

Continuous and **High Resolution** Observation of the **ME**diterranean Sea



## 7<sup>th</sup> FerryBox Workshop

**Coupling FerryBox and automated flow cytometer: a new approach for studying biogeochemical processes and CO<sub>2</sub> system variability in relation to phytoplankton community structure**

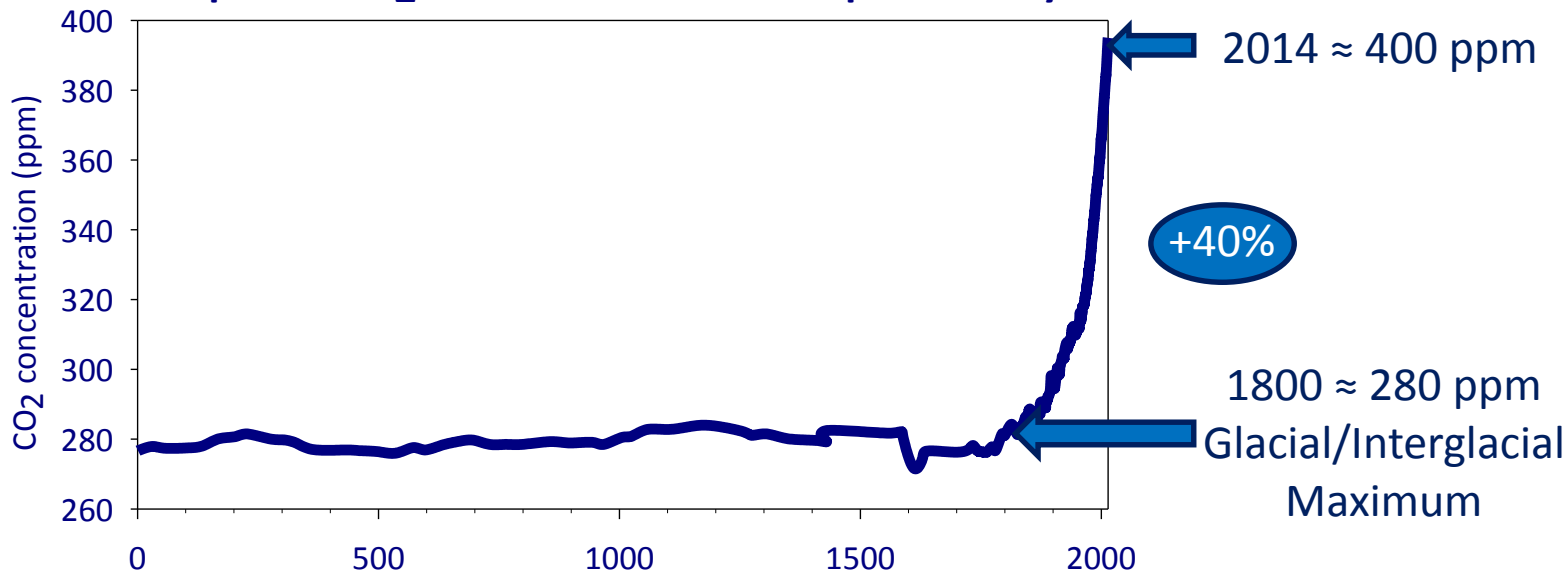
*Heraklion, 6 and 7 April 2016*

**Pierre Marrec<sup>1</sup>, Cherif Sammari<sup>2</sup>, Nagib Bhairy<sup>1</sup>, Mathilde Dugenne<sup>1</sup>, Gérald Grégori<sup>1</sup>, Sana Ben Ismail<sup>2</sup>, Soumaya Lahbib<sup>1</sup> and Melilotus Thyssen<sup>1</sup>**

<sup>1</sup> CNRS/INSU, IRD, Mediterranean Institute of Oceanography, Aix Marseille Université, Marseille, France

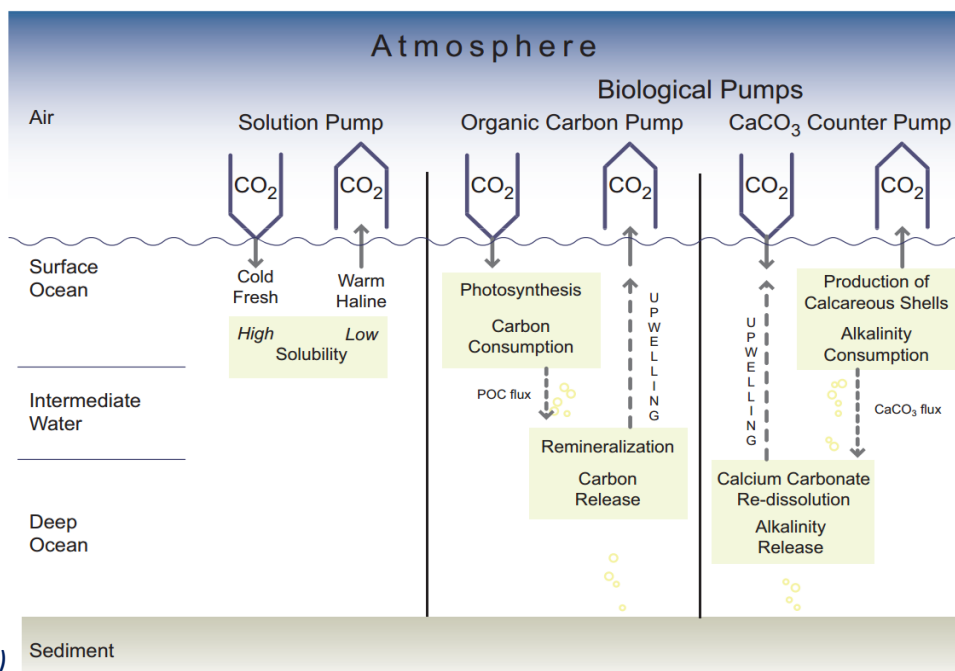
<sup>2</sup> INSTM, National Institute of Marine Sciences and Technologies, Salammbô, Tunisia

## Atmospheric CO<sub>2</sub> concentration for the past 2000 years



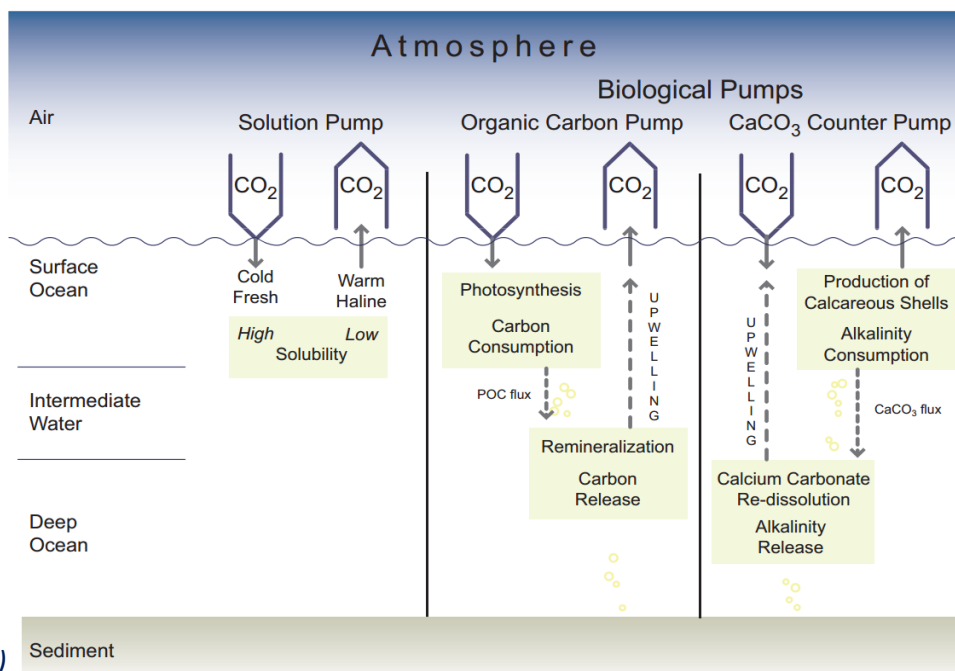
- Since the **industrial revolution**: atmospheric CO<sub>2</sub> has **increased by ≈40%**.
- **Highest atmospheric CO<sub>2</sub> concentration** over at least the last **800 000 years**.
- Essentially due to **anthropogenic activities**.
- **Increase of atmospheric GHG concentration** (CO<sub>2</sub>, as well as CH<sub>4</sub> and N<sub>2</sub>O) is “**extremely likely**” (95-100% probability) **the main driver of climate change** (IPCC, 2013).

