Phytoplankton as biological quality element in the monitoring program of Cienfuegos Bay, Cuba.

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Presenting author first: Ángel R. Moreira González

Centro de Estudios Ambientales de Cienfuegos (CEAC)
Ministerio de Ciencia, Tecnología y Medio Ambiente
Apartado Postal 5, Ciudad Nuclear 59350, Cienfuegos, Cuba.
Email: angel@gestion.ceac.cu

Additional authors: Mabel Seisdedo Losa (mabel@gestion.ceac.cu), Alain Muñoz Caravaca (alain@gestion.ceac.cu), Carlos M. Alonso Hernández (carlos@ceac.cu), Augusto Comas González. (augusto@gestion.ceac.cu). Centro de Estudios Ambientales de Cienfuegos (CEAC)

ABSTRACT

Data on phytoplankton composition, abundance and biomass in Cienfuegos Bay in Cuba were collected during 2009 at sixteen representative stations within the bay. Phytoplankton samples were collected every three months including samples from subsurface water and benthic macroalgal turfs. Physico-chemical data were also collected and analysed with respect to the diversity and abundance of phytoplankton collected.

In general, based on the analysis of plankton chlorophyll *a* concentration, dissolved oxygen and phytoplankton composition, eutrophication and harmful algal blooms appeared be of minimal significance within Cienfuegos Bay. However high chlorophyll *a*, toxic blooms and large biomass of epiphytic macroalgae were encountered within urban wastewater discharge zones especially during the late dry period and early rainy period, when the flushing potential was low and when water temperature started increasing in the ecosystem. Blooms of toxic dinoflagellates observed in these zones were linked to a small fish kill episode.

Additionally moderate abundance of potentially ciguatotoxic and ichthyotoxic epibenthic microalgae were registered on blooms of filamentous macroalgae. Twenty-six species known to be toxic and/or harmful belonging to the genera *Gymnodinium*, *Alexandrium*, *Dinophysis*, *Prorocentrum*, *Ostreopsis*, *Heterocapsa*, *Amphidinium*, *Cochlodinium*, *Gonyaulax*, *Pseudo-nitzschia*, *Microcystis*, *Oscillatoria*, *Anabaena*, *Pseudoanabaena*, *Planktothrix* and *Lyngbya* were identified in the bay. During the rainy season, higher chlorophyll *a* concentration in spatial coverage was registered, corresponding with a medium level of eutrophic conditions. In this period, an increase of phytoplankton species richness was observed and potentially toxic species were replaced by beneficial diatoms.

Key words: Cienfuegos Bay, Cuba, eutrophication, phytoplankton, toxic microalgae

INTRODUCTION

In recent years, increasing supplies of nutrients to water bodies by human activities has been associated a predominance of algal blooms, a phenomenon known as “eutrophication”. Increased nutrient fluxes to estuarine and coastal areas result from agriculture, aquaculture, and discharges of industrial and domestic wastes. The resulting blooms lead to hypoxia, fish mortalities, toxicity of food organisms, and damage to benthic communities resulting from reduced light penetration and resulting alterations in trophic interactions throughout the ecosystem. These modifications have significant direct and indirect economic, social and environmental costs.

Cienfuegos Bay, one of the most valuable natural resources of the province of the same name, is distinguished by its extent and its beauty. The variety of developments in the surroundings and exploitation of the bay itself have impacted both the water and the coastal zone, and for these reasons it has been chosen for study within the framework of the Ramal Scientific-Technical Program, “Cuban Environmental Protection and Sustainable Development”.

Estimates indicate that in Cienfuegos Bay, direct nutrient discharges from urban areas contribute 15.1% to the total discharges from the rivers (Seisdedo and Arencibia, 2009). Recent observations on areas close to industrial and domestic discharges have shown deterioration of benthic communities and dead fish, coinciding with the occurrence of toxic and noxious algal blooms.

The objective of this paper was to describe the spatial and temporal variations of composition and abundance of phytoplankton communities, in relation to physico-chemical parameters, thus utilizing phytoplankton as indicators of water quality within Cienfuegos Bay ecosystem, with an emphasis on the detection of toxic and noxious microalgae.

