Contribution of Chlorophyta and non-Chlorophyta to the picoeukaryotic community in the Gulf of Gabès (Eastern Mediterranean Sea)

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INTRODUCTION

Picoplanktonic (cells smaller than 2-3 microns) eukaryotes contribute significantly to both biomass and primary production in oligotrophic areas of the world's oceans (Campbell *et al*., 1994; Li, 1994) and can also be important in coastal waters (Courties *et al*., 1994; Campbell *et al*., 1998). They are an essential component of microbial food webs, which play important roles in mineral cycles.

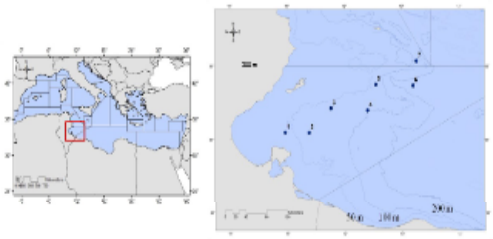
Quantifying the planktonic picoeukaryotes community is indeed crucial to understand the structure and the dynamics of marine ecosystems. Picoeukaryotic abundance was almost 103 cells mL-1 in many oligotrophic environments and reached 105 cells mL-1 in coastal waters (Worden and Not, 2008); and it ranges between 102 and 104 cells mL-1 in the photic zone of oceans (Massana, 2011).

In the Gulf of Gabès, characterized by oligotrophic conditions and a dynamic water masses circulation, information on the abundance of total picoeukaryotes is lacking although studies based on high-performance liquid chromatography and flow cytometry have revealed that picoplankton was among the main contributor to the ultraphytoplankton group (Hamdi *et al*., 2015) and with the nanoplankton can account for up to 90% of the overall chlorophyll biomass (Bel Hassen *et al*, 2009).

The aims of this study are:

- To determine the abundance and the spatial distribution of picoeukaryotes in the Gulf of Gabès.
- To investigate the contribution of Chlorophyta and non-Chlorophyta Divisions to picoeukaryotic community.
- to assess the effect of hydrographic features and nutrients availability on the spatial distribution and structure of picoeukaryotes.

METHODS



Sampling

Sampling was conducted with a 12 L Niskin bottles during a cruise (POEMM6) on board the R/V Hannibal at 7 stations from the coast to the offshore area in the Gulf of Gabès which is located south Tunisian coasts in the southern Ionian Sea. Temperature and salinity profiles were measured at each station with a conductivity, temperature and depth (CTD) probe (SBE 9, Sea-Bird Electronics, USA). Three to five depths were sampled at each station depending on bottom depth and real-time temperature profile provided by the CTD.

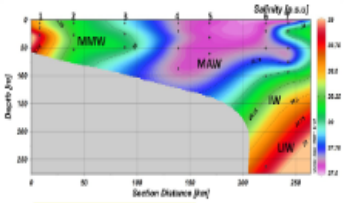
FISH associated with tyramide signal amplification (TSA-FISH)

To collect picoplanktonic cells, 250 ml of seawater was pre-filtered by gravity through 100-µm-pore-size mesh and then filtered under 200 mm Hg vacuum on firstly a GF/D membrane (whatman) and secondly a 0.45 µm polycarbonate filter (Sartorius). Cells fixation and TSA-FISH were carried out according to the protocols described by Biegala and Raimbault (2008) and Biegala et al. (2002; 2003), respectively. Two types of hybridization were carried out: an hybridization with a mix of EUK1209R (Giovanonni et *al*., 1988), CHLO02 (Simon et *al*., 2000) and NCHLO01 (Simon et *al*., 1995) probes in order to target all eukaryotic cells; and an hybridization with the CHLO02 probe targeting chlorophytes cells. The hybridized cells were counted using epifluorescence microscopy (Olympus BX41). The abundance of non-Chlorophyta cells is deduced from the counts of total picoeukaryotes and Chlorophyta cells.

Nutrient analyses

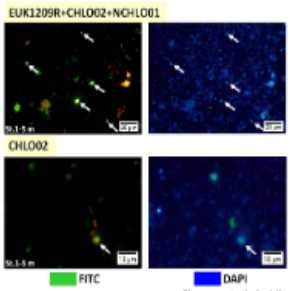
For the determination of nutrients (nitrite: NO2-, nitrate: NO3-, ammonium: NH4+ and Orthophosphate: PO43-), 250 ml of seawater were collected according to Hamdi et al. (2015). Nutrient analyses were performed with an automatic analyser type 3 (Bran & Luebbe) using standard methods (Tréguer and LeCorre, 1975).