- The world ocean plays a **fundamental role** in the **exchange of CO<sub>2</sub>** with the **atmosphere** and constitutes an **important sink** for atmospheric CO<sub>2</sub>.
- The ocean stores nearly **1/3 of anthropogenic carbon** released to the atmosphere (Sabine et al., 2004).
- **Physical** and **biological** processes control the **ocean carbon cycle** with the **solubility pump** and the **biological pumps**.





Essential to improve our understanding of the dynamics of the CO<sub>2</sub> system and to quantify air-sea CO<sub>2</sub> fluxes for global carbon cycle and climate studies.





Seawater partial pressure of CO<sub>2</sub> (pCO<sub>2</sub>) variability

Thermodynamic processes:  
- Solubility

Seawater temperature/salinity

Non-thermodynamic processes:  
- *Biological processes*  
- *Dissolution/formation of CaCO<sub>3</sub>*  
- *Lateral advection*  
- *Horizontal mixing*  
- *Upwelling*  
...

Structural phytoplankton heterogeneity  
Presence of calcifying organisms  
3D oceanic circulation (observations and models)



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**Well constrained**

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...



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3D oceanic circulation (observations and models)

**Individual process contributions difficult to assess**



Seawater partial pressure of CO<sub>2</sub> (pCO<sub>2</sub>) variability

Thermodynamic processes:  
- Solubility

Seawater temperature/salinity

Non-thermodynamic processes:

- Biological processes
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...

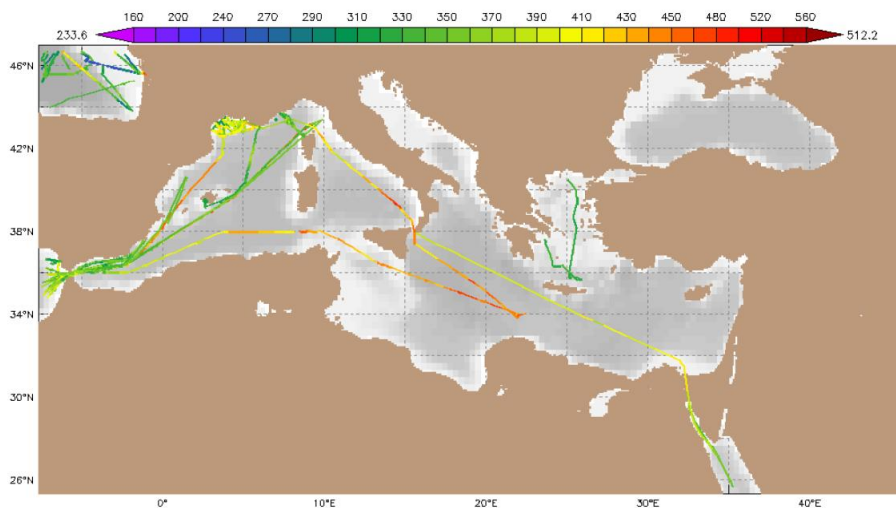
Structural phytoplankton heterogeneity  
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3D oceanic circulation (observations and models)

**CHROME project**



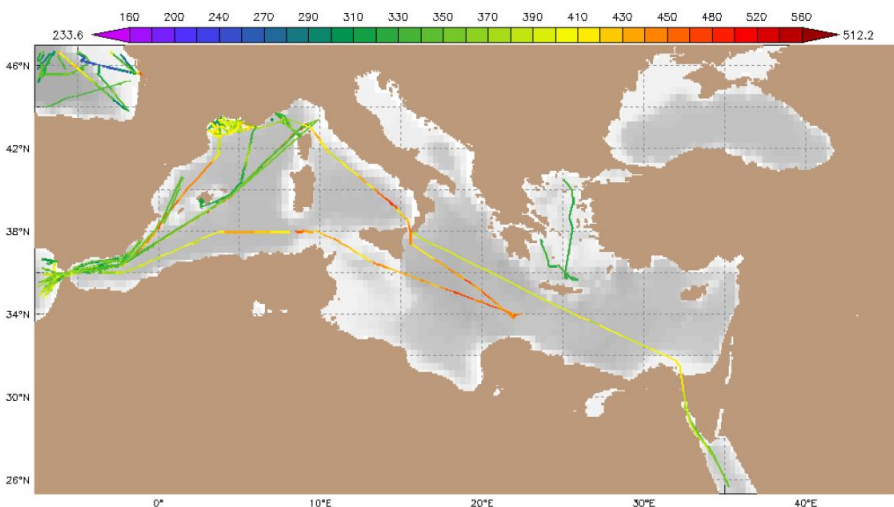
# Mediterranean Sea

- Only few pCO<sub>2</sub> data (SOCAT, LDEO databases)



# Mediterranean Sea

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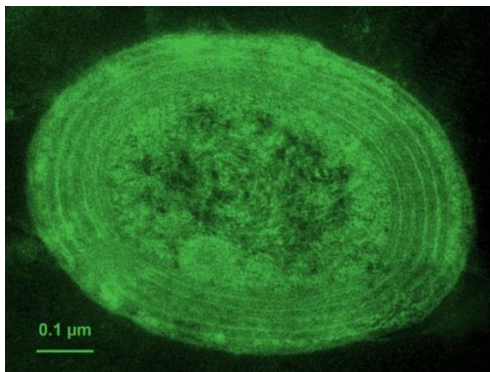


- Few phytoplankton community dataset resolving the submeso- & mesoscale :

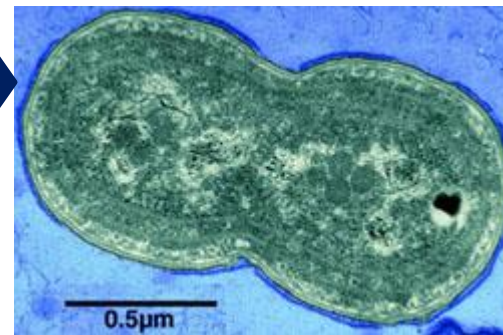
- space scales of 1-10 km and time scales of 1-10 days
- space scales of 10-100 km and time scales of 10-100 days

## Mediterranean Sea

- Oligotrophic ecosystem
- Dominated by small phytoplankton (picoplankton):
  - Cyanobacteria : *Prochlorococcus* & *Synechococcus* (0,6 – 1,2  $\mu\text{m}$ )
  - PicoEukaryotes (< 2  $\mu\text{m}$ )
- Importance of picoplankton in oligotrophic areas:
  - major primary producers
  - most abundant phytoplankton organisms on earth