METHODS AND MATERIALS

Study area

Cienfuegos Bay, situated in the southern central part of Cuba, is a semi-enclosed bay with a surface area of 90 km^2 and an average depth of 14 m. It is connected to the Caribbean Sea by a narrow channel 3 km long (Fig. 1). Several rivers flow towards the bay forming a complex estuary. The main rivers are Damují, Salado, Arimao and Caonao.

The bay is divided in two well-defined hydrographic basins, due to the presence of a submerged ridge 1m below the surface, just north of the connection channel. The bay and its coastal line represents the most important natural resource in the province, due to fishing (6%) and industrial (7%) activities, agriculture (2%), maritime transport (7%), natural parks (70%), urbanization and tourism (8%) (León, *et al*., 2000). The northern basin receives most of the anthropogenic impact from the outfall of Cienfuegos city (140 734 inhabitants), the industrial centre of the country, and the freshwater input of the Damuji and Salado rivers. The southern basin is subject to a smaller degree of anthropogenic pollution originating from the Caonao and Arimao rivers. Part of the southern basin is a natural park, an important habitat for protected migratory birds and marine species.

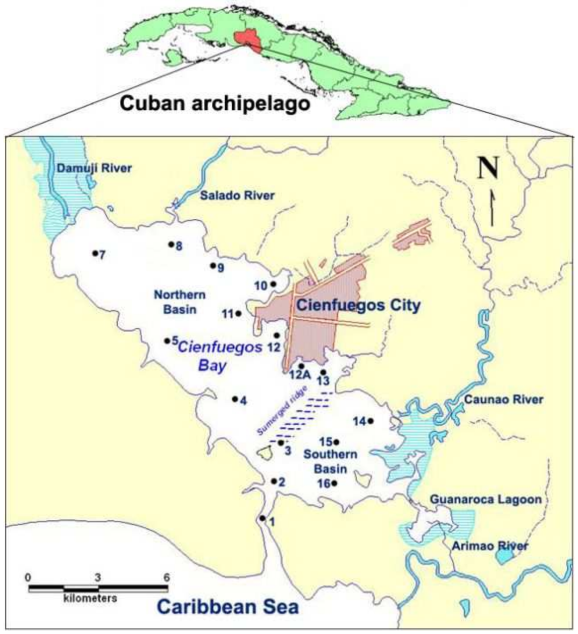

Fig. 1. Sampling stations in Cienfuegos Bay.

Sampling design and processing of samples

For phytoplankton analyses, subsurface water samples were collected three times monthly (Apr. 2009 – Nov. 2009) from 16 representative stations within the Bay. Samples were collected by submerging a 5-L Niskin bottle bellow the surface and were preserved with modified Lugol solution. Measurements of physicochemical parameters such as temperature, transparency, salinity, inorganic nutrients, dissolved oxygen and chlorophyll *a* were made concurrently at each station.

For taxonomic purposes, samples were taken with a 20 µm mesh Nytal net and were fixed with a 4 % formalin solution. Algal taxa were identified to species using a number of taxonomic texts (Hallegraeff *et*. *al*., 2004; Tomas, 1997). Abundance expressed as density (cells/L) was determined using a sedimentation technique to concentrate the phytoplankton. All cells, colonies, and filaments were counted and measured with a binocular microscope using a Sedgewick Rafter counting cell.

Overall eutrophic condition

Some parameters of ASSETS (Assessment of Estuarine Trophic Status) methodology (Bricker *et al*., 2003) such as chlorophyll *a* and dissolved oxygen were applied comparatively to rank the eutrophication status of the Bay. Chlorophyll *a* is considered a primary symptom (excessive concentration is considered to diagnose an early stage of an eutrophication problem. Low dissolved oxygen and occurrence of nuisance and/or toxic algal blooms are considered as secondary symptoms (indicators of well developed eutrophic conditions) (Table 1).

Table 1. Indicator parameters for primary and secondary symptoms of estuarine eutrophication .

