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ANNUAL NANO-MICROPHYTOPLANKTON SUCCESSION IN THE MEROMICTIC BRACKISH LAKE FARO (MESSINA - SICILY) IN RELATION TO PHYSICAL-CHEMICAL CONDITIONS

SUMMARY

A multi-disciplinary study was carried out in the meromictic brackish "Lake Faro" from May 1998 to April 1999, with the emphasis on correlation between nano-microphytoplankton monthly succession and physical-chemical characteristics of the water column. It has been stated that the seasonal dynamics of the phytoplankton community is strongly influenced by the supply of phreatic waters. A partial spring mixing sustained a summer bloom of Diatoms (*Chaetoceros* genus) and, in July, Dinoflagellates reached a maximum density at 10 m depth in microaerobic waters.

After the autumnal decrease, an input of phreatic waters gradually increases phytoplankton growth at the end of autumn. Afterwards, the winter thermal inversion and a mixing involving the upper layer of the hypolimnion in January sustained the growth, of nanophytoflagellates (6.6×10^6 cells L⁻¹) in February and Diatoms (tiny Centrales 9 x 10⁶ cells L⁻¹) in March. Nano-microzooplanktonic Ciliates, adapted to living in the redox zone, graze an abundant population of photosynthetic bacteria living in the chemocline and the other microorganisms living in the upper layer of the hypolimnion.

RIASSUNTO

Successione annuale del nano-microfitoplancton nel lago meromittico salmastro di Faro (Messina, Sicilia) in relazione alle condizioni chimico-fisiche. Sono descritte le dinamiche delle successioni mensili del nano-microfitoplancton, in correlazione con alcuni parametri fisico-chimici, rilevate in una ricerca svolta dal Maggio 1998 all'Aprile 1999 nel lago meromittico salmastro di Faro (Messina). Le dinamiche stagionali sono fortemente influenzate dall'infiltrazione di acque freatiche. In primavera una parziale miscelazione delle acque ha permesso una cospicua fioritura estiva di Diatomee (*Chae-toceros* spp.) mentre a Luglio i Dinoflagellati hanno raggiunto le massime densità in acque microae-robiche alla quota di 10 m. Dopo i bassi valori autunnali, le popolazioni fitoplanctoniche sono state stimolate dall'infiltrazione di acque freatiche alla fine della stagione.

Infine a seguito dell'inversione termica invernale e della miscelazione delle acque, che a Gennaio coinvolgeva anche il livello superiore dell'ipolimnio, sono state osservate fioriture importanti di nanofitoflagellati (6.6 x 10^6 cells L^{-1}) a Febbraio e Diatomee (9 x 10^6 cells L^{-1}), per lo più piccole Centrales, a Marzo. Infine si discute sulla presenza di numerosi Protozoi Ciliati nano-microplanctonici adattati a vivere in ambiente redox, dove si nutrono dei batteri fotosintetici del chemioclino e degli altri microrganismi viventi nello strato superiore dell'ipolimnio.

INTRODUCTION

The brackish Lake of Faro is located at the north-eastern point of Sicily, Capo Peloro, northeast of the nearby Lake of Ganzirri (Fig. 1). It is also known as "Pantano Piccolo", has a surface area of $2.63 \times 10^5 \text{ m}^2$, a volume of $2.5 \times 10^6 \text{ m}^3$ and a maximum depth of 29 meters (ABRUZZESE & GENOVESE, 1952). Lake Faro is linked to the sea and Lake Ganzirri by some artificial canals. The long, narrow "Margi" canal links the two lakes. The "Canalone di Faro" links the lake to the Ionic side of the Straits of Messina; finally, the wider "Canale degli Inglesi" opens to the north on the Tyrrhenian Sea (Fig. 3). This canal is usually silted, but for some weeks, in the summer, it is usually partial-



Fig. 1 — Sicily and the area of the Straits of Messina with the two coastal brackish lagoons: Faro and Ganzirri.

ly opened, in order to improve oxygen regime. Both lakes form the residue of a larger lagoon system which was originally formed by alluvial material brought by numerous torrents.

Lake Faro is a typical brackish meromictic basin and its aerobic epilimnion has a maximum depth of 15 m (Fig. 2). In summer the epilimnion significantly decreases due to the rise of the hypolimnion with hydrogen sulphide. Both zones are separated by the picnocline (being also a thermocline and a chemocline) in the metalimnion, forming the redox zone in a layer with a depth varying between 8 and 14 m. It harbours a dense population of anaerobic photosynthetic bacteria as was observed in other marine meromictic lakes, in different environments and latitudes In summer it slows down the spread of hydrogen sulphide on the surface layer (CRISAFI, 1956; GENOVESE, 1963; SOROKIN & DONATO, 1975).

