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Phytoplankton composition and abundance assessment in the Nador lagoon (Mediterranean coast of Morocco)

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We evaluated phytoplankton abundance, composition and trophic state of the Nador lagoon (Morocco) on the basis of data taken in the period November 2007 to August 2008. Sampling was performed at 11 stations (bottle samples at 0.5 m depth and horizontal plankton net tows with mesh size of 20 mm). Among seven identified phytoplankton classes, diatoms and dinoflagellates dominated with 133 and 169 species, respectively. Frequent phytoplankton blooms were contributed by one to three species in the lagoon. Abundance and seasonality of phytoplankton characterized the Nador lagoon as a highly eutrophicated environment.

Keywords – Mediterranean Sea, Nador lagoon, Phytoplankton, diversity, eutrophication

Environmental influences on the qualitative and quantitative structure of phytoplankton in Moulay Bousselham lagoon (Morocco) and Ghar El Melh lagoon (Tunisia)

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Seasonal dynamics of phytoplankton communities in two North African coastal lagoons: Moulay Bousselham lagoon (Morocco) and Ghar El Melh (Tunisia) was
investigated. The objective is to evaluate hydrological and other influences on the structure, composition and space-time development of these communities in each site. Phytoplankton in Merja Zerga showed a quasi-permanent predominance of marine diatoms in the open sea station and in the marine inlet channel. Dinoflagellates were abundant in summer and early autumn in the marine inlet and extended into the central lagoon station. In Ghar El Melh, marine species (especially diatoms and dinoflagellates) dominated despite occasional winter inflows of freshwater.

A total of 164 phytoplankton taxa were identified at the two lagoons. These included 63 Dinophyceae, 84 Bacillariophyceae, 8 Cyanophyceae, 7 Chlorophyceae and 2 Silicoflagellate taxa. Diatoms were dominant in terms of the number of species and abundance in the two lagoons. The highest number of species occurred in spring and summer in both lagoons, but in summer and autumn the number of Dinophyceae species usually exceeded that of the diatoms. The lowest number of phytoplankton species in Moulay Bousselham occurred in November and December. In terms of phytoplankton species succession, the diatoms were most abundant during April–May (Pseudonitzschia pseudodelicatissima, Nitzschia spp., Rhizosolenia sp.) as was the dinophyceaean Ceratium. In early April, species of Chaetoceros and Navicula were dominant in terms of both abundance and the number of the species. In summer the diatoms Licmophora spp, Navicula spp, Nitzchia spp. And Pleurosigma elongatum, together with the dinophyceans Ceratium spp and Protoperidinium spp., were most common. Nitzchia was of particular significance with 31 species identified at all the sites and Navicula with 15 species. In summer (June–August), Ceratium and Protoperidinium (Dinophyceae) were usually the dominant species. The genus Protoperidinium attained the highest diversity (11 species) at Ghar El Melh in November–January when Dinoflagellate cell numbers were at their lowest (7.9 $10^4$ cells m$^{-3}$).

Only in September did dinoflagellate cell numbers exceed those of diatoms at the open sea sampling station. This was due to a predominance of Peridinium spp. and Scrippsiaella trochoidea. In both lagoons, Dinoflagellate cell numbers reached 12,9 $10^6$ cells m$^{-3}$ at this time, but in November they declined (to c. 25.9 $10^3$ cells m$^{-3}$). The highest cell concentration in Moulay Bousselham was 7-9 $10^6$ cells m$^{-3}$ in June whereas for Ghar El Melh the highest concentration (9.9 $10^7$ cells m$^{-3}$) was in July. In Ghar ElMelh, dinoflagellates were the most abundant, especially Alexandrium
foedum, Ceratium furca, Coolia monotis, Peridinium quinquecorne, Prorocentrum compressum and Prorocentrum concavum. Some of these species are toxic. In June, the diatoms were mainly dominated by benthic species (Licmophora gracilis and Navicula cocconeiformis). The central lagoon areas were more productive during spring and summer, but Merja Zerga was more influenced by brackish species than Ghar El Melh lagoon, which was characterized by marine forms.

Toxic species of phytoplankton were recorded in all the two lagoons and, following the UNESCO classification, seven groups of harmful algae were identified. These occurred mainly during summer and autumn. Dinoflagellates and, to a lesser extent cyanophyceans, comprised the most harmful groups and particularly noteworthy are the dinoflagellate Gonyaulax spinifera in Ghar El Melh, Dinophysis species in both Ghar El Melh and Moulay Bousselham.

**Keywords:** Moulay Bousselham lagoon, Ghar El Melh lagoon, Phytoplankton, Structure, Harmful algae

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**Pseudo-nitzschia species in a coastal Mediterranean lagoon: Bloom characteristics, species identification and toxicity**

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The bloom dynamics of *Pseudo-nitzschia* species in the lagoon of Bizerte (North of Tunisia) were investigated during 3 years. Monthly sampling was carried out for 3 years (2004/2005, 2006/2007, 2008/2009) at several stations characterised by different ecological features. The causative species of blooms were isolated and maintained in batch culture. The specific identification of these species were performed by scanning electron microscopy and based on partial LSU rDNA. The toxicity of identified species was tested by HPLC analyse of domoic acid (DA) and Elisa method.