Primary symptom		Secondary symptom	
Indicator	**Thresholds and ranges**	**Indicator**	**Thresholds and ranges**
Chlorophyll *a*	Hypereutrophic: >60 µg Chl*a* L^{-1}	**Dissolved oxygen**	Anoxia: 0 mg L^{-1}
	High: >20 but ≤60 µg Chl*a* L^{-1}		Hypoxia: >0 but ≤ mg L^{-1}
	Medium : >5 but ≤20 µg Chl*a* L^{-1}		Biologically stressful: >2 but ≤5 mgL^{-1}
	Low: >0 but ≤5 µg Chl*a* L^{-1}		

RESULTS AND DISCUSION

Primary symptom

Abundance of phytoplankton (Chlorophyll *a*)

Maximum values of chlorophyll *a* were observed during the rainy period (September), with an average value of 6.56 µg/L which classifies the estuary within the "Medium" level of expression (>5 to ≤20 µg L^{-1}) of eutrophic conditions. The second peak of chlorophyll *a* occurred in early rainy period (June), the average value (3.70 µg/L) falls within the "Low" category although conditions of medium level were observed at some stations (Fig. 2).

The medium chlorophyll *a* concentrations were qualitatively related to a decrease of salinity during the rainy period which was characterized by salinity values in the range of 23.3 - 33.7 psu with a average value of 27.9 psu (Fig. 3). Generally, the freshwater discharges and the rains are associated with nutrient input, as well as with the increase of chlorophyll *a* values in the estuaries.

During the late dry period (April), although high and medium values of chlorophyll *a* were observed at three stations close to enriched areas (stations 8, 9 and 10), a "Low" level of expression of eutrophic conditions was obtained for most of estuary. In the early dry period (November), values of chlorophyll *a* classified in the "Low" eutrophic class (Fig. 2).

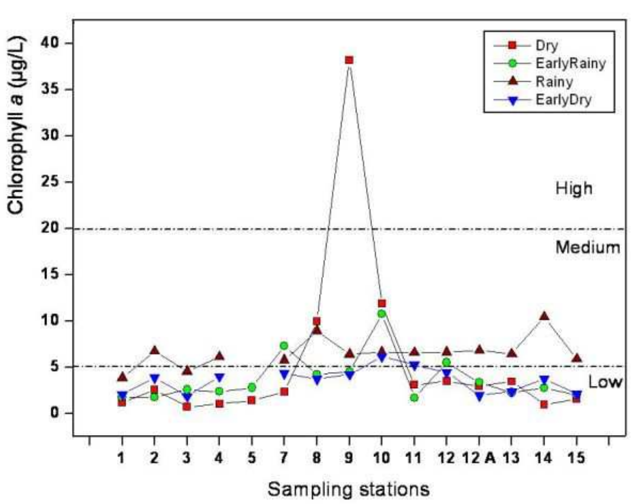

Fig. 2. Seasonal and spatial variation of Chlorophyll *a*.

The low depth, semi-enclosed morphology that produce a restricted circulation; the increase of water temperature; and the occurrence of discrete rains which result in a high stability and salinity of water, and reduced flushing potential seem to be the main factors contributing to the occurrence of peaks of chlorophyll *a* in these shallow enriched areas (stations 8, 9 and 10) during late dry period (April).

High abundance of phytoplankton (blooms) were also observed during the early rainy period (June) in similar shallow sites but out of the monitoring network, located near sewage effluents from Cienfuegos city (near station 12) The early rainy period was characterized by high salinity values (30.50 -33.90 psu), with an average value of 32.78 psu. In most of sampling stations during the four campaigns (April, June, September and November), the salinity values were above 25 psu, thus all system should be classified as a seawater zone (Fig. 3).

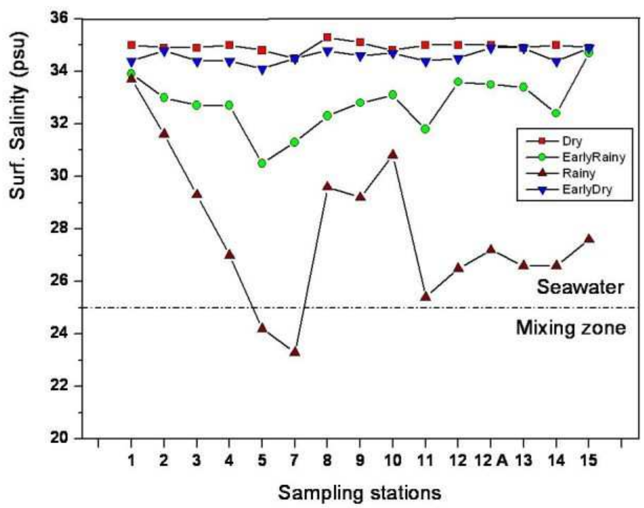

Fig. 3. Seasonal and spatial variation of salinity

A good spatial and seasonal qualitative relationship was not observed among nutrients and chlorophyll *a*, which is in correspondence with a criterion found in US-EPA (2001) about that these two types of variables (causal and response) cannot be correlated well. This could be showing a non immediate response from this aquatic system to the nutrients changes.