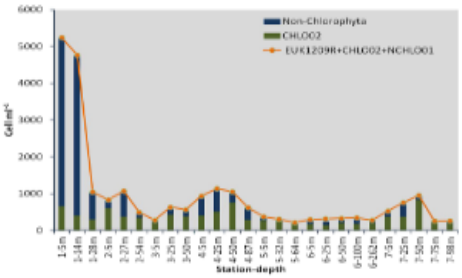
RESULTS

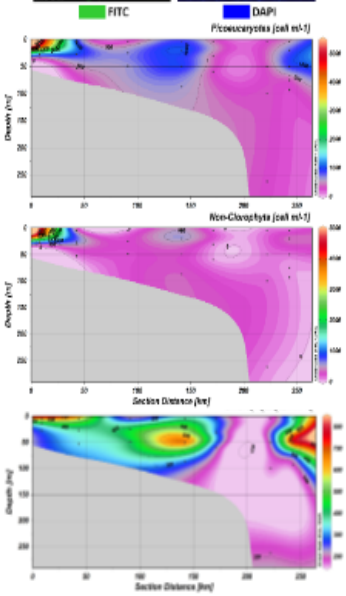


Four water masses were identified in the Gulf of Gabès:
(1) The Modified Atlantic Water (MAW) characterized by the lowest salinity and detected in the open-sea area at approximately 110 Km off the coast (2) The Levantine Intermediate Water (LIW) having the lowest temperature and the highest salinity values was observed at a depth exceeding 200 m of the station 6. The Ionian Water (IW) which is a transitional layer between the MAW and LIW masses, was located at depths between 100 and 200 m.

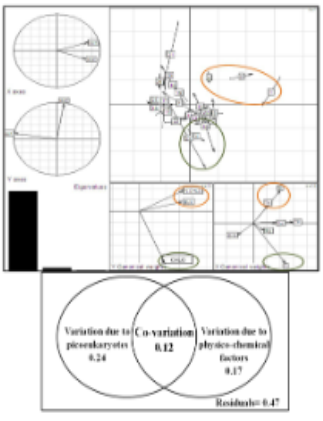
The Mediterranean Mixed Water (MMW) corresponded to the coastal waters characterized by the highest temperature values at depth up to almost 50 m.







- The mean abundance of total picoeukaryotes was 9.44 ± 12.59 102 cells mL-1, they were more abundant at coastal area where they reach 5.240 103 cells mL-1 at the surface of station 1.
- In surface waters of the Gulf of Gabès, the mean abundance of total picoeukaryotes was 1.212 103 cells mL-1.
- Non-Chlorophyta cells dominated the picoeukaryotic community, their abundance ranged from 23.36 cells mL-1 (St. 5, 64 m) to 4.058 103 cells mL-1 (St. 1, surface) with a mean value of 5.61 ± 11.70 102 cells mL-1.
- Chlorophytes were less abundant with an average value of 3.55 ± 1.84 102 cells mL-1 whereas in the offshore area(Stations 2-7) their densities were higher than those of non-Chlorophyta cells
- The densities of non-Chlorophyta cells mostly increase at 25 m depth and maximum densities of chlorophytes were observed at 50 m depths in the open sea area (Stations 4 and 7).
- The surface abundance of Chlorophyta and Non-Chlorophyta cells exhibited a decreasing gradient towards the offshore area.



- The overall model explained 53% of the total variation (permutation test, p=0.04,1000 replicates) which was due to the variability of physico-chemical facors (17%) and picoeukaryotic abundance (24%).
- The co-inertia plot illustrated close links at station 1 between total picoeukaryotes, non-Chlorophyta cells, salinity, total phosphorus, total nitrogen and orthophosphates concentrations.
- At the oceanic region, close links were detected between chlorophytes and temperature.

CONCLUSION

In the Gulf of Gabès:

- Chlorophyta and non-Chlorophyta Divisions contributed respectively to 38.5 and 61.5 % of total picoeukaryotes.
- The picoeukaryotic abundance is typical of an oligotrophic environment
- The spatial distribution of picoeukaryotic community seems to be affected by physical parameters (salinity and temperature) and nutrient availability (organic nitrogen and total phosphorus) of the water column, but no clear relation with the identified water masses was detected.

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