA.

other DSP-related HAB species is conducted in the US [2].

Now, for the first time, both elevated concentrations of *D. acuminata* and DSP intoxicated shellfish have been detected in US waters, in central Texas in the western Gulf of Mexico (Fig. 1), requiring shellfish beds to be closed to harvesting.

Early warning of this first toxic *Dinophysis* bloom was provided by a new automated particle imaging system [5]. Imaging FlowCytobot combines video and flow cytometric technology to capture images for plankton identification and to measure chlorophyll fluorescence associated with each image. From the high resolution images, organisms ranging from 10 to ~100 μm can be identified, often to genus or even to species.

The instrument was installed in the pier laboratory at the University of Texas-Marine Sciences Institute (Port Aransas, TX; Fig. 2) in September 2007, as part of a NOAA/CICEET project to monitor *Karenia brevis*, another toxic

but the imaging approach's flexibility allowed this unexpected plankton species to be studied equally well.

In early February 2008, *Dinophysis* cells (1-5 per mL) were detected through manual inspection of cell images. By late February estimates were >100 per mL (Fig. 3). Because these levels exceed the typical background level for the genus in this region, manual sampling was conducted by the Texas Department of State Health Services (DSHS) to confirm the species, toxicity and abundance in coastal waters from Port Aransas harbor and adjacent bays. The bloom consisted primarily of *D. acuminata* (Fig. 3), although *D. fortii* and *D. caudata* were also present at much lower abundance. Okadaic acid was confirmed in plankton samples collected 25 February by liquid chromatography/mass spectroscopy. Four days later, collection and analysis of shellfish samples showed significant accumulation of DSP toxins. On 7 March, the Aransas, Corpus Christi, and

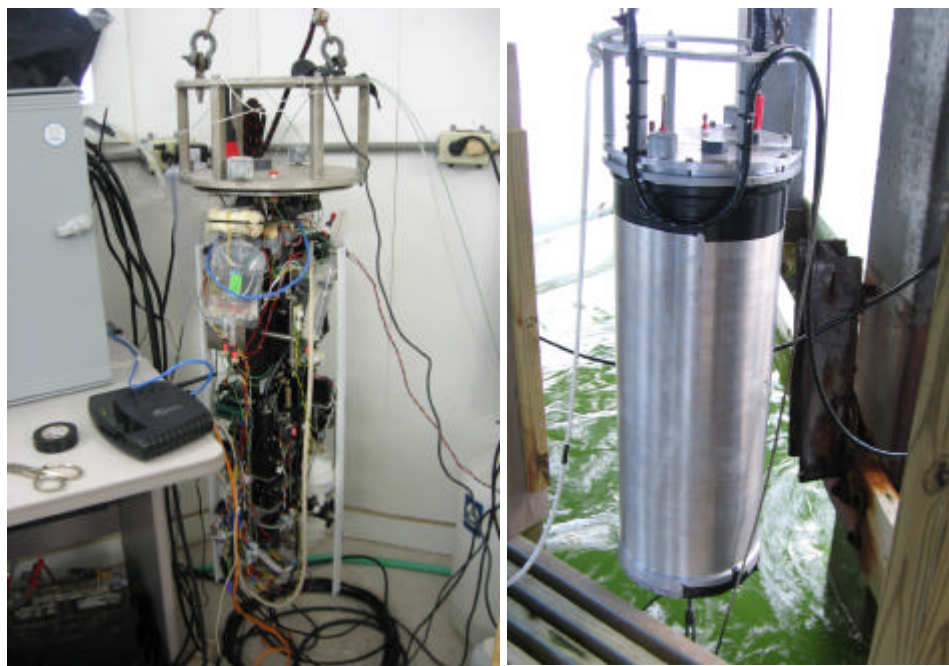


Fig. 2 Imaging FlowCytobot (version 3) as constructed for deployment from the Port Aransas Pier. Internal view (left) while being configured and aligned in the laboratory, and after enclosure in the pressure housing (right) designed for submersible applications.

Copano Bays were closed to shellfish harvesting, and shellstock harvested on or after 1 March were recalled [6]. Timing of this closure and recall of local oyster harvests was fortunate as it allowed organizers of a scheduled Oyster Festival – an annual community event that typically attracts 30,000 people—to purchase oysters from elsewhere.

Monitoring by Texas DSHS continued throughout March in the Port Aransas region. During this time, observed *Dinophysis* concentrations remained >5 cells mL^{-1} in Aransas and Copano Bays, but the bloom did not appear to impact other regions of the

Texas coast. At the Port Aransas pier, *Dinophysis* counts from Imaging FlowCytobot exceeded 20 cell mL^{-1} during most of March, with the exception of a few days mid-month when counts decreased to <1 cell mL^{-1} (Fig. 3). Imaging FlowCytobot sampling shows that the bloom is continuing in April and the 3 bays remain closed to shellfish harvesting.