Throughout the years, species of *Pseudo-nitzschia* accounted for a large fraction (50-70%) of total microalgae and thus appeared as a regular component of the
phytoplankton. During the first year, the abundance picked (25 $10^4$ cells l$^{-1}$) in July 2004. The causative species of this bloom was identified as *P. calliantha*. During the second year, two blooms were detected on March 2006 (1.5 $10^4$ – 2.5 $10^6$ cells l$^{-1}$) and October 2006 (3.5 – 5 $10^6$ cells l$^{-1}$), with highest abundance at stations located in shellfish farming sectors. Diatoms responsible for these blooms were *P. delicatissima* and *P. brasiliana*. In the third year, a very pronounced bloom (3 $10^6$ cells l$^{-1}$) was observed in August 2008 and was probably triggered by *Nitzschia subtilis*.

The ACC analyses of environmental factors revealed that bloom of *P. calliantha* occurred in warmer Si-rich water characterized by high salinity, whereas *P. delicatissima*, *P. brasiliana* and *N. subtilis*. bloomed under high P or N-nutrient concentrations.

The amnesic toxin was detected in some strains of the four identified species (9.8 – 149.1 ng DA ml$^{-1}$ culture), with the highest level in the culture of *P. calliantha* isolated from the 2004 July bloom. Moreover, the DA was measured for the first time in seawater during the 2006 October bloom of *P. brasiliana*. The study showed that nutrient-rich waters of the lagoon are suitable for the growth and proliferation of several toxic species of *Pseudo-nitzschia/Nitzschia*. This may constitute a threat to further aquaculture activity development in the Lagoon. Therefore, a rigorous monitoring for ASP toxins, as well as an accurate examination of *Pseudo-nitzschia* dynamics with related environmental factors are required.

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**Response of the genus *Pseudo-nitzschia* H. Pergallo to increased water temperature in a coastal Mediterranean lagoon (Bizerte Lagoon, North Tunisia)**

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Occurrence and abundance of the potentially toxic genus *Pseudo-nitzschia* were studied in Bizerte lagoon during two annual cycles (from March 2004 to March 2005 and from March 2006 to February 2007) that significantly differed in water temperature. Sampling was conducted at three stations inside the lagoon and at one
coastal station located in the Bizerte Bay (Mediterranean Sea). The results indicated that the genus *Pseudo-nitzschia* was significantly more frequent and more abundant during the second study period characterized by the highest water temperature. During 2006-2007 sampling period, the genus was present at higher frequency (in > 90% of the samples) and was more abundant (1.9 x 10^4 – 4.9 x 10^6 cells l^-1) when compared with the values obtained during the 2004–2005 survey (in 70% of the samples; 3.01 x 10^3 - 2.31 x 10^5 cells l^-1). Accordingly, a significant correlation was recorded between water temperature and *Pseudo-nitzschia* abundance (r_x= 0.552; p < 0.01). The genus *Pseudo-nitzshia* was also more diverse during the second study period, when three representative groups were observed (*N. seriata*, *N. delicatissima* and *P. americana* complex). Conversely, throughout the 2004-2005 period, only two taxonomic groups (*N. seriata* and *N. delicatissima*) were present. These results suggest that the increase of water temperature may affect the dynamics of the potentially toxic *Pseudo-nitzschia* and may promote their proliferation in coastal Mediterranean lagoons.

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**Occurrence and environmental controls of potentially harmful dinoflagellates blooms in SW Mediterranean lagoon (Bizerte Lagoon, Tunisia)**

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Abundance of the potentially harmful (PH) dinoflagellates was examined in relation with environmental factors in the Bizerte Lagoon from November 2007 to February 2009. The investigation was carried out at six sampling stations characterised by different ecological features. Ten potentially harmful taxa were observed during the sampling period; *Alexandrium tamarense*, *Ceratium lineatum*, *Cochlodinium polykrikoides*, *Dinophysis* spp., *Gonyaulax* spp., *Gymnodinium mikimotoi*, *Gymnodinium sanguineum*, *Protoperidinium* sp. and *Prorocentrum* spp. Blooms of PH dinoflagellates were highly variable in intensity (4 x 10^3 - 7 x 10^5 cells l^-1), time (November 2007, January, February, May, August, September and October 2008) and space.
The most abundant (2 \(10^4\) – \(4 \times 10^5\) cells l\(^{-1}\)) and frequent PH dinoflagellates, as *Prorocentrum* spp., *Dinophysis* spp. and *C. lineatum*, exhibited high densities picks, particularly in shellfish stations during November 2007 and January 2008. The other dinoflagellates (*Protoperidinium* sp., *Gonyaulax* spp., *G. mikimotoi*, *G. sanguineum* and *C. polykrikoides*) were also present during several sampling mouths, but at lower concentrations (\(4 \times 10^3\) – \(4 \times 10^4\) cells l\(^{-1}\)). *A. catenella* was present at very low levels, except in November 2007 when it performed a sudden pronounced bloom (\(2.5 \times 10^5\) – \(7 \times 10^5\) cells l\(^{-1}\)) at stations located in conchyliculture sectors. ACC analyses of environmental factors revealed that all blooms of PH dinoflagellates were related to low temperature, turbidity and ammonium concentrations. Some dinoflagellate taxa (*A. catenella* and *C. polykrikoides*) were associated with high salinity, Chl *a* and phosphate levels, whereas others (*C. lineatum*, *Prorocentrum* spp., *Protoperidinium* sp., *Gonyaulax* spp., *G. mikimotoi*, *G. sanguineum* and *Dinophysis* spp.) were found in samples with high availability of nitrate and nitrite. The widespread distribution of the PH dinoflagellates in eutrophic waters of the Bizerte Lagoon and the frequent presence of blooms, particularly in shellfish areas, suggest a potential thread of the harmful species in this coastal Mediterranean lagoon.