The higher concentration of chlorophyll *a* during the rainy period at the station 10 and 12 suggests the importance of the urban inputs at this time at these stations, in spite of the increased dilution in this system. Although both nutrients, during the rainy season, showed lower average values (Table 2) than those obtained during the other periods considered, the maximum value of dissolved inorganic phosphorus (DIP) of analyzed period was obtained in one of these two stations (station 12) at the onset of the rainy season.

Despite the maximum values of nutrients found during the rainy season, eutrophic conditions were not observed in any of the stations during the rainy period. This could be due to low residence time, which according to US-EPA (2001), when it is lower than the time of algae cellular division, algal bloom formation is inhibited.

Table 2. Results of nutrients concentrations in waters of Cienfuegos Bay (2009)

Nutrients (umol/L)	**Rainy period**			**Dry period**		
	Mean	**Min.**	**Max.**	**Mean**	**Min.**	**Max.**
DIP	0,93	<0,52	9,32	1,48	<0,52	3,90
DIN	4,13	<2,57	11,6	4,40	<2,57	12,0

DIN: dissolved inorganic nitrogen
DIP: dissolved inorganic phosphorus

Secondary symptoms

Dissolved Oxygen

Most data of surface-water dissolved oxygen are higher than the threshold for "Biological stress" (>2 to ≤5 mgL^{-1}). Only one value fell within this category. This value was obtained in station 15 during the early rainy period. No problems with oxygen depletion in the surface water due to excesive growth of microalgae have been observed at stations near urban wastewater sources. Although most of the bottom-water oxygen values are within the range of 5 to 7 mg/L, stations 4, 5, 8, 9, 11, 13 and 15 showed some values below this threshold, particularly stations 5, 9 and 11 during the early rainy and rainy periods when the organic enrichment processes are increased (Fig. 4).

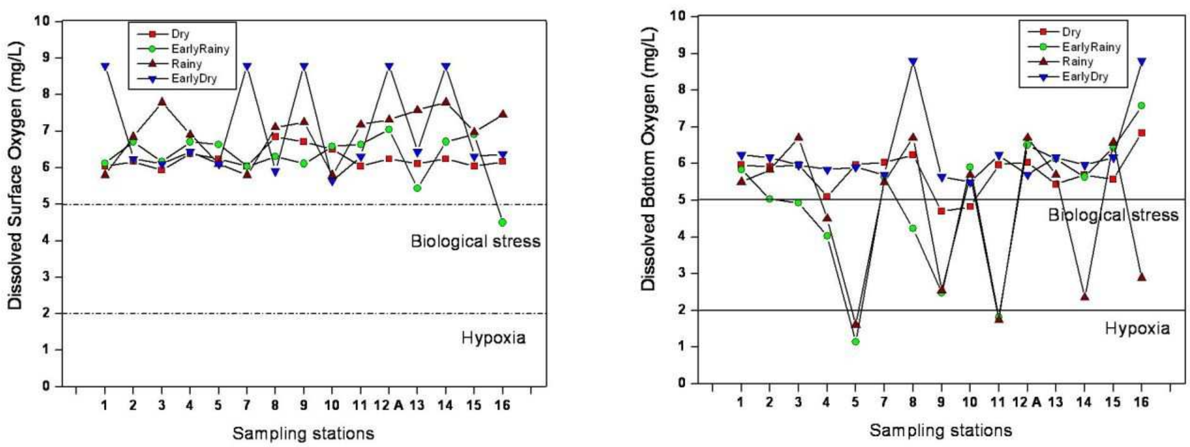

Fig. 4. Seasonal and spatial variation of surface and bottom water dissolved oxygen

The low bottom-water oxygen concentration at stations 5, 9 and 11 could be due to organic accumulation associated with the estuarine hydrodynamic movement in the north lobule of this bay (station 5) and to organic enrichment associated with discharges of industrial and urban wastewaters (stations 9 and 11). These stations are deep which contribute to favourable conditions for accumulating and decomposing organic matter. Higher water circulation exhibited at station 11 is likely to be the cause of e of phytoplankton bloom in this area. However, depletion of bottom-water oxygen concentration at station 9 could be associated with the occurrence of dinoflagellate blooms observed in this area of more restricted water circulation.