Because Imaging FlowCytobot has collected over 31 million images at the Port Aransas pier since 1 January 2008, automated image analysis and classification is essential for application to taxon-specific bloom detection. A preliminary set of images selected from

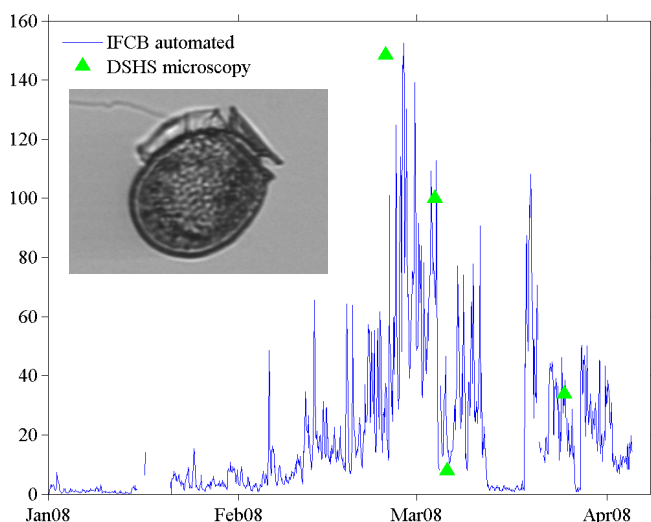


Fig. 3. Preliminary time series of *Dinophysis* abundance at the Port Aransas pier from automated analysis and classification of images collected by Imaging FlowCytobot. DSHS counts from conventional microscopic analysis are shown for comparison. Inset:

the time series have been inspected, manually identified, and then used to train a machine learning algorithm for automated classification, as described in Sosik and Olson [7]. This analysis approach not only provides taxon-specific abundance estimates at times of specific interest, but also permits detailed bloom dynamics to be investigated over extended periods of time (Fig. 3). From this information it will be possible to study aspects of bloom ecology such as initiation and patchiness.

This successful event response has demonstrated that continuous and automated methods for monitoring coastal waters can provide real-time detection and early warning of harmful algal bloom (HAB) events.

Further development of automated image classification, involving extraction of image features and supervised machine learning algorithms, will enhance our ability to monitor the abundance of individual phytoplankton taxa at relevant temporal scales for HAB prediction.

Acknowledgements

We thank E. Buskey, C. Hyatt and the Mission-Aransas NERR program, T. A. Villareal, K. Wiles and Texas DSHS for additional sampling and cell counts.

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• India

“Green tide” of *Noctiluca miliaris* in the Northern Arabian Sea

On the west coast of India, discoloured water events caused by a variety of organisms and sometimes associated with mass mortality of local marine fauna, have frequently been reported [1-7].

During an oceanographic cruise onboard *FORV Sagar Sampada* in the second week of March 2007, an intense green colouration accompanied by soup like consistency was observed in the upper layers of Northern Arabian Sea. The phenomenon in its full intensity with intermittent patches were found to be spread over an area of around 30 km². The physical and chemical characteristics of two stations were sampled along with a non-blooming region. The location of the blooming area was in Lat. 21° 50' 32 N, Long. 66° 09' 38 E and Lat. 21° 50' 63 N, Long. 67° 03' 55 E, and that of the non-bloom region was in Lat. 21° 00' 33 N, Long. 65° 01' 21 E. The bloom organism was *Noctiluca miliaris* Suriray, a nonphotosynthetic, heterotrophic and phagotrophic dinoflagellate. Innumerable motile prasinophyte endosymbionts (*Pedinomonas noctilucae* (Subr.) Sweeney) were found within the *Noctiluca* cells giving the deep green hue to the surface waters.

Water samples were analysed for pH, dissolved oxygen (DO), chlorophyll *a* and nutrients according to Parson *et al* (Table 1). The CTD profiler recorded temperature and salinity. Phytoplankton samples preserved in 1-3% neutralised formalin were used for qualitative and quantitative analysis.

Cells have a size range from 200-

600 µm in diameter. *Noctiluca* density was 4x 10⁶ cells m⁻³ in bloom stations. *Noctiluca miliaris* was found along with 19 other species of phytoplankton, which included nine species of diatoms (*Chaetoceros brevis*, *C. didymus*, *Navicula digitoradiata*, *N. elegans*, *N. lyra*, *Rhizosolenia alata*, *R. hebetata*, *R. imbricata*, *Thalassionema nitzschioides*) and ten species of dinoflagellates (*Ceratium furca*, *C. trichoceros*, *C. vultur*, *Diplopeltopsis minor*, *Diplopsalis lenticula*, *Prorocentrum micans*, *Protoperidinium obovatum*, *P. oceanicum*, *P. pyriforme*, *P. steneii*). Among the diatoms, *Rhizosolenia* formed the major constituent. Co-occurrence of *Noctiluca miliaris* and *Rhizosolenia* spp. has been reported previously [8].

Dissolved oxygen values ranged between 4.96 and 5.09 ml L⁻¹. Inorganic phosphate was below the detection limit, presumably due to consumption by the bloom species. Nitrate values were higher and might be due to the presence of *Noctiluca*. Prasad and Jayaraman [9] reported a similar trend in nitrate concentration during *Noctiluca* swarming. The relatively high chlorophyll *a* value at the bloom stations (21.9 and 21.3 mg m⁻³) compared to the non-bloom station (0.97 mg m⁻³) is due to the flagellates associated with *Noctiluca*. Relatively stable low temperature, high salinity, calm sea and muggy weather are known to favour proliferation and blooming of *Noctiluca*.