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**Potential threat of the benthic dinoflagellates Gambierdiscus and Ostreopsis to North Africa: a review of their ecology and toxicology**

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Ciguatera fish poisoning (CFP) is a widespread food-borne syndrome caused by the consumption of contaminated fish. The incidence and worldwide distribution of CFP appear to be increasing paralleling a worldwide increase in toxic or harmful algal bloom (HAB) events. The primary toxin involved with CFP is ciguatoxin, produced by the dinoflagellate of the genus *Gambierdiscus*. However, there are reports of multiple toxins other than ciguatoxin from both moray eel viscera and Spanish mackerel. These additional toxins, along with the complex symptomology of CFP, suggest the involvement of more than one organism and/or toxin. Subsequent ecological surveys have consistently observed a number of other toxic dinoflagellates,
including the genus *Ostreopsis, Prorocentrum, Coolia,* and *Amphidinium,* in higher concentrations than *Gambierdiscus.* These toxic dinoflagellates share similar habitats whose complex interactions make the CFP problem more complex. Together this collection of epiphytic dinoflagellates can be referred to as the “epiphytic toxic phycosphere”. Of the known benthic, toxigenic microalgae, species from the genera, *Gambierdiscus* and *Ostreopsis,* present the biggest threats to human and environmental health. This review will discuss the threat of benthic dinflagellates to North African ecosystems including a review of toxins and toxin analysis.

**POTENTIALLY HARMFUL EPiphytic MICROALGAE IN ALEXANDRIA WATERS**

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The purpose of this work is to carry out a survey of the potentially harmful microalgae epiphytic on macroalgae in Alexandria waters. The survey focused on the occurrence, composition and abundance of the microalgae in relation to the main ambient physico-chemical conditions, on one hand, and on the host preference on the other hand.


Benthic harmful microalgae have caused fish kills in Alexandria in more than one instance. In the year 2004, mass mortality of the fish Siganus spp. happened in the Eastern harbour (Ismael and Halim, 2007). Siganus spp. are known to be voracious feeders on the algae Ulva spp., the preferred host of certain harmful microalgae. On another instance, fish kills happened more than once in the public aquaria of the National Institute of Oceanography and Fisheries. The aquaria are known to be fed by water pumped in from the bottom of the Eastern harbour and therefore containing benthic microalgae. The latter case is still under investigation.
Macroalgae were collected from three random quadrates of 0.1 m$^2$ from the hard substratum and all biota inside the quadrate completely removed by scraping the hard substrates.

The survey yielded 175 microalgal species both harmful and harmless – epiphytic on macroalgae. They consist of 90% diatoms, 6% dinoflagellates, 2.3% cyanobacteria and 1% euglenophytes. This inventory comprises 17 potentially harmful species.

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**New assemblage of dinoflagellate cyst in surface sediment of Moroccan marine ecosystem**

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The distribution and abundance of cysts assemblages were investigated in surface sediment from Walidia lagoon on Moroccan atlantic coast since 2006 to 2011.

From 2006 to 2008, the surface sediment showed the presence of the same assemblage, composed of 3 cyst: *Lingulodinium machaerophorum* (cyst of *Lingulodinium polyhedrum*, indicator of lagoon ecosystem), *Spiniferites ramosus* and *Operculodinium centrocarpum* (cyst of *Protoceratium reticulatum*, indicator of marine effect), with a dominance of *Lingulodinium machaerophorum*. Since 2009, the palynofacies change, a new assemblage of cyst was observed, *Lingulodinium machaerophorum*, *Protoperidinium sp*, and *Polykrikos schwartzii*. This assemblage is correlated with a high activity of upwelling system (Targarona *et al*, 1999). The new cyst association was noted at the entire samples sites of Oualidia lagoon. The changement in surface sediment composition is correlated with an modification of hydrographic conditions at this lagoon.