*Synechococcus*  
Photo: John Waterbury,  
Woods Hole Oceanographic Institute

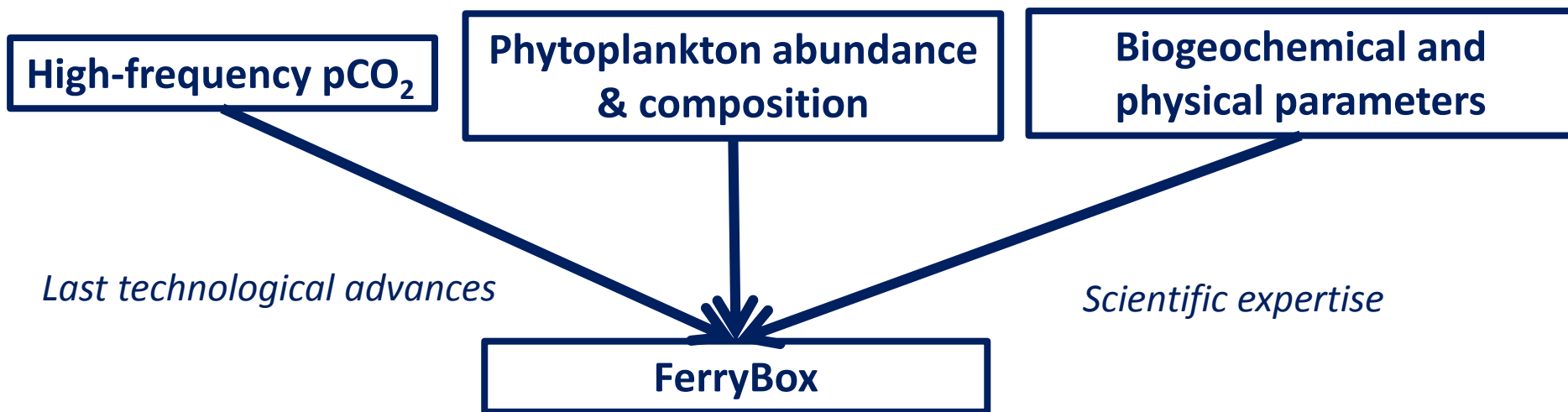


*Prochlorococcus*  
Biodiversity project – Austin Chiang



# CHROME project

Unravel the role of phytoplankton community structure on biogeochemical variability and CO<sub>2</sub> system dynamics at high spatio-temporal resolution





# CHROME project

High-frequency pCO<sub>2</sub>

Phytoplankton abundance & composition

Biogeochemical and physical parameters

Last technological advances

Scientific expertise

FerryBox

## PhD Thesis

Yann Bozec & Pascal Morin



Contents lists available at SciVerse ScienceDirect

**Marine Chemistry**  
journal homepage: [www.elsevier.com/locate/marchem](http://www.elsevier.com/locate/marchem)

Seasonal and latitudinal variability of the CO<sub>2</sub> system in the western English Channel based on Voluntary Observing Ship (VOS) measurements

P. Marrec<sup>a,\*</sup>, T. Cariou, E. Collin, A. Durand, M. Latimier, E. Macé, P. Morin, S. Raimund<sup>1</sup>, M. Vernet, Y. Bozec

CNRS, UMR 7144, Equipe Chimie Marine, Station Biologique de Roscoff, Place Georges Teissier, 29680 Roscoff, France  
UPMC Univ. Paris 06, UMR 7144, Adaptation et Diversité en Milieu Marin, SDR, 29680 Roscoff, France

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Contents lists available at ScienceDirect

**Journal of Marine Systems**  
journal homepage: [www.elsevier.com/locate/jmarsys](http://www.elsevier.com/locate/jmarsys)

Spatio-temporal dynamics of biogeochemical processes and air-sea CO<sub>2</sub> fluxes in the Western English Channel based on two years of FerryBox deployment

P. Marrec<sup>a,\*</sup>, T. Cariou, M. Latimier, E. Macé, P. Morin, M. Vernet, Y. Bozec

CNRS, UMR 7144, Equipe Chimie Marine, Station Biologique de Roscoff, Place Georges Teissier, 29680 Roscoff, France  
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Biogeosciences, 12, 5371–5391, 2015  
[www.biogeosciences.net/12/5371/2015/](http://www.biogeosciences.net/12/5371/2015/)  
doi:10.5194/bg-12-5371-2015  
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**Dynamics of air-sea CO<sub>2</sub> fluxes in the northwestern European shelf based on voluntary observing ship and satellite observations**

P. Marrec<sup>1,2</sup>, T. Cariou<sup>1,2</sup>, E. Macé<sup>1,2</sup>, P. Morin<sup>1,2</sup>, L. A. Salt<sup>1,2</sup>, M. Vernet<sup>1,2</sup>, B. Taylor<sup>3</sup>, K. Paxman<sup>3</sup>, and Y. Bozec<sup>1,2</sup>

<sup>1</sup>CNRS, UMR7144, Equipe Chimie Marine, Station Biologique de Roscoff, Place Georges Teissier, 29680 Roscoff, France  
<sup>2</sup>Sorbonne Universités, UPMC, Univ. Paris 06, UMR7144, Adaptation et Diversité en Milieu Marin, Station Biologique de Roscoff, 29680 Roscoff, France  
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Accepted: 25 August 2015 – Published: 18 September 2015



CNRS UPMC  
Station Biologique  
Roscoff



# CHROME project

High-frequency pCO<sub>2</sub>

Phytoplankton abundance & composition

Biogeochemical and physical parameters

Last technological advances

Scientific expertise

FerryBox



Melilotus Thyssen & Gérald Grégori

frontiers in MICROBIOLOGY

ORIGINAL RESEARCH ARTICLE published: 15 September 2014 doi: 10.3389/fmicb.2014.00485

Consequence of a sudden wind event on the dynamics of a coastal phytoplankton community: an insight into specific population growth rates using a single cell high frequency approach

Mathilde Dugenne\*, Melilotus Thyssen, David Nerini, Claude Mante, Jean-Christophe Nicole Garcia, Fabrice Garcia and Gérald J. Grégori

Aix Marseille Université, Université de Toulon, CNRS/INSU, IRD, Mediterranean Institute of Oceanography, UM 110, Marseille, France

frontiers in MICROBIOLOGY

ORIGINAL RESEARCH ARTICLE published: 12 August 2014 doi: 10.3389/fmicb.2014.00387

Onset of the spring bloom in the northwestern Mediterranean Sea: influence of environmental pulse events on the *in situ* hourly-scale dynamics of the phytoplankton community structure

Melilotus Thyssen<sup>1\*</sup>, Gérald J. Grégori<sup>1</sup>, Jean-Michel Grisoni<sup>2,3</sup>, Maria Luiza Pedrotti<sup>2,3</sup>, Laure Mousseau<sup>2,3</sup>, Luis F. Artigas<sup>4</sup>, Sophie Marro<sup>2,3</sup>, Nicole Garcia<sup>1</sup>, Ornella Passafiume<sup>2,3</sup> and Michel J. Dents<sup>1</sup>

<sup>1</sup> CNRS/INSU, IRD, Mediterranean Institute of Oceanography, Aix Marseille Université, Marseille, France  
<sup>2</sup> Sorbonne Universités, UPMC Univ. Paris 06, UMR 7093, LOV, Observatoire Océanologique, Villefranche-sur-Mer, France  
<sup>3</sup> CNRS, UMR 7093, LOV, Observatoire Océanologique, Villefranche-sur-Mer, France  
<sup>4</sup> Laboratoire d'Océanologie et Géosciences, Univ. du Littoral Côte d'Opale, CNRS, UMR8157, Wimereux, France