Composition of phytoplanktonic community (Nuisance and toxic blooms)

From the overall point of view, throughout the year, at this ecosystem, the phytoplankton growing season starts in the early rainy period (June) and its maximal density occurs in the rainy period (September). The presence of a moderate density and diversity of diatoms characterized the early rainy and rainy period in all the Bay. The most abundant species in these periods were *Dactyliosolen fragilissimus* (3.4 x 10^7 cells/L), *Rhizosolenia hebetata* f. *semispina* (7.5 x 10^5 cells/L), *Thalassionema nitzschioides* (5.1 x 10^5 cells/L) and *Chaetoceros lorenzianus* (4.6 x 10^4 cells/L), typical species of Cienfuegos Bay (Moreira *et al*., 2007). Generally, moderate freshwater discharges and the rains are associated with an increase of phytoplankton diversity in the estuaries.

However, a low phytoplanktonic diversity and the occurrence of blooms in areas with sewage influence characterized the late dry period (April). Blooms of the non toxic dinoflagellates *Gymnodinium estuariale* and *Gyrodinium instriatum* (Fig. 5) were observed in stations 8 and 9. The presence of a high concentration of the potentially amnesic toxic diatom *Pseudo-nitzschia multistriata* (Fig. 5) was detected with a density of 1.1 x 10^6 cells/L, at station 10. This species was present but in moderate concentrations at several stations during this period.

Blooms of other non toxic dinoflagellates such as *Peridinium quinquecorne* and *Prorocentrum compressum* (Fig. 5) are also frequent at the same areas influenced by urban wastewater (near station 9, 10 and 12), during early rainy period (June), early (November) and late (April) dry period (Moreira *et al*., 2009).

It should be noted that during the early rainy period (June), in a shallow site with restricted circulation, out of the monitoring network and located near domestic effluents from Cienfuegos city (near station 12), a observed bloom of the toxic dinoflagellate *Heterocapsa circularisquama* (Fig.6) was associated with a small episode of fish kill. Some potentially toxic dinoflagellates (Fig. 6) were found in this zone but in low concentration, including *Alexandrium* cf *minutum* and *Gymnodinium catenatum*, potentially producers of paralytic shellfish poisoning (PSP); *Dinophysis caudata and D. ovum*, potentially producers of diarrhetic shellfish poisoning (DSP); *Amphidinium carterae*, *Cochlodinium* cf. *polykrikoides*, *Gonyaulax polygramma* and *G. spinifera*, potentially producer of icthiotoxic compounds; *Prorocentrum minimum*, potentially producer of neurotoxic compounds; and *P.* r*athymum*, potentially producer of hemolytic compounds that could increase the effects of ciguatera due to its benthic life.

In the year 2009, despite intermittent rains, an increase of phytoplankton species richness was observed and potentially toxic and noxious species were replaced by non toxic diatoms during the rainy period.

Potentially toxic freshwater cyanobacteria were detected in very low concentrations at the stations with river influence (stations 7, 8 and 14). However, in the last years, the impact of tropical storms and hurricanes (e.g. Dennis Hurricane in 2005) on the bay and its associated excess of freshwater loading have caused high concentration of small freshwater chlorophyceans and cyanobacteria, including toxic species (e.g. *Planktothrix isothrix* and *Pseudoanabaena* sp.) in all the Bay; and a shift in marine phytoplankton species composition was observed. In these events, other potentially toxic cyanobacteria species (e.g. *Microcystis aeruginosa*, *Oscillatoria* sp. and *Anabaena* sp.) (Fig. 7) were registered in moderate concentration.