The typical conditions of a meromictic environment are partially disturbed by the limited entrance of Tyrrhenian seawater through an artificial channel. Exceptional cases such as a complete opening of this channel by mussel farmers or extremely violent tides cause accidental alteration of the anoxic layer at deeper levels with temporary disastrous consequences on all aerobic organisms (GENOVESE, 1962, 1963; BRUNI & GRILLO, 1981).

Although many studies have been carried out in the lagoons of Ganzirri and Faro, few have involved the production of the first trophic levels. A trophic model representing Lake Faro (PUCCIA & HULL, 1988) was present-



Fig. 2 — Faro Lake section W-E with frustum cone trench.

ed in 1988 and overall features of its biological and physical-chemical regime has been described (Bruni *et al.*, 1976), photosynthetic activity of picoplankton (PLATT *et al.*, 1985; ACOSTA POMAR *et al.*, 1988) and seasonal observations (GIUFFRĚ, 1991) on phytoplankton composition were investigated.

This research, on the monthly succession and distribution of some components of the first levels of trophic net, was carried out as part of a global national study project on Italian brackish environments financed by MURST (Ministero dell'Università e della Ricerca Scientifica e Tecnologica). From early results it has been seen how samplings, carried out every 15 days, showed lower summer levels of chlorophyll "*a*" and POM in Lake Faro than in the nearby marsh of Ganzirri (CORTESE *et al.*, 2000). The research on picoplankton, carried out seasonally, has shown a higher picophytoplanktonic biomass in summer, due to the presence of Cyanobacteria in the deep layer of Lake Faro (MAUGERI *et al.*, 2000 a, b).

Method

Monthly phytoplankton sampling was carried out over a year, every 15 days, together with physical-chemical observations, using a multiparametric Idronaut Ocean Seven mod. 401 probe, and with meteorological data. Four stations were sampled: station 13 (5.80 m depth), station 15 (1.80 m), station 17 (10 m) and station 18 (29 m) (Fig. 3). Samples were taken with Niskin bot-



Fig. 3 — The meromictic Faro lagoon with the sampling stations and the bathymetric lines (solid lines 5 m, dotted lines 1 m).

tles from the surface at all stations; at the deepest station 18, depths 5, 10, 15 and 20 m were also sampled. Samples were fixed with formaldehyde, buffered in hexamethylenetetramine, at a final concentration of 1.6%. 100 ml of every sample was sedimented, in conformity with the Utermöhl method for 48 hours, and analysed using a Zeiss invertoscope, with a 40 x objective.

Haptophyceae and Chrysophyceae, found occasionally, were described, but were counted within "Unidentified phytoflagellates". The nano-microzooplankton was not a target of this research. Nevertheless, the numerous ciliated Protozoa were counted in the slides for phytoplankton, sedimented in conformity with the Utermöhl method.

RESULTS

Hydrographic parameters

Salinity and rainfall. A decrease in salinity at the intermediate layer (15–20 m) was observed in September and November (Fig. 4) due to the inflow of the ground water table. However, in September this was fed not so much by the little rainfall of the previous months as by the more important inflow in November (Fig. 5), but by increased anthropic activity, starting in June. This facilitated a later sinking of denser surface waters. The increase in rainfall at the end of 1998 (Fig. 5) established a permanent stratification in October with less salty surface waters which lasted until spring of the following year. The thermal homogeneity in November and the inversion in January gave rise to the homohaline waters mixing phase (Fig. 4).

Temperature, oxygen and pH. The gradual summer increasing of temperature and decrease of pH and O_2 values (Figs 4 and 6) have been transitorily stopped by the partial opening of the artificial "Canale degli Inglesi" canal in July, with the input of colder and more haline sea water sinking in the depth.

At the end of the summer thermic stratification (Fig. 4) an increase of pH and oxygen values (Fig. 6) was observed in the metalimnion and in the intermediate deep layers of the epilimnion too. Nevertheless, the phase of thermal homogeneity and the inflow of phreatic waters in November favored the spread of poorly oxygenated water towards the surface and the decrease of pH and oxygen values in the epilimnion (Fig. 6).