<http://www.frontiersin.org/abstract>



# INSTM FerryBox system

1 Marseille ↔ Tunis  
& 1 Genova ↔ Tunis  
crossings per week.

Continuous measurements:

- Temperature
- Salinity
- Dissolved Oxygen
- Fluorescence
- Turbidity

Our sensors CHROME/MIO

- pCO<sub>2</sub>
- pH
- Automated Flow Cytometer (FC)



C/F Carthage



41

JENA ENGINEERING

FerryBox installation: February 2016  
 pCO<sub>2</sub>, pH and FC implementation: March 2016  
 But the system is not entirely operational yet

## MIO /CHROME sensors



SubCtech OceanPack MK2:

- Flat-Membrane-Equilibrator
- LI-COR LI-840x inside
- Auto-zeroing calibration
- Span gas calibration



## MIO / CHROME sensors



### SubCtech OceanPack MK2:

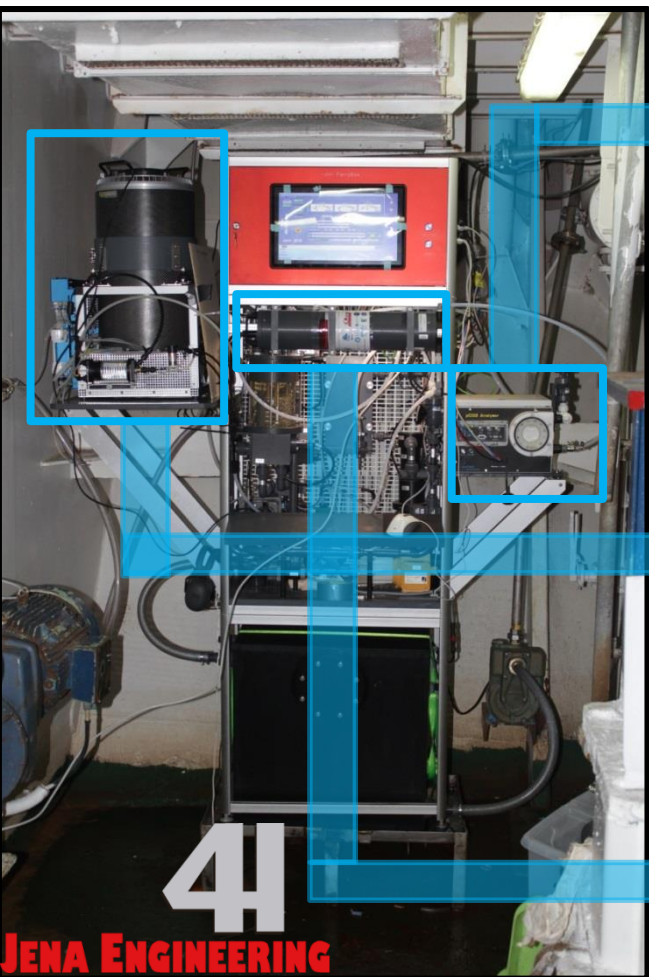
- Flat-Membrane-Equilibrator
- LI-COR LI-840x inside
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- Span gas calibration



### CytoBuoy CytoSense Flow Cytometer

- Single cell analysis
- Automated and remotely controlled
- Image in flow acquisition
- Run with embedded computer

# MIO / CHROME sensors



**JENA ENGINEERING**



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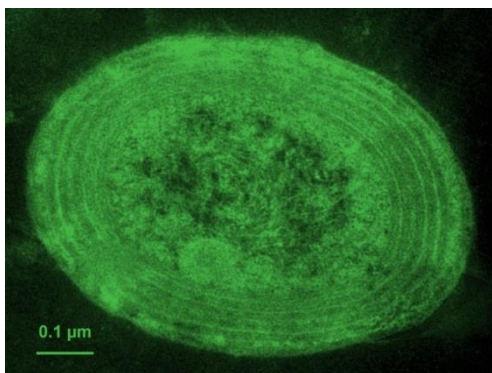
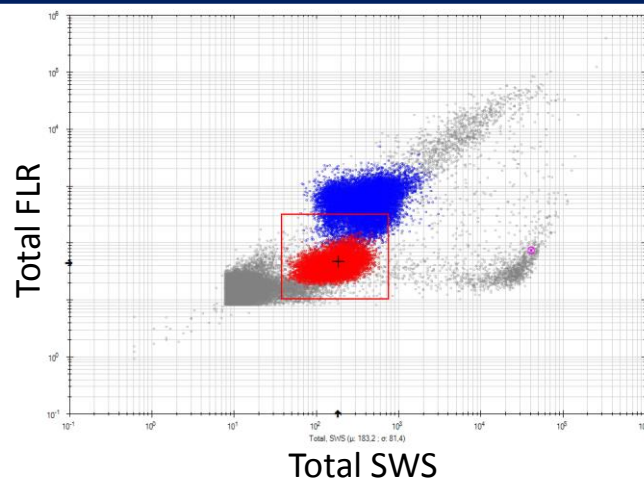
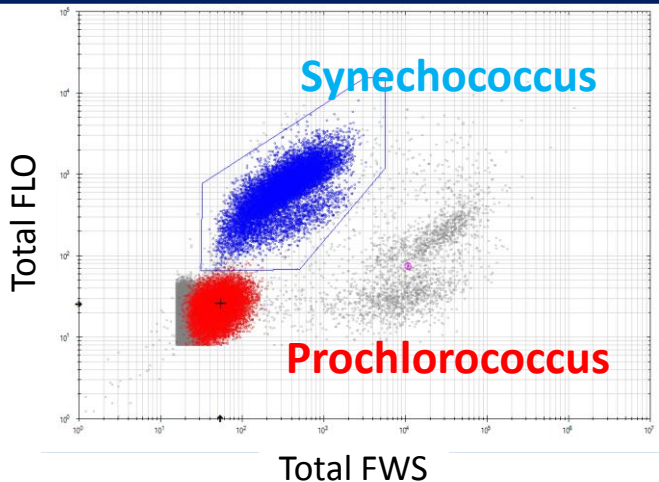


Satlantic SeaFET:

- ion sensitive field effect transistor (ISFET)
- Our first deployment

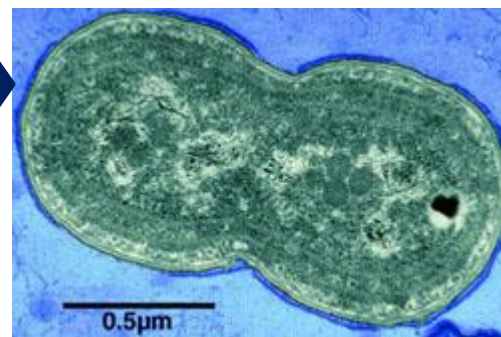


Thanks to recent advances, we can now observe prochlorococcus with the CytoSense, in addition to larger phytoplankton cells.