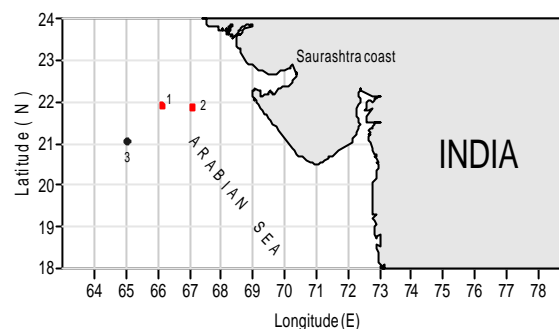


Fig. 1. Map of study area.

Zooplankton biomass was very high at the bloom stations (6090 ml 1000 m⁻³ and 7790 ml 1000 m⁻³) compared with the non-bloom station (985 ml 1000 m⁻³). Copepoda, Chaetognatha, Cladocera, fish eggs, Amphipoda, Heteropoda, *Lucifer*, *Oikopleura*, salps, doliolids, and siphonophores were abundant.

Surveys in this area during the last seven years have shown that *Noctiluca* blooms in the Northern Arabian Sea every year during this period. The regular occurrence of the bloom in the same area, the role of wind in nutrient enrichment, and the role of the symbiont *Pedinomonas noctilucae* in *Noctiluca* blooms need detailed investigations.

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Table 1. nd* - not detectable

Parameter	<i>Noctiluca</i> bloom stations		Non bloom region
	Station 1	Station 2	Station 3
Temperature (°C)	25.71	25.56	26.01
Salinity (ppt)	35.96	35.98	36
pH	8.33	8.31	8.36
Dissolved oxygen (ml l ⁻¹)	4.96	5.09	5.38
Nitrate (µmol l ⁻¹)	4.735	5.038	1.678
Phosphate (µmol l ⁻¹)	nd*	nd	nd
Silicate (µmol l ⁻¹)	0.201	0.015	1.602
Chlorophyll <i>a</i> (mg m ⁻³)	21.9	21.3	1.24
Cell count (Cells m ⁻³)	4x 10 ⁶		<i>Noctiluca miliaris</i> absent

Book review

Thronsen J, Hasle GR and Tangen K 2007. *Phytoplankton of Norwegian Coastal Waters* (Almater Forlag AS, Oslo) 343 pages, b&w illustrations, photos, color plates, soft cover.

Price £ 96 at www.nhbs.com/title.php?tefno=157807. Also available from Balogh International (www.balogh.com/~balogh) for \$ 205. ISBN (13) 978 82-7858-086-8.

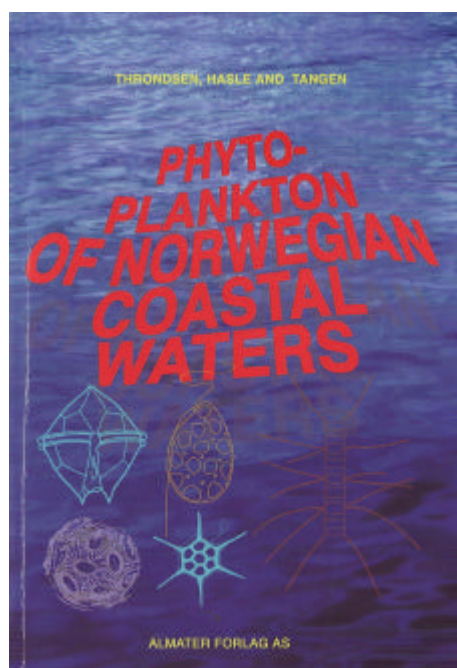
A few years ago, on a visit to Oslo, Jahn Thronsen showed me a book to which he contributed: "Norsk Kystplankton Flora". I marveled at the wonderful drawings of phytoplankton it contained. A year later, on a visit to Roscoff, Jahn pulled out a copy and offered it to me, autographed. I was thrilled, but at the same time frustrated because Norwegian does not make easy reading, at least for me. I was wondering whether this book would someday be translated into English. This is now done and the result is outstanding.

This book is limited to the phytoplankton of coastal waters around Norway but still should be extremely useful for European readers since Norwegian coasts are influenced by quite different water masses including Arctic, Atlantic, and Baltic ones. Still, as Jahn Thronsen cautions in his introduction, "users abroad (should) be especially careful when identifying the species and seek comparison and advice from local floras and other taxonomical publications".

Those familiar with the book "Identifying Marine Phytoplankton" (IMP) coordinated by Carmelo Tomas, probably currently one of the most used taxonomical reference for phytoplankton, will feel at home, since the three authors of the present book were also major contributors to IMP. The book is organized along division and classes with major sections dealing with dinoflagellates, diatoms and coccolithophorids. However, while in IMP, flagellates are grouped together at the end of the book, here they are grouped according to divisions. For example, Chrysophyceae are treated next to diatoms in division Heterokontophyta and Pavlophyceae next to coccolithophorids in division

Haptophyta. This is much more coherent but make things a little more difficult for novices. The taxonomy is really up to date as recently established groups such as Pinguiphyceae are mentioned, even though they do not contain any genera reported for Norwegian waters. Even species that have been described after the publication of the Norwegian edition (e.g. the Dictyochophyceae *Verrucophora farcimen*) have been included in the English version.