These conditions, together with high water turbidity and the winter low sun irradiation, did not support phytoplankton growth. However, it is evident that the nutrient enrichment induced by the phase of mixing in January, with a massive input of N-NO₃ and P (CORTESE *et al.*, 2000), triggered a later gradual phytoplankton growth and oxygen values (Fig. 6).



Faro Lake - T (°C) - St. 18



Fig. 4 — Water salinity and temperature: annual vertical distribution (station 18).



Fig. 5 — Annual rainfall. The histograms show monthly rainwater millimetres before sampling.

Phytoplankton

Numerous planktonic and also tychopelagic species belonging to Bacillariophyceae, Dinophyceae, Euglenophyceae, Haptophyceae and Chrysophyceae were found in the waters sampled.

Bacillariophyceae and unidentified nanoplanktonic phytoflagellates. Two maxima of unidentified nanoplanktonic phytoflagellates were recorded at the surface, one in early summer and the second in winter. Over the year, in the surface layer, the populations of phytoflagellates and of the Diatoms alternated (Figs 7 and 8). At the end of spring 1998 a diatom bloom of *Chaetoceros lacinio-sus* and *C. curvisetus* (6×10^6 cells L⁻¹) was observed on the surface at station 17.

The surface water exchanges with the Ionic side of the Straits of Messina (Canalone di Faro) and the lake of Ganzirri (Canale Margi) (Fig. 3) due, not only to winds, but also to tidal dynamics, caused a maximum of unidentified phytoflagellates, reaching a density of 6.6×10^6 cells L⁻¹ at station 13 in February 1999, and after, in March at station 17, together with Diatoms which attained their maximum density (9 ×10⁶ cells L⁻¹), due to tiny Centrales.

In summer a general increase in diatom density was recorded all over the lake, even in intermediate deep waters (Fig. 8), with a peak on August 26^{th} in



Faro Lake - O₂ (ml/l) - St. 18



Fig. 6 — Water pH and oxygen: annual vertical distribution (station 18).

superficial layers. The average density in surface was close to 5×10^{6} cells L⁻¹, with a dominance of *Chaetoceros* cfr. *brevis* and *C. minimus*, but a decrease in diatom density was seen at a depth of 10 meters, correlated to the rise of the anoxic hypolimnion (Fig. 6). Bacillariophyceae were often dominant with a few species, nevertheless numerous taxa were identified.

After the winter mixing phase, the spring rise of phytoplankton density reached its maximum rate in March (Figs 7 and 8), when the thermic stratification was re-established and solar irradiation had increased.

Dinophyceae. Over the year a modest presence of Dinoflagellates was observed. Gynmodinium sanguineum Hiras was noted in many samplings with, occasionally, G. pygmaeum Lebour; less frequently the genera Oxytoxum, with O. variabile Schiller, O. adriaticum Schiller, O. mediterraneum Schiller and O. milneri Murr., Protoperidinium, with P. cfr. pellucidum Balech, P. breve (Paulsen) Bergh, P. bipes (Paulsen) Balech and P. quinquecorne Abé, Dinophysis, with D. acuminata Clap. & Lach. and D. sphaerica Stein and, finally occasionally Prorocentrum triestinum Schiller and Oxyphysis oxytoxoides Kofoid were seen.

The Dinophyceae maximum density $(5 \times 10^5 \text{ cells } \text{L}^{-1})$ was reached at the beginning of July at 10 meters (Fig. 9). Nevertheless they were often present in surface and in deep waters and it is proved that poorly oxygenated inter-



Fig. 7 — Phytoplankton (Bacillariophyceae and all phytoflagellates taxa) annual superficial trend (average) of stations 13, 15, 17 and 18.

Faro Lake - Unidentified Phytoflagellates - St. 18

Faro Lake - Bacillariophyceae - St. 18



Fig. 8 — Unidentified Phytoflagellates and Bacillariophyceae: annual vertical distribution (station 18); numbers in the legend are the cube root of cells l^{-1} .



Fig. 9 — Dinophyceae, Euglenophyceae and Protozoa Ciliata: annual vertical distribution (station 18); numbers in the legend are the square root of cells l^{-1} .

mediate deep layers are the most favourable. In August, during the enhancement of the anoxic deep layer (Fig. 6), Dinoflagellates density weakly increased in microaerobic waters at 5 m (Fig. 9) and decreased at 10 m.