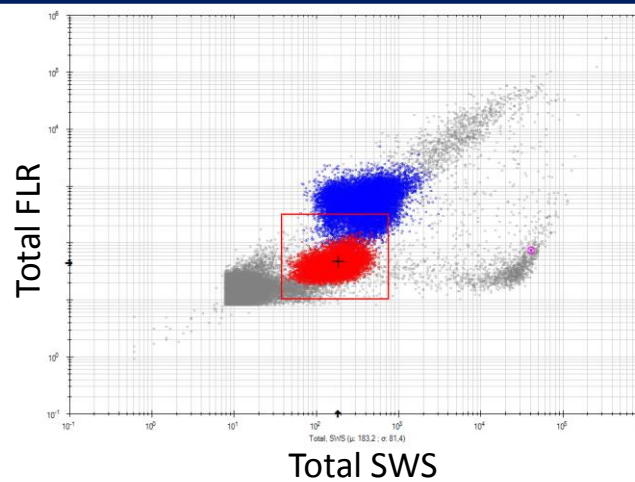
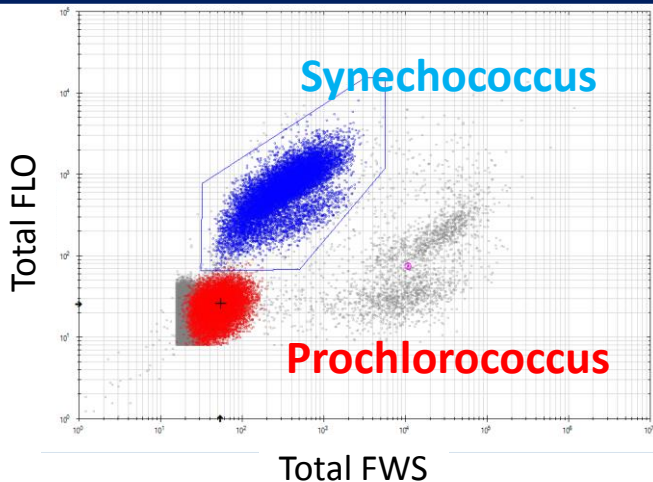


Synechococcus  
*Photo: John Waterbury,  
Woods Hole Oceanographic Institute*

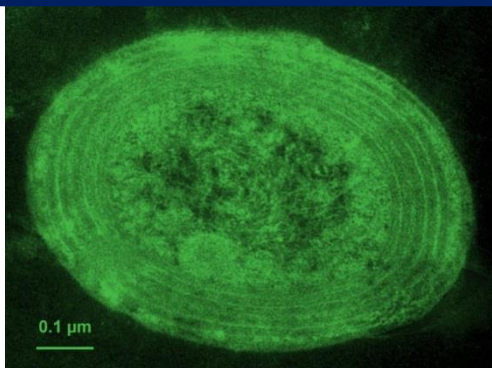
Prochlorococcus  
*Biodiversity project – Austin Chiang*



Thanks to recent advances, we can now observe both with the CytoSense, in addition to larger phytoplankton cells.

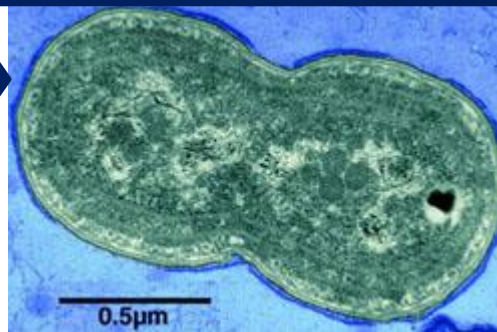


It will allow us to better understand the spatio-temporal dynamics of these major photosynthetic contributors in relation to air-sea CO<sub>2</sub> fluxes.



Synechococcus  
*Photo: John Waterbury, Woods Hole Oceanographic Institute*

Prochlorococcus  
*Biodiversity project – Austin Chiang*

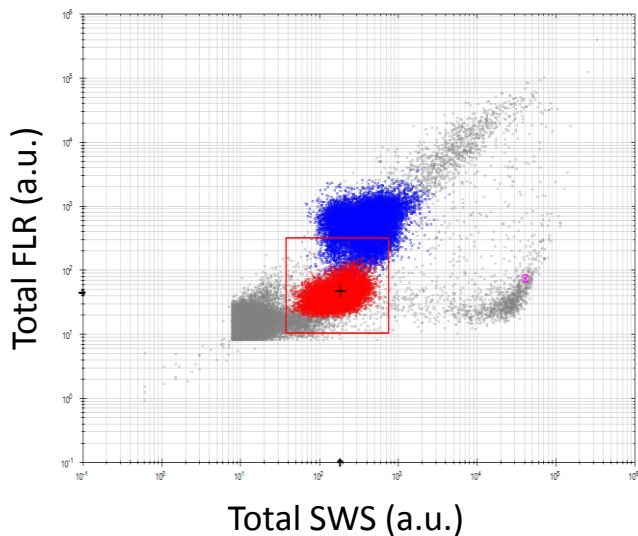




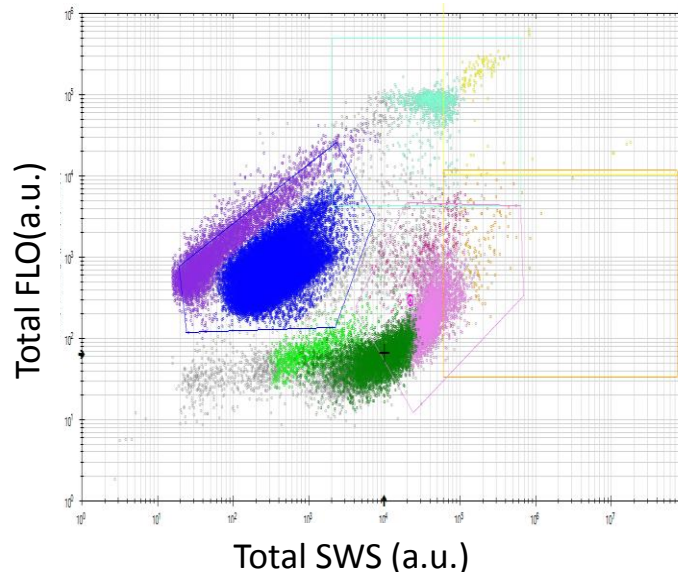
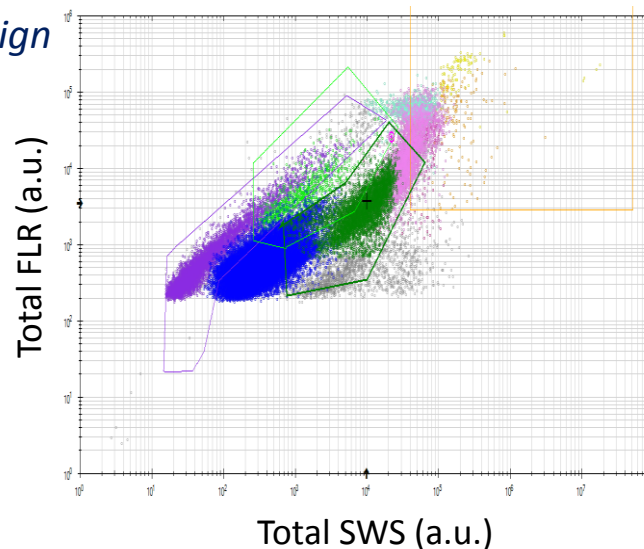
FLR5

# Example of clustering

FLR30



*OSCAHR campaign*



10 phytoplankton groups defined:

Prochlorococcus

Synechococcus

Picoplankton & Picoplankton High FLR

Cryptophytes like

Nanoplankton 1 & Nanoplankton High FLO

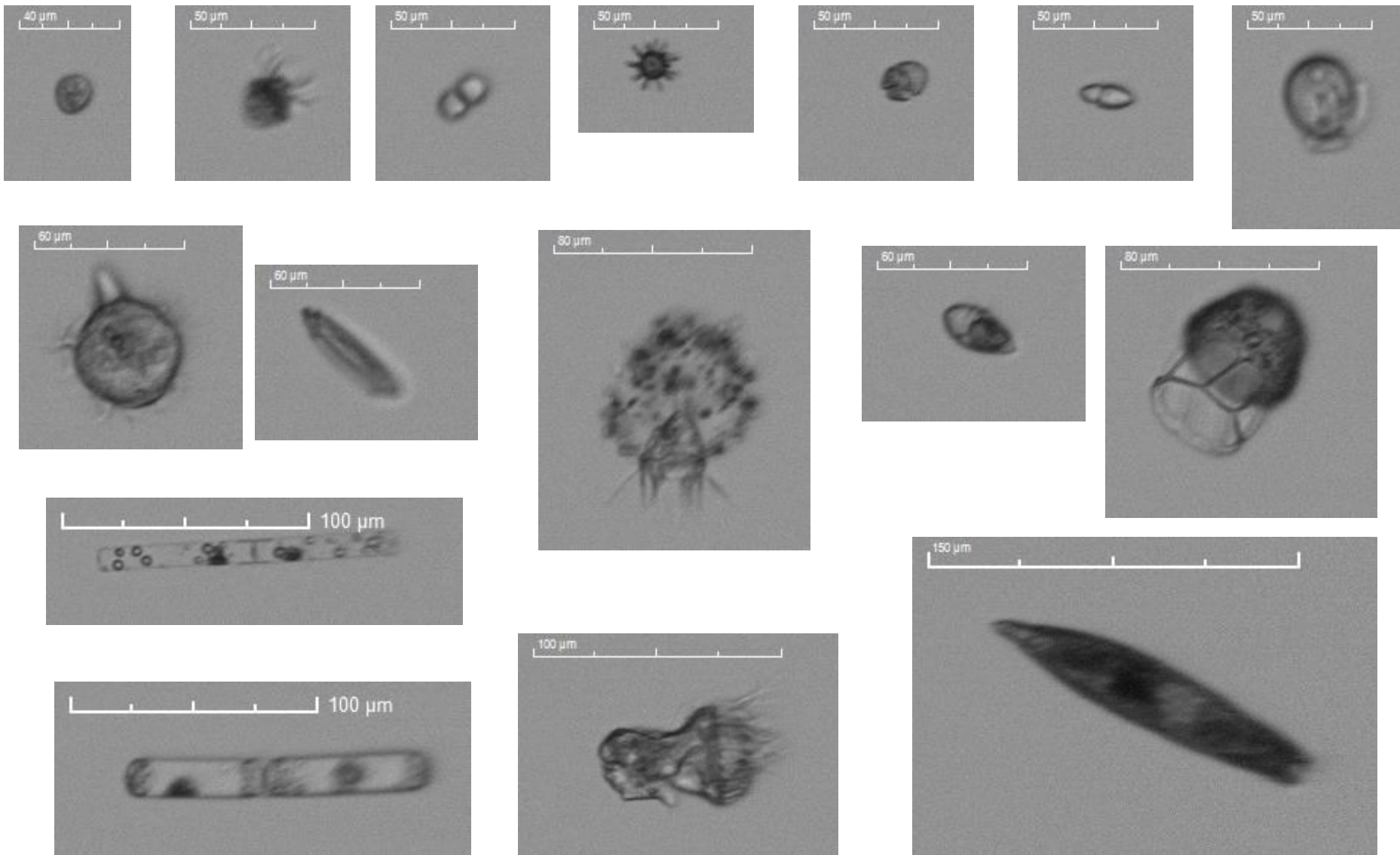
Microplankton & Microplankton High FLO

Unidentified (for the moment)

Calibration with beads (1-10  $\mu\text{m}$ ) to class phytoplankton clusters according to their size, and use of pictures for microphytoplankton ( $>20 \mu\text{m}$ ).

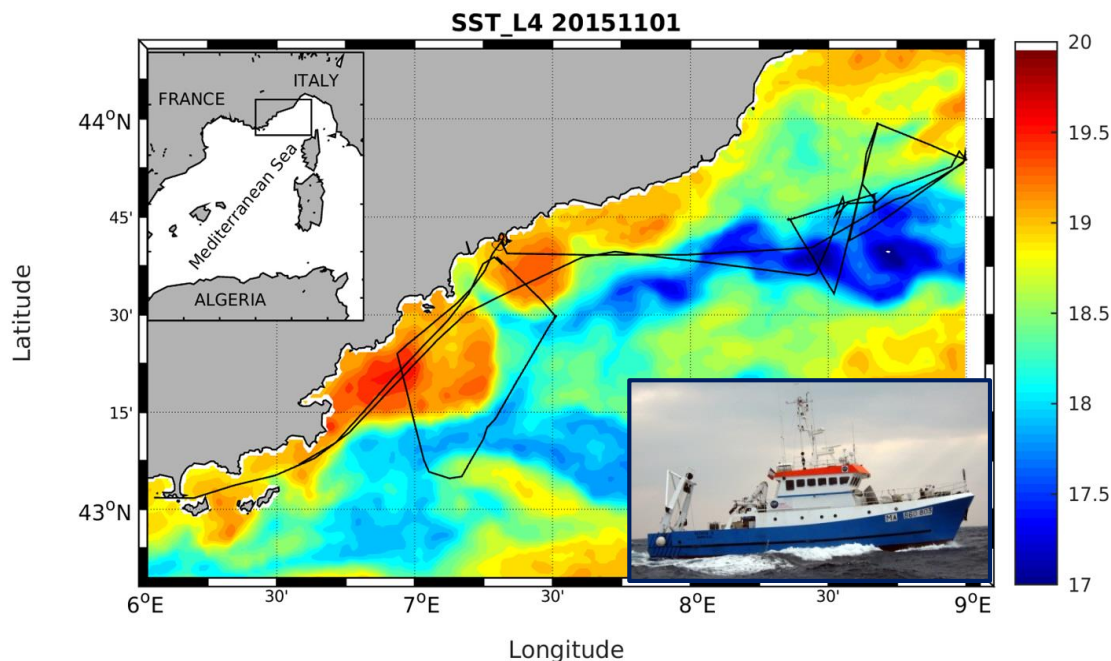


## Image in flow (IIF) acquisition (>20 μM)



## Pre-FerryBox automated flow cytometer deployment: OSCAHR

- Observing Submesoscale Coupling At Hight Resolution, *Pls Andrea Doglioli & Gérald Grégori*, 2015-10-29 to 2015-11-06 onboard the RV Tethys II.
- Last generation Cytosense flow cytometer with IIF deployment with a -4H-Pocket FerryBox (SST, SSS, O<sub>2</sub>, FLUO) and a subCtech pCO<sub>2</sub> sensor.
- Similar configuration as in the **CHROME** project.