This study suggest that the marine sediments in atlantic coast should be monitored for the presence of Dinoflagellate cyst to give ample warning of the presence and abundance of toxic species, and to get all the new cyst assemblages, indicator of a new morphology and oceanographic conditions in marine ecosystem.
Monitoring of toxic Dinoflagellates species along the Atlantic coastline El Jadida-safi (Morocco)

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Under the surveillance program of toxic phytoplankton conducted by Department of safety and quality of the marine environment of the National Institute for Fisheries Research along the Atlantic coastline Eljadida-Safi, analysis of samples of seawater collected shellfish production areas during the period 2009-2010, revealed the presence of 8 species of the genus Dinophysis (Dinophysis acuminata, D.acuta, D. caudata, D.fortii, D.rotandata, D.sacculus, and D. hastata and D. diegensis), known for their ability to produce toxin (DSP: Diarrheic Shellfish Poison). These species were found in 4 areas of shellfish production (Jemaa Ouled Ghanem, Dar Hamra, Oualidia Lagoon and Cap Beddouza). The presence of these toxic species presents a potential danger to consumers of the shellfish.

The cell density of these species exceeded 2.10³ cells / liter, this proliferation was accompanied by contamination of mussels in areas Jamaa ouled Ghanem, Dar Hamra and Cap Beddouza what caused the prohibition of exploitation of these shellfish products, this in order to protect consumers against possible poisoning.
Évaluation de la biodiversité phytoplanctonique dans la lagune de Oualidia (Maroc)

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Afin de caractériser les espèces phytoplanctoniques potentiellement toxiques et les éventuelles efflorescences nuisibles, une étude a été entreprise de février à mai 2010 au niveau de la lagune de Oualidia abritant les stocks conchylicoles les plus productifs à l’échelle du Maroc (Huître et palourde) et présentant une biodiversité marine très riche qu’il est nécessaire de préserver contre les différentes formes de contaminations, y compris celle par les phycotoxines.

La richesse spécifique des quatre points de prélèvement (Passe principale, Parc 7, Parc 1 et l’Ancien Parc Marost) est 105 taxa répartis en 72 espèces de Diatomées, contre 27 espèces de Dinoflagellés et 3 espèces aussi bien chez les Silicoflagellés et que chez les Phytoflagellés.

L’étude spatio-temporelle de la composition du phytoplancton a révélé que ce milieu paralique est essentiellement caractérisé par les Diatomées et les Dinoflagellés, avec une nette dominance aussi bien qualitative que quantitative des Diatomées avec une abondance relative de 91% due, principalement, aux conditions environnementales particulières qui y règnent.

Au cours de la période d’étude, il n’a pas été noté de proliférations massives du phytoplancton toxique ou nuisible. Cependant, certains genres renfermant des espèces toxiques ou nuisibles ont été rencontrés, mais à de faibles concentrations situées au dessous du seuil de toxicité.

Il s’agit d’Alexandrium, Dinophysis, Gymnodinium, Lingulodinium, Pseudonitzschia, Prorocentrum, Ceratium, Chaetoceros et Protoperidinium.

**Mots clés**: Lagune de Oualidia, Phytoplancton potentiellement toxique, Efflorescences algales nuisibles, Diatomées, Dinoflagellés, Paramètres environnementaux.
HAB blooms in Moroccan Atlantic Agadir coasts during the monitoring program (2003-2011)

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During the HAB monitoring program, carried out from January 2003 to August 2011, along the Agadir coastal area, the increase of occurrence and diversity of HAB species blooms has been observed. Species identified in this blooms belong to three different groups: Diatoms (\textit{Pseudonitzschia}), Ciliates (\textit{Mesodinium rubrum}) and Dinoflagellates (\textit{Dinophysis}, \textit{Lingulodinium polyedrum}, \textit{Prorocentrum micans}, \textit{P. scutellum}, \textit{Prorocentrum gracile}, \textit{P. triestinum}, \textit{Alexandrium minutum}, \textit{Gymnodinium catenatum}, \textit{Karenia mikimotoi} and \textit{Ostreopsis siamensis}). The most of these blooms occurs from June to September. \textit{Dinophysis}, \textit{Alexandrium} and \textit{Gymnodinium} were present at low cell concentration in surface waters (maximum $10^3$ cell.l\textsuperscript{-1}). Recently, the Ostreopsis blooms became frequent and increased in concentration. This concentration varied from $12 \times 10^3$ cell.l\textsuperscript{-1} in 2008 to $10^5$ cells.l\textsuperscript{-1} observed in 2009, 2010 and 2011 at summer- autumn seasons. Some of these blooms were associated with the PSP, LSP and ASP toxicity in mussels and others were accompanied by the mortality of fish and sea urchins in contaminated areas. Some oceanographic parameters (winds, upwelling, sea surface temperature and surface circulation patterns) were used to explain the Ostreopsis bloom’s dynamics.
Dinophysis blooms in Tunisian coastal waters
(Gulf of Tunis, SW Mediterranean Sea)