For each major algal class, a compact introduction is provided summarizing the systematics of the



class, the cell morphology and the specialized nomenclature (for example plate patterns for dinoflagellates); A general key follows for the class. Each species is then described and illustrated by at least one drawing and sometimes by a microscopy picture. Information on the species habitat in Norwegian coastal waters is also provided. For many genera, detailed keys are also provided. This organization (i.e. description and illustration grouped together) is in fact more handy than the one used in IMP where illustrations are organized in plates resulting in physical separation between the species descriptions and drawings, forcing the reader to move back and forth between the pages of the book.

As with all productions, there are of course a few things that could be improved. A short presentation of the

current view of protist phylogeny could have been useful to illustrate the fact that phytoplankton is highly diverse and spread over many protist groups. Also it would seem more logical to place upfront methodological considerations concerning plankton sampling and plankton preparation for microscopy, instead of sending them to the end of the book. Concerning the material covered by the book, the inclusion of some heterotrophic groups such as choanoflagellates or ciliates could be questioned. Obviously, they are often present in phytoplankton samples and the book will help people to identify them. However since the coverage of these groups is much less exhaustive than that of the photosynthetic groups, it may induce some readers to downplay their actual diversity or to wrongly identify some species. Finally there are a few new minor errors or omissions. For example p.12, the Prochlorophyta (cyanobacteria containing chlorophyll *b*) have been known for quite a while to be polyphyletic, all genera are in fact *bona fide* cyanobacteria, and Prochlorophyta are best forgotten. Also *Telonema* (p. 279) is no longer *incertae sedis* but belongs to a new phylum called Telonemia.

I think all plankton taxonomists should be very grateful to Grete Hassle, Karl Tangen and Jahn Thronsen for having offered to the scientific community and beyond, this compilation resulting from the many years they spent looking under the microscope, taking pictures and making these wonderful drawings. If phytoplankton taxonomists

PHYCOTOXINS

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FAO-SIDA-UNESCO-MIRCEN International Workshop on Shellfish Safety from Harmful Algae and Biotoxins

Harmful algal blooms and shellfish toxicity are a global problem affecting shellfish safety and international trade. It is estimated that about 2000 cases of shellfish toxicity occur annually with 15% mortality. The economic impact of this problem has been estimated to be US\$ 82 million in United States alone. In south and south-east Asia, there have been several reports of fish kills and shellfish poisoning. India had two recorded outbreaks of paralytic shellfish poisoning (PSP) and toxin involved in diarrhetic shellfish poisoning (DSP) has been detected in shellfish in India. Malaysia, Indonesia and China have experienced outbreaks of PSP. However, the dinoflagellate species involved in toxic episodes vary in the region. Bivalve production by aquaculture has been growing and in 2005, 84% of bivalve production came



from aquaculture. China is emerging as a major producer of bivalves; China presently accounts for 80% of world bivalve production by aquaculture. To ensure the safety of shellfish, it is important to have monitoring programmes for harmful algae and biotoxins. However, a major concern has been the expertise required to identify the small proportion of toxic species in water samples. Technology for detection of biotoxins has been undergoing rapid developments with improvements in analytical capability. Sophisticated analytical procedures such as Liquid Chromatography with tandem mass spectrometry (LC-MS-MS) and immunoassays are being applied for toxin detection.

Considering the global importance of the problem and the need for capacity building in this area, an International workshop on “Safety of Shellfish from Harmful Algae and Biotoxins” was held at the UNESCO Microbial Resources Center (MIRCEN), College of Fisheries, Mangalore, India from January 21 to 25, 2008. Dr. Indrani Karunasagar was the Workshop Coordinator. The workshop was jointly sponsored by FAO, Swedish International Development Agency (SIDA) and UNESCO MIRCEN. The workshop included lectures and practical sessions on identification of harmful

algae and their cysts; biotoxins and their detection by bioassay, immunoassay, HPLC and liquid chromatography mass spectrometer (LC-MS) techniques, and monitoring as a component of integrated shellfish safety management. The workshop included 23 participants from Vietnam, Malaysia, Indonesia, Thailand, China, Morocco, Yemen, Sweden and India. The faculty for the workshop included Dr. Y. Fukuyo from University of Tokyo, Japan, Dr. J. Larsen, IOC Center on Harmful Algae, Copenhagen, Denmark, Dr. Dave Clarke from Marine Institute, Galway, Ireland, Dr. Iddya Karunasagar from FAO, Rome, Dr. Ann-Sofi Rehnstam-Holm and Dr. Anna Godhe from Sweden, Dr. Indrani Karunasagar and Dr. B.B. Nayak from India. The sessions were highly interactive and the Workshop material included Manuals and CDs. In addition to lectures and practical sessions, the programme included presentation of the status of work on harmful algae and biotoxins in the countries represented in the workshop. Experiences from 10 countries were presented.

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from other countries (and I am thinking here of my own country, France) had left similar legacies, our field would be much richer. One would only wish that all this information soon becomes available on the Internet either through a dedicated website or even better through collaborative initiatives such as Plankton*Net (e.g. planktonnet.sb-roscoff.fr) or the Encyclopedia of Life (www.eol.org). Such dissemination would probably not hurt book sales since people who can afford it would want to buy a hard copy, but would help many field taxonomists throughout the world who may have little access to books.