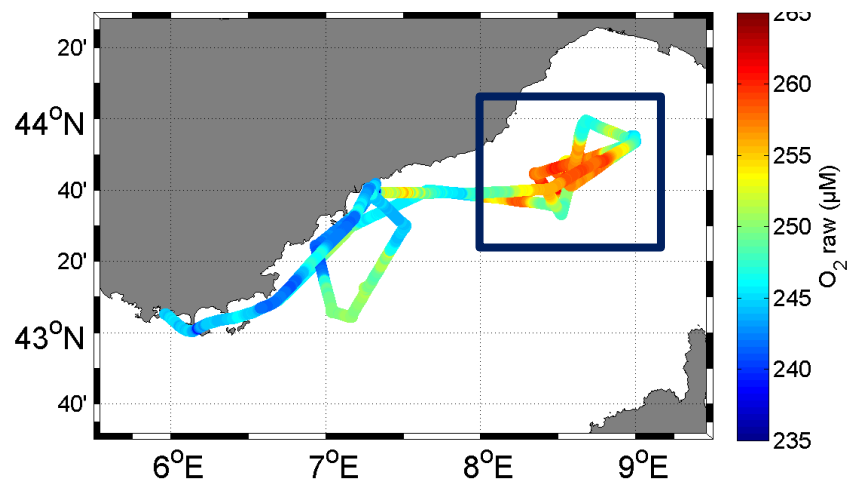
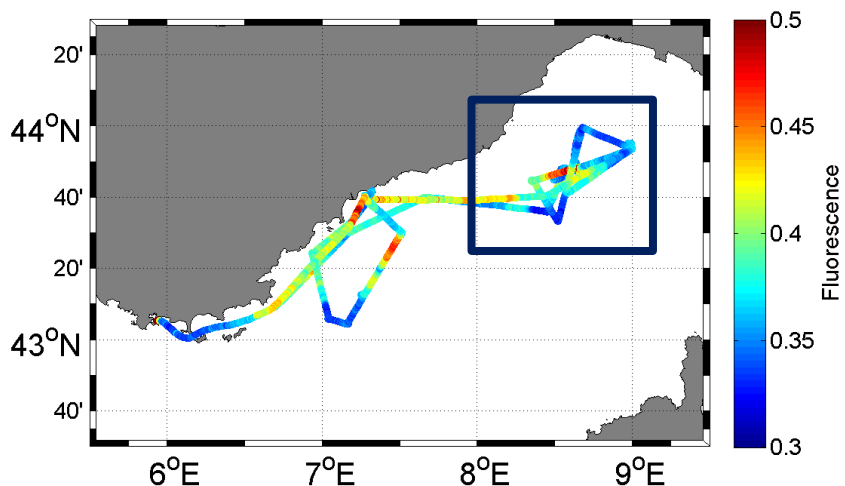
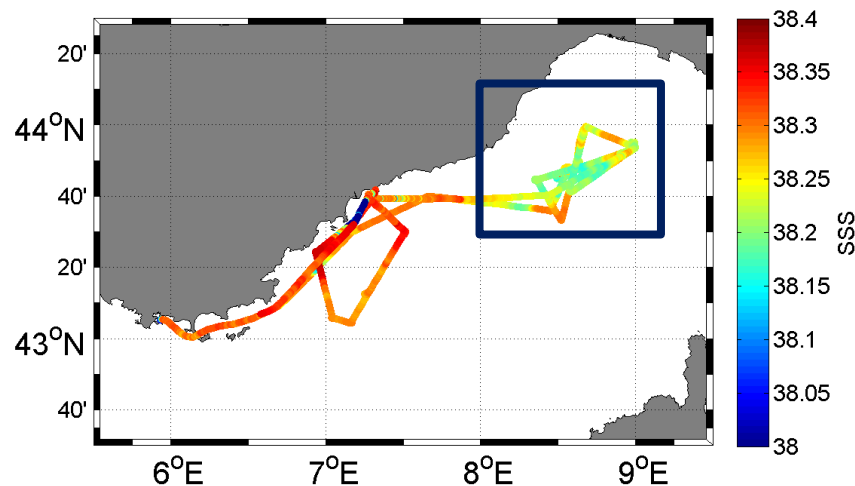
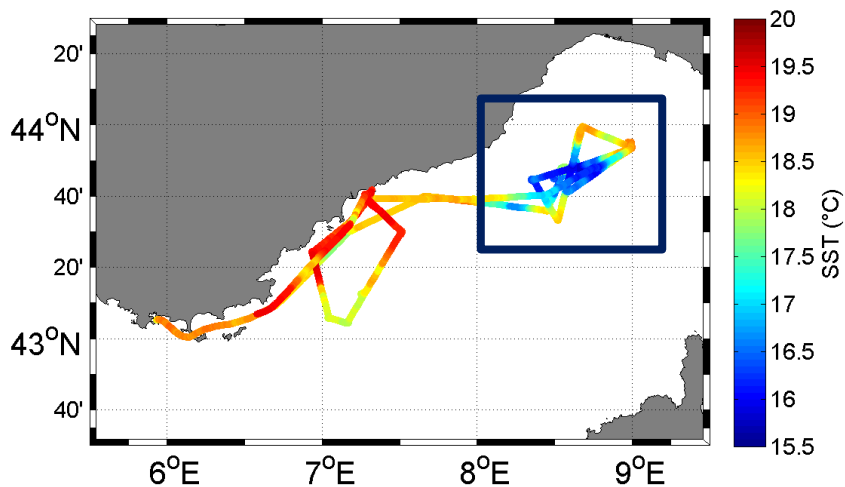




Continuous and High Resolution Observation of the **ME**diterranean Sea



7<sup>th</sup> FerryBox Workshop



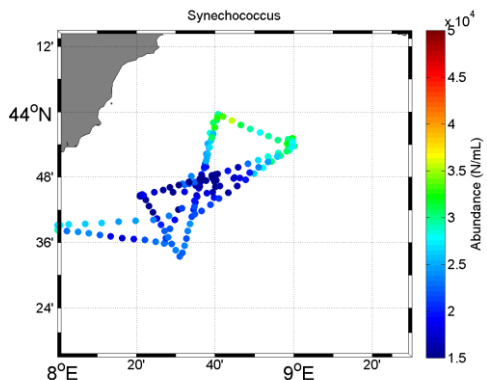
Unfortunately no pCO<sub>2</sub> data due to technical issues.



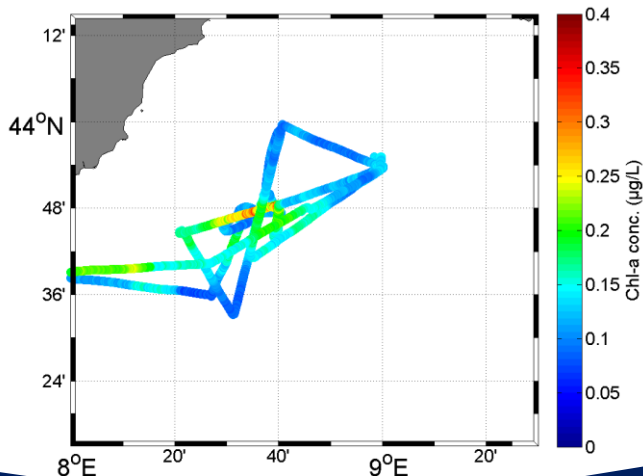


7<sup>th</sup> FerryBox Workshop

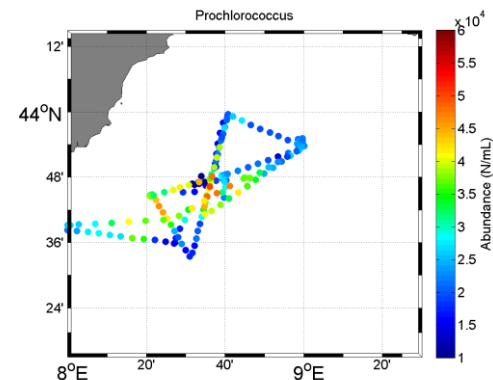
### Synechococcus



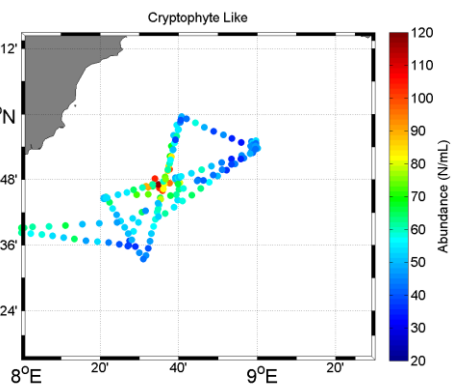
### Chl-a conc. ( $\mu\text{g L}^{-1}$ )