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The first documented Dinophysis bloom from Tunisian coastal waters was recorded in June 2009 during the monitoring program in the Gulf of Tunis. A species with morphological features similar to D. acuminata Claparede and Lachmann dominated this bloom. Maximum D. cf. acuminata abundance (130 x 10³ cells L⁻¹) appeared in early June 2009. In 2008, high numbers of D. cf. acuminata (6 x 10³ cells L⁻¹) were recorded in September under conditions of high temperature and salinity, while maximum numbers (38 x 10³ cells L⁻¹) of the same species were found in December under low temperature levels (11.5 - 12.5°C). All Dinophysis blooms persisted for no more than 4 months. Cluster analysis of measured specimens revealed the presence of three forms, F1, F2 and F3, within the D.acuminata complex, F2 and F3 were fairly close and similar to the original description of D.acuminata, whereas F1 was quite distinct with characteristics similar to D.sacculus Stein. These different forms were observed throughout the whole period at different stages of the blooms, but each bloom was dominated by certain of them. Other observations include couplets of large and small-sized cells, and also a larger robust form. The D. cf. acuminata population always presented a stratified vertical distribution with vertical peaks positioned in or just above the pycnocline. Among the physico-chemical parameters, water temperature appears to be the most important factor influencing the distribution of Dinophysis abundance.

Keywords: D. acuminata complex, bloom, forms, water temperature.
Harmful Algal blooms in Tunisian coasts: Genesis, socio economic impact and assessment issues

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Harmful Algal Blooms (HABs) represent a significant and expanding threat to human health and fisheries resources throughout the world. These phenomena have increased steadily in both species complexity and geographical extent over the last several decades. In turn, the range of harmful effects and the magnitude of economic costs have also widened.

During the last two decades, HABs events have been observed in more locations than ever before throughout the Tunisian coasts. Some toxic microalgal species are involved and few toxins have been uncovered, but against many fisheries resources have been affected. This increase in HABs events seems to be a real phenomenon in our coasts. The role of monitoring is important in the increase of detection of these phenomena but we believe that anthropogenic nutrient loadings, marine traffic (ballast water) and development practices of aquaculture and fishing can be responsible of the spread of HABs.

Unfortunately the problem isn’t only ecological and harmful algal blooms have also significant economic impacts. Furthermore, it is important to begin to understand the scale of the economic costs of such natural hazards and to assess the balance between the costs of shellfish production, HABs monitoring and management of the problem. This study aims at providing elements for the analysis the sources of increase of HABs in our coasts and their impacts first for the ecological of view but essentially for socio-economic point. We will try to compile disparate estimates of the economic effects of HABs during 1995–2010. We will compare looses of fisheries, recreation and tourism, and the coasts of classic monitoring and management. Some assessments issues including new monitoring methods and exploitation operations will be also proposed.
Bloom forming microalgae in Algiers coastal waters (SW Mediterranean sea).

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A harmful microalgal study conducted in two harbours, two enclosed beaches and a shellfish area of Algiers Wilaya during spring and summer 2008, 14 potentially harmful species have been collected. Among forming bloom species: the dinoflagellates *Prorocentrum triestinum*, *P micans*, *Scrippsiella spp*, *Protoperidinium quinquecorne*; the diatoms *Leptocylindrus danicus/minimus*, *Chaetoceros spp*. and the raphidophytes *Eutreptiella sp*. Among potentially toxic species: *Pseudo-Nitzschia spp.*, *Dinophysis rotundata* and *Ostreopsis spp.*

Les dinoflagellés dans la baie de Bousmail (Algérie)

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Nous présentons dans ce travail, un inventaire des dinoflagellés de la baie de Bousmail (Algérie) durant l'année 2009. Différents paramètres physico-chimique ont été réalisés.

Les analyses qualitative et quantitative des peuplements phytoplanctoniques révèlent la présence des principaux genres de dinoflagellés:*Ceratium*; *Protoperidinium*, *Dinophysis*, *Noctiluca*, *Scrippsiella*.

*Mots clés*: Dinoflagellés; Algérie; Méditerranée
PSP and DSP toxins in four species of shellfish from south Atlantic Moroccan coasts

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Paralytic (PSP) and some lipophilic shellfish toxins (LSTs) can cause human illness due to eating shellfish containing some algal toxins. A mouse bioassay (MBA) is used in Morocco to detect phycotoxins in regulatory monitoring of shellfish established by INRH, the methods used are AOAC 2000 for PSP toxins and Yasumoto 1984 for LSTs. This test has proven his efficiency in the surveillance of paralytic toxins for several years. But in case of LSTs detection, the MBA is imprecise giving only a positive or negative result and is prone to interference from other compounds. The search for alternative methods to the mouse bioassay for detection of LSTs was necessary before 31 December 2014 according to commission regulation (EU) No 15/2011 of 10 January 2011.

The present work analyzes PSP and LSTs in four different species of shellfish (mussels, oysters, cockles, and razor clams) sampled from 2003 to 2007 in 2 areas at Agadir, one at Laâyoune and eight at Dakhla using MBA, samples taken during toxic events were analyzed through liquid chromatography-mass spectrometry (LC-MS/MS) for LSTs and by HPLC-FD for PSP toxis.