In conclusion, although I have not yet used this book myself to practice phytoplankton identification, I am deeply convinced that this book is soon going to be sitting next to numerous microscopes (and probably often stolen...) along with IMP, and I can only highly recommend purchasing it.

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Towards measuring mucus events

Are aquatic “mucus”, “slime” and “froth” events, like harmful algal blooms [1], increasing worldwide in both extent and intensity? The last six numbers of *Harmful Algae News* (HAN) Nos. 31-36 (2006-2008), report the following mucus events:

- “Cream or beige coloured foam” produced by *Alexandrium monilatum* in Costa Rica [2];

- “Abundant foam forming stripes parallel to the coast” in a bloom of *Chattonella marina* and *C. cf. ovata* in the Gulf of California, Mexico [3];

- Three reports of benthic, pelagic and neustonic occurrences of polymer matrices apparently produced by *Ostreopsis ova* as well as *O. cf. siamensis* in the Mediterranean. Cogestri *et al.* [4], showed mucilage flocs and cells detaching from the substrate and floating in the water, while Totti *et al* [5] found *O. ova* to produce a brown velvet-like mat, shown by scanning electron microscopy to consist of a matrix of fibrils, covering rocks, seaweed and mollusc shells. Barone [6], furthermore, demonstrated that *O. ova* cells use this network, to entangle and trap prey, rather like spiders use their webs except that, unlike spiders, they do it collectively. These webs may also entrap potential predators, thus protecting their creators;

- Along a 65-km stretch of the coast of NE Spain large patches of floating green-brownish mucus were reported [7]. Dominant organisms in the mucus included *Gonyaulax fragilis* and *Pseudo-nitzschia*, but since *G. fragilis* was shown to secrete mucoid material from its apical pore, it seems likely that this species may be the origin;

- In a report on *Microcystis aeruginosa* in a city reservoir in Colombo, Sri Lanka, it is pointed out the species may form thick layers or mats at the water surface [8], underlining the inevitability that surface algal and cyanobacterial blooms to some extent control air-water fluxes. There are three examples in the present issue:

- “Soup-like consistency” in green *Noctiluca* blooms covering around 30 km², in the Arabian Sea, off NW India [9];

- The pelagophycean *Chrysophaeum taylorii* was found in 2007 investing substrates of rock, sand and seagrass on the NE coast of Sardinia, with a yellow-green-brown mucilaginous cover up to 5-8 cm thick. Bubbles formed in this cover and caused it to float up to the surface, producing visible patches. It was sufficiently abundant to alarm tourists and local people. [10];
- In the Sea of Marmara, Turkey, from December 2007 to March 2008, white mucilaginous aggregates were noticed near the surface. SCUBA divers then found that similar mats also occurred on the bottom, covering benthos [11]. This phenomenon, newly reported for the area, was compared to that of “mare sporco” that has occurred sporadically for centuries in the Northern Adriatic [12].

These reports of mucous events linked to algal blooms are of course only a tiny proportion of the literature. Much more exists on this subject, and still more on the properties of small mucous particles, including transparent exopolymeric particles (TEP) and marine and lake snow [13] as well as models of their flocculation and break-up [14-16].

Progress has been better in understanding and measuring the mechanical properties of the air-water surface through 2-dimensional (2D) shearing [17], and more particularly during 2D compression-dilation cycles, accompanied by electrochemical measurements showing how the molecules restructure and exchange with the bulk phase [18, 19].

Yet it is in the bulk phase of the medium, where changes in the mechanical properties of the water are likely to impact plankton ecology most. Progress in this field is slow both at scales of mm to cm [20-23], and, using at atomic force microscopy at those of polymer molecules [24], and little understood by many HAB research teams.

Like marine algal slime, blood too is thicker than water, and huge advances were already being made nearly 30 years ago in the rheology of blood [25]. Qualitative descriptions of

mucus are extremely valuable, but were sufficient training in marine rheology methods available to teams working on HABs, we might see comparable advances in understanding HAB dynamics. To this end, I propose that a Working Group on marine mucus properties be set up. This will be particularly focussed on algal blooms, as it is where algae are most concentrated that they have most potential to thicken the water. Anyone interested is welcome to contact the author directly, Esther at the XVth International Conference on Rheology, Monterey, USA, or the 13th International Conference on Harmful Algae, Hong Kong, November 3-7 2008.

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Reflections from the GEOHAB Scientific Steering Committee: Ocean Colour Remote Sensing and Harmful Algal Blooms: a Rush of Blood?

Development and application of optical sensors and remote sensing is central to many marine research programmes, agencies and activities. GEOHAB addresses development of improved remote observation systems for harmful algae, and activities have included a science workshop on real time observation systems for harmful algae and publication (early 2008) of an UNESCO monograph on oceanographic methodology entitled 'Real-time Coastal Observing Systems for Marine Ecosystem Dynamics and Harmful Algal Blooms: Theory, Instrumentation and Modelling' edited by Marcel Babin, Collin Roesler and John Cullen. A critical role of these activities is to identify gaps in knowledge, and to stimulate development and improved application of relevant techniques.