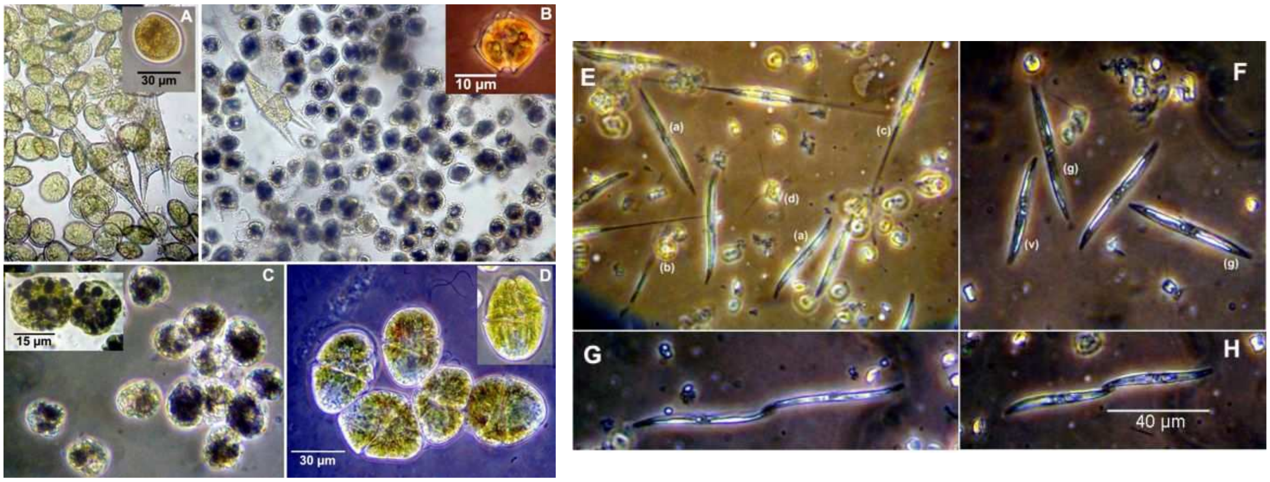

Fig. 5. Non toxic dinoflagellates (A-D) and potentially toxic diatom *Pseudo-nitzschia multistriata* (F-H) which form blooms in discharge areas; A. *Prorocentrum compressum*, B. *Peridinium quinquecorne*, C. *Gymnodinium estuariale*, D. *Gyrodinium instriatum*.

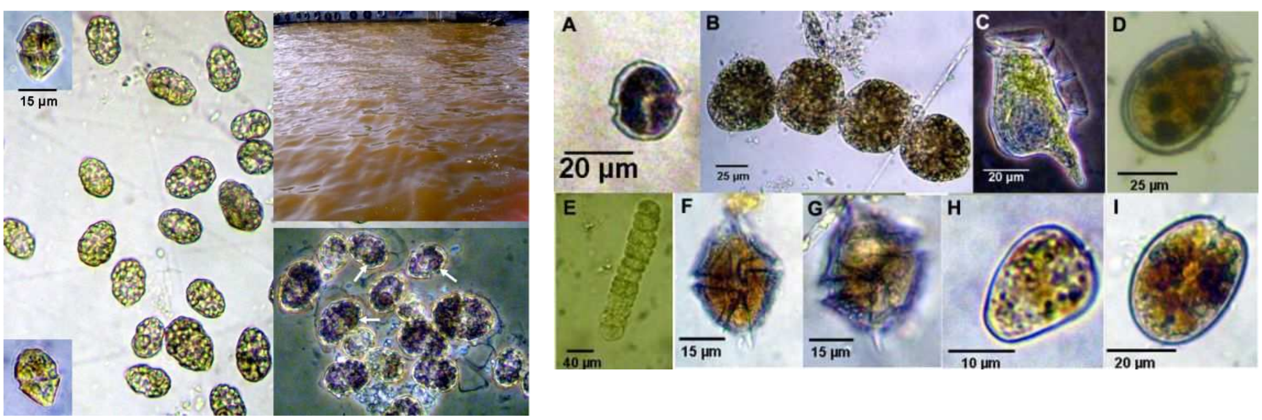

Fig. 6. Bloom of the toxic dinoflagellate *Heterocapsa circularisquama* (Left) and other toxic or noxious species associated in waste discharge areas; A. *Alexandrium* cf *minutum*, B. *Gymnodinium catenatum*, C. *Dinophysis caudata*, D. *D. ovum*, E. *Cochlodinium* cf. *polykrikoides*, F. *Gonyaulax polygramma*, G. *G. spinifera*, H. *Prorocentrum minimum*, I. *P.* r*athymum*.