The LSTs studied were OA, DTX-1, YTX, 45-OH- YTX, Homo-YTX, 45-OH-Homo-YTX, DA, GYM, PXT1, PTX-2, PTX-2sa by Multiresidue Method for Determination of Algal Toxins in Shellfish (Holland 2003, Goto 2001) and PSP toxins studied were STX, GTX (1,2,3 and 4), Neo-STX, dcGTX (2 and 3), B (1 and 2) toxins and C toxins (1,2,3 and 4) by Post-column-periodate oxidation (Thielert et al. (1991) modified by Hummert et al. (1997) and Yu et al.(1998.).

The results showed a summery apparition of PSP toxins at Agadir and Laâyoune’s shellfish and autumnal at Dakhla’s shellfish, the majors toxins detected were carbamats toxins, mainly GTX1, 2, 3, 4 and a minority of STX. The LSPs toxins
detected were OA, DTX-1 and its associated esters (DSP toxins); no seasonality of these DSP toxins was distinguished. The results obtained in a comparative study of PSP and DSP toxic shellfish using the two methods showed that some MBA negatives samples were positives by HPLC-FD for PSP and LC-Ms for DSP. The comparison between the STX concentrations obtained by these two methods revealed a very highly significant difference was found between the results of MBA-PSP and HPLC/FD-PSP with t = 5.12 (P<0.001) and a negative correlation between OA concentrations and death times of MBA (r=-0.78). The comparison between the toxicity of different shellfish revealed that the benthic species especially Solen marginatus and cockles were more toxic than mussels and oyster. The PSP shellfish’s contaminations were mainly associated with the detection in seawater of Alexandrium minutum and the DSP contamination with a various species of Dinophysis.

Etude des mécanismes de contamination des mollusques bivalves par des neurotoxines à action rapide (FAT) & développement de procédés de détoxification

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This study aimed to determine the optimal conditions of growth and toxin production in Karenia selliformis and Alexandrium ostenfeldii in order to produce cells at a high concentration with known toxicity to perform further contamination and detoxification trial on edible shellfish. Moreover, this work aims to study the impact of non-toxic microalgae on the detoxification kinetics of clam Ruditapes decussatus contaminated by gymnodimines and oyster Crassostrea gigas contaminated by spirolides. The final goal of this study is to determine the physiological impact of toxic microalgae in the shellfish tissues.
Initially, results revealed that the growth performance of *K. selliformis* (growth rate and maximum concentration) is obtained at a temperature of 22°C and at a salinity of 36 psu when using the f/2 medium, while those of *A. ostenfeldii* are obtained at a temperature of 16°C and at a salinity of 35 psu using the L1 medium. Gymnodimine concentration increases with the age of the culture of *K. selliformis* while spirolide concentration decreases during cell growth of *A. ostenfeldii* especially during the culture in a photobioreactor of 100 l. Secondly, the experimental studies focused on the interaction between toxic microalgae with molluscs bivalves. The results revealed i) a major accumulation of GYMs and SPXs in digestive gland of clams and oysters ii) a rapid detoxification of contaminated shellfish when adding food.

Finally, this study addressed an important issue on the impact of toxic algae on shellfish. The exposure to *A. ostenfeldii* showed i) a decrease in the digestive gland tubule thickness and the percentage of active digestive tubules ii) an inflammatory response consisting of hemocyte infiltration and diapedesis into the intestinal tract of the oysters. Concerning the natural and experimental contamination of clams, histological analysis did not reveal any alteration of the digestive gland as demonstrated in oysters contaminated by *A. ostenfeldii*.

**Keywords:** *Karenia selliformis*, gymnodimine, *Ruditapes decussatus*, *Alexandrium ostenfeldii*, spirolide, *Crassostrea gigas*, culture, detoxification, histology.
The potent neurotoxin domoic acid (DA), produced naturally by several species of the diatom genus *Pseudo-nitzschia*, is the major causative agent of amnesic shellfish poisoning. Occurrence of harmful algal blooms (HABs) has become a major concern for several threatened regions in Morocco, Tunisia and Egypt. To address the adverse effects of harmful diatom blooms, contributions are being made by our agencies to support an algal toxins monitoring programme. Regulatory levels have been established and several technologies were adopted. Liquid chromatography –tandem mass spectrometry is the standard method usually used for confirmation of domoic acid contamination of seawater and shellfish matrices. This method is recognised as sensitive and serves as an important tool for investigating DA residues and its congeners in marine food webs. This approach is preferred since it avoids the use of live animals; however, this technology presents some limitations for unknown toxins and those for which standards are unavailable. To enhance the sustainable development and management of the marine coastal environment through the application of isotope techniques, a rapid and sensitive receptor binding assay technology has been developed for DA detection. This method, known as a competitive binding assay, uses the recombinant GluR6 glutamate receptor and microplate liquid scintillation counting for domoic acid detection in shellfish and algal extracts. This *in vitro* technology is rapid, reliable, cost effective and particularly suitable for determination of the integrated toxic potency of a sample. It’s also an additional option for countries with legislation restricting the use of live animal bioassays for regulatory applications.
**Keywords**: domoic acid; harmful algal blooms; liquid chromatography–tandem mass spectrometry; receptor binding assay; shellfish.