Ocean colour remote sensing offers considerable potential for the observation of harmful algal blooms (HABs); such potential remains largely unfulfilled due to the sizeable uncertainties associated with applications in the coastal zone. Recent studies suggest that space-based ocean colour sensors allow the routine coastal detection of high biomass phytoplankton blooms, or the identification of phytoplankton functional types.

However, there are still fundamental problems with the application of rigorous ocean colour techniques in optically complex coastal waters. The ocean colour and harmful algal bloom scientific communities would gain a great deal by identifying and addressing these problems, rather than prematurely adopting under-developed techniques.

Ocean colour radiometry has several fundamental steps: Radiance is measured at the top of the atmosphere; to get the oceanic signal the atmospheric signal has to be removed (typically accounting for more than 80% of the total signal). The derivation of geophysical parameters such as chlorophyll *a* concentrations from this corrected signal, uses various empirical or analytical algorithms. Further ecologically meaningful products, such as anomalies, can then be calculated. In the HAB-prone coastal zone, all of these steps suffer from considerable uncertainty. Atmospheric correction schemes considered suitable for the open ocean often perform poorly in turbid, atmospherically complex coastal waters where a lack of suitable validation data hampers our ability to analyse and improve these vital procedures. In addition, an incomplete understanding of the underlying causes

of ocean colour variability means that the various techniques typically employed to derive HAB products are at best ambiguous.

There are five broad techniques employed with regard to HAB or phytoplankton assemblage type detection:

1. Bright water is historically often used to discriminate non-harmful coccolithophore blooms and can be used for operational, i.e. non-quantifiable, detection of abnormally high turbidity waters that can circumstantially be associated with given groups such as harmful cyanobacteria (e.g. *Nodularia spumigena* in the Baltic Sea).

2. Discrimination based on spectral signature of targeted phytoplankton species or groups is successful for a small number of cases e.g. cyanobacterial blooms containing the spectrally distinctive phycocyanin pigment (see Figure 1).

3. Several approaches based on chlorophyll anomalies relative to climatologies have been proposed as a means to raise warning of possible high biomass HAB events. While operationally promising to some extent, those approaches are subject to ambiguities caused by the anomalies being partially or wholly due to variability

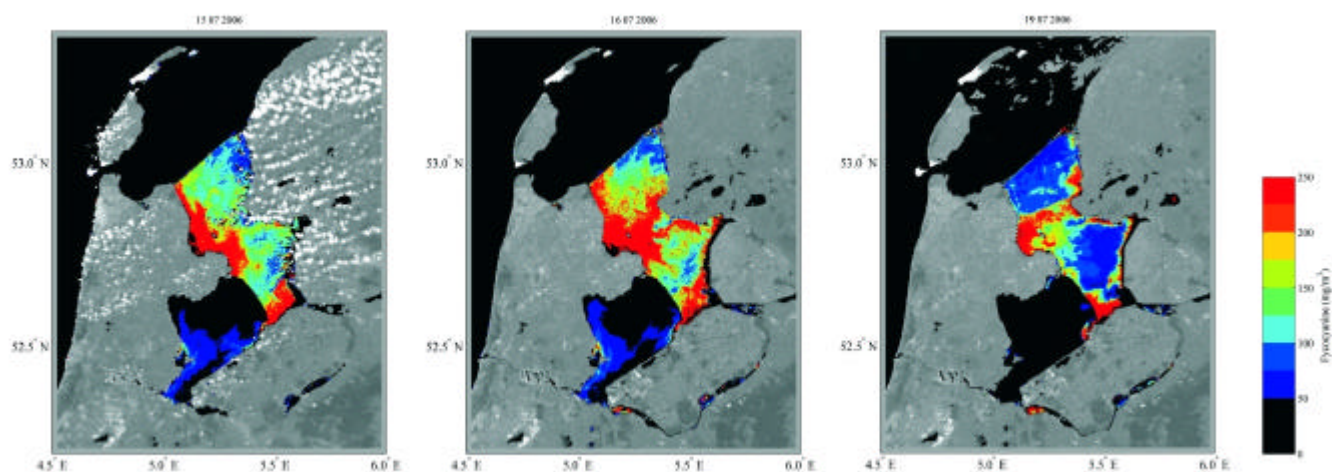


Fig. 1. Spatial distribution of the phycocyanin pigment in the Netherlands eutrophic Lake IJsselmeer (courtesy of Stefan Simis). Phycocyanin is associated with the presence of cyanobacteria such as *Aphanizomenon flos-aqua* and *Microcystis* sp. Those results were obtained using the algorithm from Simis et al. (2007) applied to imagery from the European sensor MERIS (Full resolution), collected over three days during the July 2006 heatwave. This short time series illustrates the potential of ocean colour remote sensing for monitoring with great spatial detail the temporal evolution of highly dynamic phytoplankton blooms. Reference: S.G.H. Simis, et al., Remote Sensing of Environment 2007, 106: 414-427.

in atmospheric properties, dissolved organic colour, or non-chlorophyllous particles. As illustrated in Figure 2, the use of ocean colour to detect phytoplankton is often, in coastal waters, limited by the fact that other seawater constituents (e.g. coloured dissolved organic matter) are present in such high quantities that they simply hide any other constituents. Other approaches alternative to the use of the blue and green part of the reflectance spectrum may allow circumventing this problem.