Additionally, coinciding with the occurrence of dinoflagellates blooms at stations near sewage effluents, moderate abundance of potentially ciguatotoxic and ichthyotoxic epibenthic microalgae were registered on blooms of filamentous epiphytic macroalgae in shallow zones near these stations (e.g. stations 12, 12A, 13) during the late dry and early rainy periods .

Prorocentrum concavum, *P. emarginatum*, *P. lima*, *P. belizeanum*, *Ostreopsis ovata* and *O. siamensis* (Fig. 8) were the most abundant species associated to the ciguatera syndrome, while *Gambierdiscus toxicus*, principal agent of ciguatera was not registered within the Bay, it was only found in the connation channel of the Bay with the Caribbean Sea.

Lyngbya majuscula (Fig. 7) was the most abundant dermatotoxic species in the Bay. This species could be responsible for contact irritation (known as "swimmer itch") illness that affects to swimmers at the beaches of Cienfuegos Bay, principally between the early rainy and the rainy periods. Blooms of *Lyngbya majuscula* have been found to be neurotoxic and are also related to gastrointestinal diseases (Nogle *et al*., 2001).

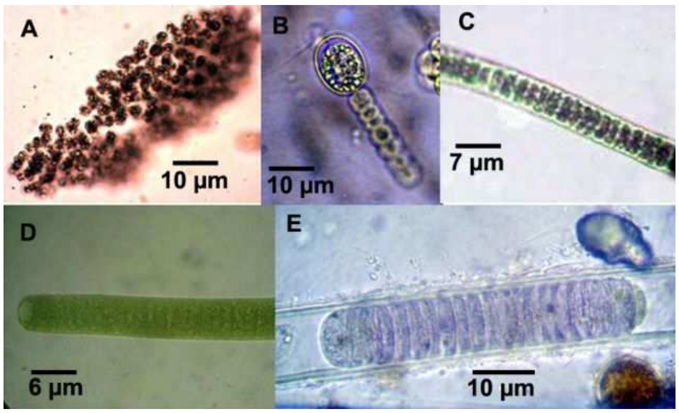

Fig. 7. Potentially toxic cyanophytes, A. *Microcystis aeruginosa*, B. *Anabaena* sp., C. *Planktothrix isothrix*, D. *Oscillatoria* sp., E. *Lyngbya majuscula*.

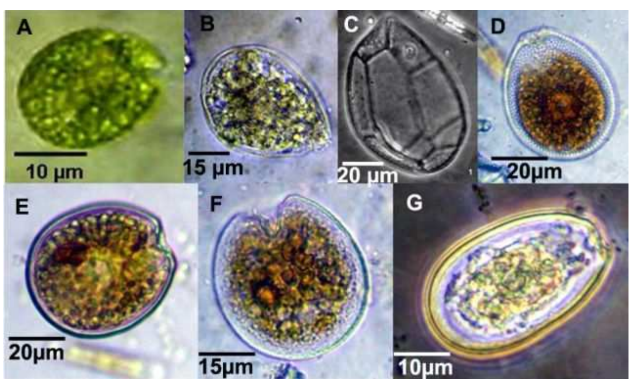

Fig. 8 Potentially toxic benthic dinoflagellates, A. Amphidinium carterae, B. *Ostreopsis ovata* , C. *O. siamensis*, D. *Prorocentrum belizeanum*, E. *P. concavum*, F. *P. emarginatum*, G. *P. lima*.

Epiphytes blooms were dominated by the filamentous brown macroalgae *Feldmannia irregularis* and *Hincksia mitchelliae*. These epiphytes growth principally on *Halodule wrightii*, which is the main seagrass and most abundant macrophyte in Cienfuegos Bay. The overgrowth of epiphytes and dinoflagellates are the main cause of submerged aquatic vegetation depletion in areas with sewage influence in Cienfuegos Bay due to reduced light availability and smothering. During the rainy period the opportunistic brown macroalgae were being gradually replaced by the coarse red macroalgae *Hypnea spinella*, *Acanthophora spicifera*, *Gracilaria pauciramosa*, *G. blodgettii* and *Padina sanctae-crucis*, most abundant seaweeds of Cienfuegos Bay.

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