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**Gymnodinium catenatum** Grahams blooms in Moroccan Western Mediterranean coasts & depuration of shellfish species contaminated by Paralytic Shellfish Poisoning (PSP) toxins

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The study of *Gymnodinium catenatum* was conducted across two sampling stations; M'diq bay and Oued Laou estuary during the period from July 2007 to May 2009. The results showed that the *Gymnodinium catenatum* blooms occurred during the autumn period and early winter. During January 2008, the presence of *Gymnodinium catenatum* in high concentrations resulted in contamination of shellfish species cockles and sweet clam by Paralytic Shellfish Poisoning (PSP) toxins. In the Oued Laou estuary, the levels of these toxins in shellfish went beyond the normative threshold for consumption of shellfish, (80 µg SXTeq. (100g)⁻¹ of meat) and reached (710 ± 82.07) and (198 ± 6.56) µg SXTeq. (100g)⁻¹ of meat in cockles and sweet clam respectively. In M'diq bay, levels of PSP toxins in the meat of these two shellfish were relatively less important: 256.57 ± 12.22 µg SXTeq. (100g)⁻¹ and 80.66 ± 8.14 µg SXTeq. (100g)⁻¹ of meat in cockles and sweet clam respectively. The depuration of cockles and sweet clam was conducted in laboratory conditions. The results showed partial and progressive elimination of PSP toxins in two shellfish species in time. In the cockle, the elimination of PSP toxins appears to be slower compared with the sweet clam.

**Keywords**: Morocco, Gymnodinium catenatum, PSP toxins, Acanthocardia tuberculata, Callista chione and depuration.
Variability of *Pseudo-nitzschia* Peragallo (Bacillariophyceae) and domoic acid in M'diq Bay, Morocco.

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A study of the *Pseudo-nitzschia* genus was conducted in the M'diq Bay (35°43', 425 N – 05°19', 841 W), during the year 2007. The evolution of the *Pseudo-nitzschia* was studied in relation with the local evolution of the domoic acid (DA) in two shellfish species; cockle (*Acanthocardia tuburculatum*) and sweet clam (*Challista chione*). The obtained results showed that the highest concentrations of *Pseudo-nitzschia* are more important during the period from March to October, with peaks occurring in spring and autumn. However, toxin analysis by High-performance liquid chromatography (HPLC) showed an increase of domoic acid accumulation in Shellfish sampled during spring and autumnal periods, coinciding with the periods of the maximal proliferations of *Pseudo-nitzschia*. The maximum toxin concentration was 4.9 µg DA g⁻¹ of whole tissue recorded in Sweet clam in spring period. Using scanning transmission electron microscopy, thirteen species were identified, among which nine are considered as producers of the domoic acid. These are *Pseudo-nitzschia multistriata*, *P. cuspidata*, *P. galaxiae*, *P. multiseries*, *P. delicatissima*, *P. pseudodelicatissima*, *P. pungens var. aveirensis*, *P. calliantha* and *P. fraudulenta*. The other four not toxic species are *P. subpacifica*, *P. dolorosa*, *P. sub-fraudulenta*, and *P. cf. caciantha*.

*Keywords*: Morocco, M'diq Bay, Shellfish, Domoic Acid, *Pseudo-nitzschia spp.*
Monitoring of Harmful Algal Toxins Using the Radioligand Receptor Binding Assay
An IAEA Technical Cooperation, UNESCO-IOC and NOAA Joint Project

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Harmful algal blooms (HABs) may produce potent toxins that are transferred through marine food webs, threatening wildlife diversity and, when accumulating in seafood, human health, international trade and sustainable coastal fisheries development. In the context of climate change, benthic HABs appear to be more frequent and widespread globally and in particular for countries ad joining the Mediterranean. In recent years, Ciguatera Fish Poisoning (CFP) has been reported for the first time in North Atlantic Ocean, near North Africa, in the Macaronesian islands (Madeira, Canary Islands…).

In partnership with NOAA-NOS and IOC-UNESCO, the IAEA Technical Cooperation (TC) department has been supporting technology transfer and capacity building to assist member states in managing HABs through toxigenic species monitoring, biotoxins analysis, and dating of sediment cores. Technologies involve the radioligand receptor binding assay (RBA) for toxins associated with Paralytic Shellfish Poisoning (PSP), currently undergoing an AOAC inter-laboratory collaborative trial, and with Ciguatera Fish Poisoning (CFP). Other method include radiometric sediment core dating combined with fossil cyst abundance to allow reconstruction of the prior history of blooms, such as those of \textit{Gymnodinium catenatum}, and their relationship to climate. Following a successful project in Southeast Asia, implementation of these technologies has been initiated in Tunisia through the African regional project and is currently underway in Latin America.
Detection and impact of the Atlantic flow on the phytoplankton richness in the Sicily Channel during summer cruise

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During the “SEASAME” project, a cruise has investigated eight stations in the Sicily channel along a radial Tunisia-Sicily in September 2008.
The summer season is marked by a strong “thermocline” at the mean depth of twenty meters. The following profiles of different abiotic parameters: temperature, salinity, dissolved oxygen and water density detected the presence of a vein of Atlantic current.