4. Analytical algorithms, based upon inverse optical modelling of water column constituents, are promising as alternatives to standard geophysical algorithms but also suffer from poor atmospheric correction, and an incomplete understanding and parameterisation of the variable optical properties of phytoplankton and other water constituents.

5. The use of anomalies in the reflectance spectrum has recently been proposed to distinguish broad phytoplankton groups, based primarily on statistical relationships with chemotaxonomic data derived from High Performance Liquid Chromatography (HPLC) pigment measurements. However, recent studies suggest that the variability in the reflectance targeted by such algorithms is primarily associated with the varying concentration of non-algal constituents and that such methods are not robust.

Aside from atmospherically-related problems with absolute radiometric measurements, many of the problems lie in the fundamental question of identifying phytoplankton groups or species, and whether such groups can be distinguished using ocean colour. For example, HPLC pigment methods are potentially a very precise means of measuring phytoplankton pigment concentrations, but variations in accessory pigmentation are often only a minor source of ocean colour variability, and the chemotaxonomic interpretation of HPLC data can often be ambiguous. Allometric or size based ocean colour techniques also show promise, but whilst phytoplankton size variability across marine systems is characterised to some degree, our ability to translate this into useful information

is limited by a lack of routine particle size observations. There is therefore a need for the ocean colour community to engage constructively with the phyecological community, to adopt less ambiguous routine means of identifying phytoplankton community structure, e.g. traditional cell counts, flow cytometry, genetic techniques, or cell sizing techniques if it is to pursue HAB or phytoplankton functional type research in an ultimately meaningful way.

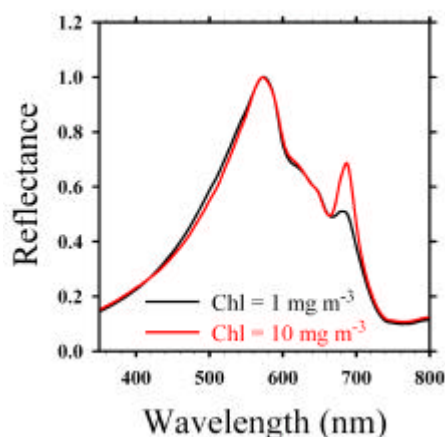


Fig. 2. Spectra of the water-leaving reflectance calculated using a radiative transfer code (Hydrolight 4.2) and optical properties for coloured dissolved organic matter and suspended non-algal particles typical of the Baltic Sea. Two different concentrations of chlorophyll *a* were used to set the optical properties of phytoplankton. Note that the two spectra were normalized to their respective area under the curve. This figure illustrates the fact that increasing the amount of phytoplankton by an order of magnitude in such coastal water has only subtle effects on the shape of the reflectance spectrum in the blue and green domains. It has, however, a very significant impact on reflectance in the red part of the spectrum where chlorophyll *a* fluorescence stimulated by sunlight can be readily observed (peak centred around 683 nm).

The way forward

The way forward lies in a suite of measures: true validation rather than rationalisation of results as plausible; sensitivity studies exploring the causal influences and assemblage detection limits across a wide variety of water types; a drive to put confidence limits on ocean colour products from a regional perspective. Consequently there is a requirement for a skill assessment strategy for ocean colour products which requires both the quantification of

product uncertainty and the definition of minimum acceptable performance levels. Ocean colour techniques that do not outline plausible causal mechanisms may be of regional operational value, but are unlikely to be useful across a variety of marine ecosystems, and are therefore of limited value in advancing our understanding and global observation of HABs as ecologically prominent phenomena. The greater involvement of multi-disciplinary scientific communities is extremely important – most notably HAB phyecologists and oceanographers, in addition to ecophysiological modelers, who have specific and theoretically demanding requirements with regard to error analyses. The challenge is to give these distinct scientific communities some degree of ownership of ocean colour products as experimentalists. Considerable gains can be made from better understanding mechanisms underlying variability in the red portion of the ocean colour spectrum, most sensitive to phytoplankton backscattering at high biomass as demonstrated by research in hypertrophic freshwater systems. Red wavelengths may also yield valuable information on physiology and phytoplankton functional type through sun-induced fluorescence, currently measured by several ocean colour sensors but remaining very much an underexploited phenomenon. However, the starting point almost certainly lies not only in the ocean, but in the ocean-atmosphere system: validation and improvement of the atmospheric correction across a variety of coastal regions is critical.

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GEOHAB developments

Five new people were welcomed onto the Scientific Steering Committee (SSC) of GEOHAB by chairman Robin Raine (Ireland) at the recent SSC meeting in Annapolis, USA between 9-11 April 2008. Elisa Berdalet (Spain), Suzanne Roy (Canada) and Icarus Allen (UK) now act as liaison between the SSC and the GEOHAB Core Research Projects on Stratified Systems, Coastal Bays and Fjords, and Eutrophic Systems respectively. Stewart Bernard (South Africa, remote observations) and Liam Fernand (UK, physical oceanography) will participate on a Task Team on Observations through which formal links between GEOHAB and GOOS can be established. This team, led by Bengt Karlson (Sweden), will initially update the IOC-IPHAB-GEOHAB recommendations for real time HAB observation systems, and make recommendations regarding HAB-observations in the ChloroGIN (<http://chlorogin.org/contact.php>) pilot areas among other activities.