Euglenophyceae. Euglenophyceae appeared in the plankton in October-January (Fig. 9) and then again in spring. They were present with the *Eutreptiella* genus, mainly *E.* cfr. *marina* Cunha, but also *Euglena* cfr. *acusformis* Schiller and *E.* cfr. *acus* Ehr., were seen. Euglenophyceae reached maximum values in October in the surface layer at station 18 with 10^5 cells L⁻¹.

Chrysophyceae and Haptophyceae. Chrysophyceae were found in May with numerous colonies of *Dinobryon* in all stations and at nearly all depths with 3.7×10^5 cells L⁻¹ and later for all the summer with a few specimens belonging to the *Calycomonas* genus.

In May 1998 there was an occasional presence, at 10 meters, of Haptophyceae with *Calyptrosphaera superba* Lecal and then, in July, with *C. sphaeroidea* Schiller, probably residue traces of denser water coming from the sea and then sunk. Finally, single specimens of *Umbelicosphaera sibogae* (Weber-van Bosse) Gaarder, *Ophiaster hydroideus* (Lohm.) Manton and Oates and *Emiliania huxley* (Lohm.) Hay and Mohler were noted.

Protozoa Ciliata. At the start of the summer rise of hypolimnion, ciliated Protozoa were totally absent only at the beginning of July (Fig. 9) when, at 10 meters, a marked increase in water temperature was observed (Fig. 4), together with the maximum number of Dinoflagellates (Fig. 9), while oxygen and pH values (Fig. 6) decreased throughout the water column.

Nevertheless, after a partial opening of the "Canale degli Inglesi", about the middle of July, it is worth noting that Protozoan Ciliates clearly dominated at deeper intermediate layers, particularly at 15 meters (29 July; $3,1x10^5$ cells L⁻¹), often in anoxic conditions, and were also frequently found at 20 meters. Tintinnida, belonging to the *Favella* and *Tintinnus* genera, were identified on only a few occasions.

Organic detritus. Abundant tiny organic detritus was often found in the samples and, from October 1998 to the following spring, a large amount of transparent exopolymeric particles (TEP), probably polysaccharide and glycoprotein algal exudates, were present.

DISCUSSION

The annual phytoplankton succession showed an alternate predominance between Bacillariophyceae and Phytoflagellates (Fig. 7) with two main growth phases, one in spring-summer and the other at the end of the winter, with the latter being more important, due to the nutrient increase originating from the mixing and the autumnal rains with the consequent input of phreatic water at all levels.

The lake generally shows a notable succession variation with a moderate richness in species and some blooms of phytoplankton populations. This can be expected in a coastal brackish environment with a clear dominance of the nanophytoplanktonic dimensional fraction belonging, above all, to the phytoflagellates, but sometimes also to Diatoms, as in March 1999. Here some Dinoflagellates species were shown to prefer microaerobic conditions, with a higher density at 10 meters, near the metalimnion, up to the anoxic hypolimnion and this can be explained only in part by sinking.

The comparison between annual trends of Dinoflagellates and Ciliates, particularly in summer, would seem to alternate, but above all both taxa prevalently live at different deep layers (Fig. 9).

A large number of Dinoflagellates are mixotrophic or eterotrophic and it is possible to hypothesize that some species of Faro Lake are also able to compete with the Ciliates. There is evidence that some protozoan species prefer the relatively very deep layers even if they are anoxic. Actually, ciliated Protozoa, found during our research, above all in the deep layer, had already been observed in Lake Faro and it has been hypothesized (SOROKIN & DONA-TO, 1975) that some benthic genera (Hypotrichida: *Trachelostyla caudata; Urostyla* sp.; 1 g/m³ and 2 ×10⁴ cells L⁻¹) are able to "graze" while swimming in the metalimnion, which is extremely rich in photosynthetic anaerobic bacteria; the digestive vacuoles of the Ciliates, in fact, were red coloured for the presence of numerous specimens of *Chlorobium phaeobacteroides*. Therefore SOROKIN & DONATO (1975) showed the capacity of these Hypotrichida to abandon typical movements on a substratum and to swim in the upper part of the anoxic layer, tolerating a concentration of even 40 mg L⁻¹ of H₂S.

A significant presence of benthic Ciliates living in the plankton $(0.5 - 1 \text{ g/m}^3)$ of Faro Lake was again observed by SOROKIN & GENOVESE (1982) with numerous Zooflagellates, including *Bodo* and *Monas* genera, reaching a biomass of 0.5 g/m³ at depths of 13.5 – 14.5 m.