This Atlantic current is identified in the first hundred meters deep and characterized by a low temperature and salinity.

The qualitative and quantitative study of phytoplankton at these depths showed low species richness and a low density compared to that determined for the Mediterranean waters. During this study, we conclude that the Atlantic vein which flows into the Sicily channel do not enrich the Mediterranean waters with new phytoplankton species.

Keywords: Sicily Channel, Atlantic Current & phytoplankton.
Cultures of micro-algae from Moroccan Atlantic and Mediterranean coast
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HAB is in increasing in the world, can cause severe economic losses to aquaculture, and human health impacts.
At Moroccan coasts, potentially toxic species are present and always blooming and closing areas for harvesting for many weeks.
The are a national program to isolate and purify the Moroccan potentially toxic species, to obtain cultures of harmful species from local areas they infest.
The cultures will help to determine not only the systematic position but pigment content, genetic studies, the toxins production and the toxins identification.
Furthermore, it deals with screening, isolation, purification and identification of species having direct impact on shellfish products. To access to this resource one requires a best mastering and a deep knowledge of their culture conditions at the laboratory scale on artificial medium and production conditions as well. The results presented here are related to the isolation, purification and culture conditions of 4 marine toxic species of flagellate, diatoms and Dinoflagellate.

\textbf{Keywords}: Isolation- Purification- Microalgae – Identification
Preliminary study on epiphytic dinoflagellates at Algiers coastline
(Algiers Bay and Bou Ismail Bay)

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A potentially toxic epiphytic dinoflagellate assemblage on macroalgae was studied from March 2010 to October 2011 in a shallow protected rocky habitat in Algiers Bay and Bou Ismail Bay (West of Algiers bay, Algerian bassin). The assemblage was monitored on Rhodophyte macroalgae: Corallina elongata. The dominant dinoflagellates were Ostreopsis cf. Ovata, and the accompanying species were Prorocentrum lima, Coolia monotis and Amphidinium cf cartereae. The diatom Coscinodiscus sp. and Lichmophora sp. was an abundant component of the assemblage. The morphometric measurement obtained from specimens collected in Bou Ismail Bay (Algerian coast) are indicative to Ostreopsis cf. Ovata with DV : AP ratio close to 2.94. The dinoflagellate assemblage follows a clear seasonal pattern, achieving maximum cell concentration during spring (the maximum value was 61x10^3 cells/gr Fr w of macroalgae) and summer with significant relative changes in the species composition. Negative correlation was recorded between the cell concentration of Ostreopsis cf ovata and water temperature.
dinoflagellés qui dominent. A la station S3 en face de la STEP trois groupes principaux se partagent la dominance : les Prasinophycées avec l’espèce *Tetraselmis sp* (en été) qui donne la couleur verte à l’eau, les diatomées avec principalement l’espèce *Nitzschia longissima* (printemps) indicatrice de milieu confiné et les Euglenophycées avec l’espèce *Eutreptiella sp* (printemps). Le maximum de la richesse spécifique est observé à la station de la passe marine S6 et le minimum au niveau de la station S3 en face de la STEP.


*Mots clés* : phytoplancton, efflorescences algales, lagune, diversité.

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**Occurrence of potentially toxic cyanobacteria in the reservoir of Sidi El barrak (Nefza, North West of Tunisia)**

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A study of phytoplankton populations was carried out during the period of 4 seasons between 2008 and 2009 in the Sidi El Barrak reservoir (North West of Tunisia). This dam is one of the most important reservoirs used for drinking, irrigation and aquaculture in Tunisian inland waters. The aim of this work is to define seasonal succession patterns of phytoplankton in six different areas in this reservoir characterized by different environmental factors.
Analysis of phytoplankton showed a total of 167 species. There were 79 species of Chlorophyta, 61 species of Cyanophyta, 28 species of Bacillariophyta, 5 species of Chrysophyta, 4 species of Euglenophyta, 2 species of Dinophyta and 2 species of Xanthophyta within proportions of 99.58 % for Chlorophyta, 0.40 % of Cyanophyta and 0.01 % for Bacillariophyta. Among the cyanobacteria, *Aphanocapsa* and *Microcystis* were dominant, with the highest densities of the main species: *Aphanocapsa holsatica* and *Microcystis flos-aquae*. This later species was characterized by a periodic appearance in the pelagic zone and was sometimes the dominant species. The max. Concentrations about $13 \times 10^7$ cells l$^{-1}$ were recorded during autumn. Correlations between these phytoplankton community and nutrients loading were investigated.

**Keywords:** Phytoplankton- Cyanobacteria - *Microcystis* - nutrients - seasonal succession - Sidi El Barrak reservoir