Discussion on progress in regional research focused on the success of the 2nd GEOHAB Asia meeting, organised by Ken Furuya (Japan) and hosted at the Vietnamese Institute of Oceanography in Nha Trang by Nguyen Ngoc Lam. At this meeting, and subsequently, a document was developed on the likely effects of large scale

nitrogen additions in the sea, following a proposal to dump large amounts of urea in the Sulu Sea (Glibert et al. 2008 Marine Pollution Bulletin, doi 10.1016/j.marpolbul.2008.03.010). The SSC were delighted to learn that the government of the Phillipines had refused permission for this activity. GEOHAB has issued a statement on this activity which is posted on the website (www.geohab.info).

Robert Magnien and Quay Dortsch (NOAA) were among agency representatives who were invited to the SSC meeting for a briefing on US HAB programs. An evening outreach session was also organised by Pat Glibert (Horn Point, MD, USA) at the Chesapeake Bay Program Office in Annapolis Maryland where four presentations on GEOHAB were made followed by a small reception.

Further points under discussion at included arrangements for a workshop on modelling the biophysical interactions relevant to the promotion of HAB events is being organised by the GEOHAB SSC and will be held in summer 2009. The formal announcement for this will be in June 2008.

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The GEOHAB SSC in front of the Annapolis Synthesis Center of the University of Maryland, Center for Environmental Science, Annapolis, USA.

ISSHA President's Corner

Exciting things are happening. We are less than 6 months away from the 13th International Conference on Harmful Algae 3-7 November 2008 in Hong Kong and there are a number of very busy local and international organizing committee members making preparations.

ISSHA Auction

Plans for the ISSHA Auction are underway. We are soliciting items for the ISSHA Auction. If you can recommend a vendor you would like ISSHA to contact for a contribution, please send me contact information. Also, be thinking about you might bring to the Auction. I know that Allan Cembella will have the now famous Florida Red Tide - Mote Sunglasses Case back to see if he can recover his investment. Books, reprints and prints are always popular items and they travel well. See you at the ISSHA Auction in November!

Awards

Just a reminder that by 1 June 2008 Lifetime Achievement Award and Young Scientist Award Nominations should be sent by e-mail to Marina Montresor (mmontr@szn.it), Chair of the Committee on Achievement Awards.

Officer and Council Nominations

These nomination are due on 3 July 2008. Dr. Karen Steidinger has agreed to help vet the nominations. Please submit candidates to her at Karen.Steidinger@MyFWC.com. You may also send nomination to the ISSHA Secretary, Dr. Tracy Villareal at T.Villareal@mail.utexas.edu.

*Pat Tester, ISSHA President,
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Future events

Events in Food Safety, Toxins and Related Areas, AOAC Toxins Task Force 2008:

1) Dallas, Texas Sept 21-24, AOAC Int. Annual Meeting:

The 2008 AOAC International Annual Meeting brings together prominent senior-level analytical chemists and microbiologists to present research and share ideas with common scientific interests in the areas of biodefense, agricultural, food, drug, and environmental sciences.

There will be an open session of the Marine and Freshwater Toxins Task Force Meeting.

More info at: www.aoac.org/meetings1/122nd_annual_mtg

2) Seattle and Tacoma, Washington, June 15-20.

Rapid Test Kit Workshop, Seattle, WA Shoreline Laboratory and Hotel Nexus, June 15-17.

ELISA and Dipstick Tests for Natural Toxins, Vet. Residues & Allergens - Including lectures and extensive hands-on experience, kits from several vendors, this session in association with Pacific NW AOAC Annual Meeting. Spots are available on limited basis, so if you are interested in the workshop please contact:

James_Hungerford@hotmail.com

3) Pacific NW AOAC Annual Meeting, Tacoma, WA, June 18 - 19.

There will be a specific seminar on Marine and Freshwater Toxins.

More info at: www.aoacpacnw.com



OCTOBER 2008

Ciguatera and related biotoxins

Nouméa, New Caledonia, 21-31 October 2008.

A workshop on the latest developments in ciguatera research, covering environmental influences, causative organisms and socio-economic and medical aspects.

Deadlines:

- Submission of abstract: 1st June.
- Early registration: 1st June.
- Late registration: 15th September.
- Submission of full papers: 31st December.

For additional information: nwww.ird.nc/ciguatera

NOVEMBER 2008

The 13th International Conference on Harmful Algae 2008

Hong Kong - China, 3-7 November 2008.

The International Society for the Study of Harmful Algae (ISSHA) is pleased to announce that the 13th International Conference on Harmful Algae will take place in Hong Kong on 3-7 November, 2008. Supporting organizations include School of Science & Technology, the Open University of Hong Kong and the Association of

Harmful Algal Bloom of South China Sea (AoHABSCS).

The conference will address all issues related to the causes and effects of marine and freshwater harmful microalgae and, to serve as a forum for the exchange of new research results and relevant ideas among researchers, industry, government and all other interested groups.

Important dates:

- Abstract submission deadline: 1st June.
- Early registration: before 1st June.
- Late registration: 1st June - 15th Sept.
- Deadline for Submission of Full Papers: 31st December.

More info at: www.hab2008.hk

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