They can, therefore, exploit an abundant food resource nearly in the absence of any of their competitors or predators. Actually, the metalimnion layer, with brownish-red waters, is predominantly populated by numerous *Chlorobium phaeobacteroides* Pfennig (Chlorobiaceae), photosynthetic sulphur bacteria with bacteriochlorophyll "*d*" (TRÜPER & GENOVESE, 1968) but other genera were also found such as *Chromatium, Thyopolicoccus, Thyocistis* (GENOVESE, 1963), *Rhodopseudomonas* and *Prosthecochloris* (BRUNI *et al.*, 1978). Photosynthetic production rate was estimated from 2.5 to 5 μ M of C daily (SOROKIN & DONATO, 1975) and a notable biomass (30 g/m³) of photosynthesising bacteria *Chlorobium phaeovibrioides* was identified (SOROKIN &

GENOVESE, 1982). This layer of "red waters" is an ecological barrier and contributes to hinder the spread of hydrogen sulphide.

There is evidence that Protozoa Ciliata were not temporarily "swimming" in the chemocline, but adapted to live in deep anoxic layers. Such a tendency to live in a reducing environment has sometimes been described in the sulphide-rich hypolimnion and, for example, in a lake in Spain, at least seven species of Ciliates (FINLAY *et al.*, 1991) were found. Here different combinations of symbiotic bacteria were found within the various identified Ciliates species, but methanogenic autofluorescent bacteria were present in all.

Microrganisms living in microaerobic or anaerobic environments, such as anaerobic fungi and protozoa, do not generally use mitochondria as the powerhouses of the cell, but hydrogenosomes, membrane-bounded organelles, about 1 μ m in diameter that resemble mitochondria, from which they have possibly derived as an adaptation to anaerobic or microaerobic environments. They produce ATP and molecular hydrogen, reducing protons because they cannot use oxygen as an electron acceptor (EMBLEY & MARTIN, 1998). They are surrounded by endosymbiotic methanogenic archaea because of the presence of the hydrogen (FENCHEL & FINLAY, 1995).

The singular characteristics of Lake Faro, which distinguish its composition, distribution and dynamics of biotic components from the nearby Lake Ganzirri, are the result, not only of various links with the sea, but basically of physicalchemical and biotic effects which derive from its bathymetric profile. Winds and tides cause the circulation of surface water layers between the "Canalone di Faro" and the "Margi" canal (Fig. 3) on the south-eastern side, and marginally, in the central zone, but a trench cuts the shallow bottom in a slightly decentralized position (Fig. 2) and hinders hydrodynamic action at deeper layers.

Seasonal variations and meteorological conditions can only influence the metalimnion and the most superficial layers of the hypolimnion, which can then rise or sink only a few meters. The organic detritus, the phototrophic, chemorganotrophic (heterotrophic) and chemolithotrophic prokaryotes, the nano-microphytoplankton eukaryotes and the abundant nano-microzoo-planktons, such as Ciliates, activate a "microbial loop" which maintains a striking amount of energy, organic substance and nutrients circulating within it. The biological characteristics and productivity of Lake Faro can be better understood in the context of a "micro-litrosphere", the short microbial trophic network present in the marine environment (SIEBURTH & DAVIS, 1982), as defined in its autotrophic, heterotrophic and mixotrophic components (FOGG, 1986a, b).

The "microbial loop" significantly affects the lake productivity due to the more rapid metabolism of its tiny components, even though these components are not always present in considerable biomass (Fig. 10).



Fig. 10 — Simplified microbial loop drawing of microlitrosphere (marked by thicker arrows) in the trophic net of the Faro marsh. (APB = AnoxicPhotosynthetic Bacteria; HB = Heterotrophic Bacteria; **PP** = PicoPhytoplankton; NMP NanoMicroPhytoplankton: **PC** = Protozoa Ciliata; $\mathbf{N} = \text{Nutrients}; \mathbf{MA} =$ MacroAlgae; \mathbf{E} = Herbivores; C = Carnivores; FF = Filter Feeders).

The intense biological cycles of micro-organisms can therefore satisfy the trophic needs of a large quantity of filter-feeders which, in this environment are made up, not only of bivalves introduced by mussel farmers, but also of native populations, such as numerous Ascidians, Polichaeta worms and finally Porifera, all of which often encrust submerged surfaces in a noticeable way.

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