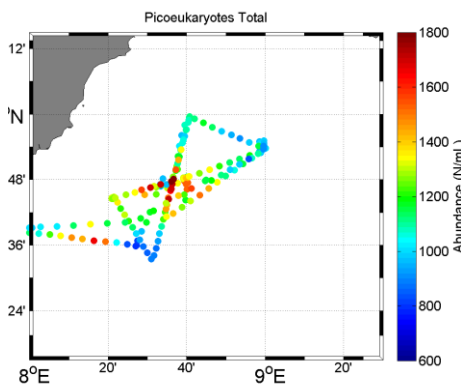
### Prochlorococcus



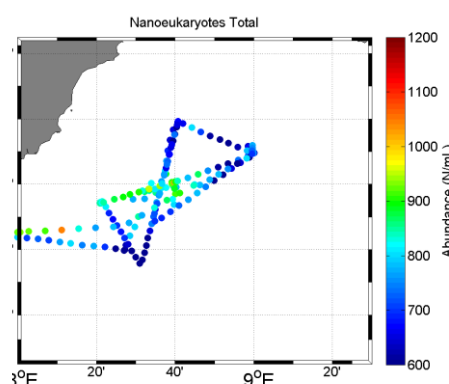
Measurements every 20 minutes



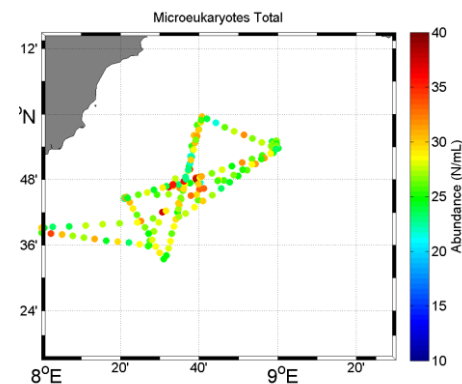
Cryptophytes like



Pico-Eukaryotes



Nano-Eukaryotes



Microphytoplankton



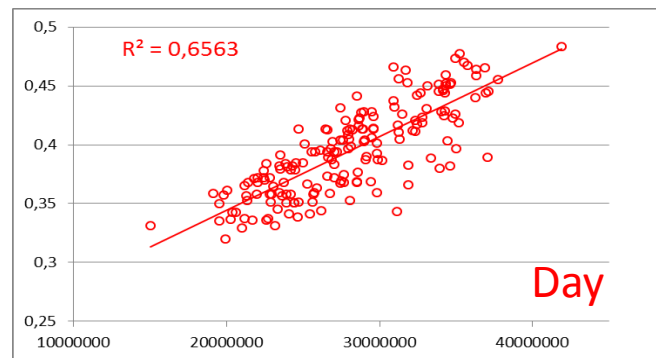
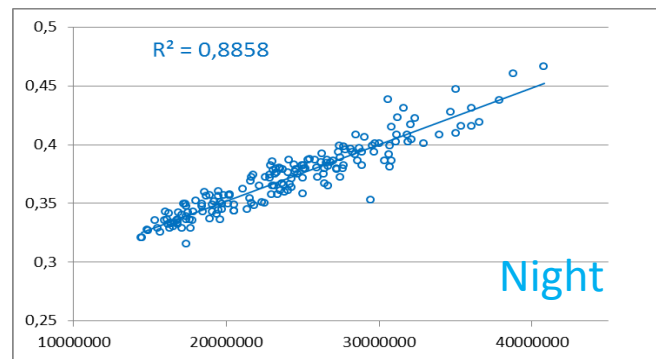
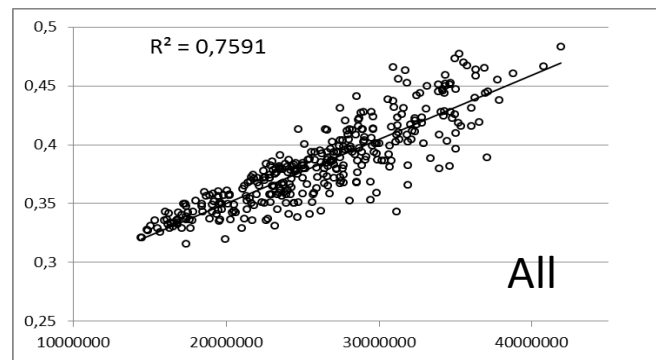
Red fluorescence (FLR): 1 of the optical parameter recorded by the cytometer (FLO, SWS, FWS).

Strong correlation between:

- The sum of the total red fluorescence recorded for all the clusters
- And the fluorescence recorded by the research vessel fluorimeter
- Quenching during daytime

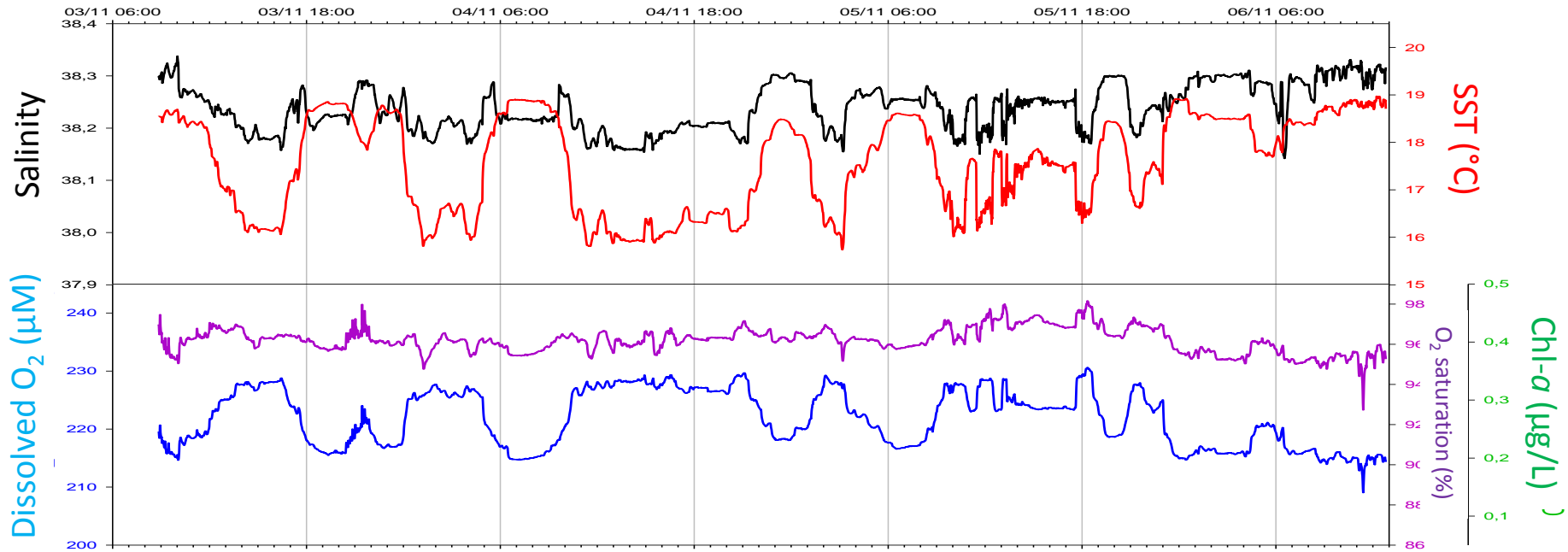
FWS: forward scatter, SWS: sideward scatter,  
FLR: red fluorescence, FLO: orange fluorescence

Fluorescence from the Fluorimeter (a.u.)

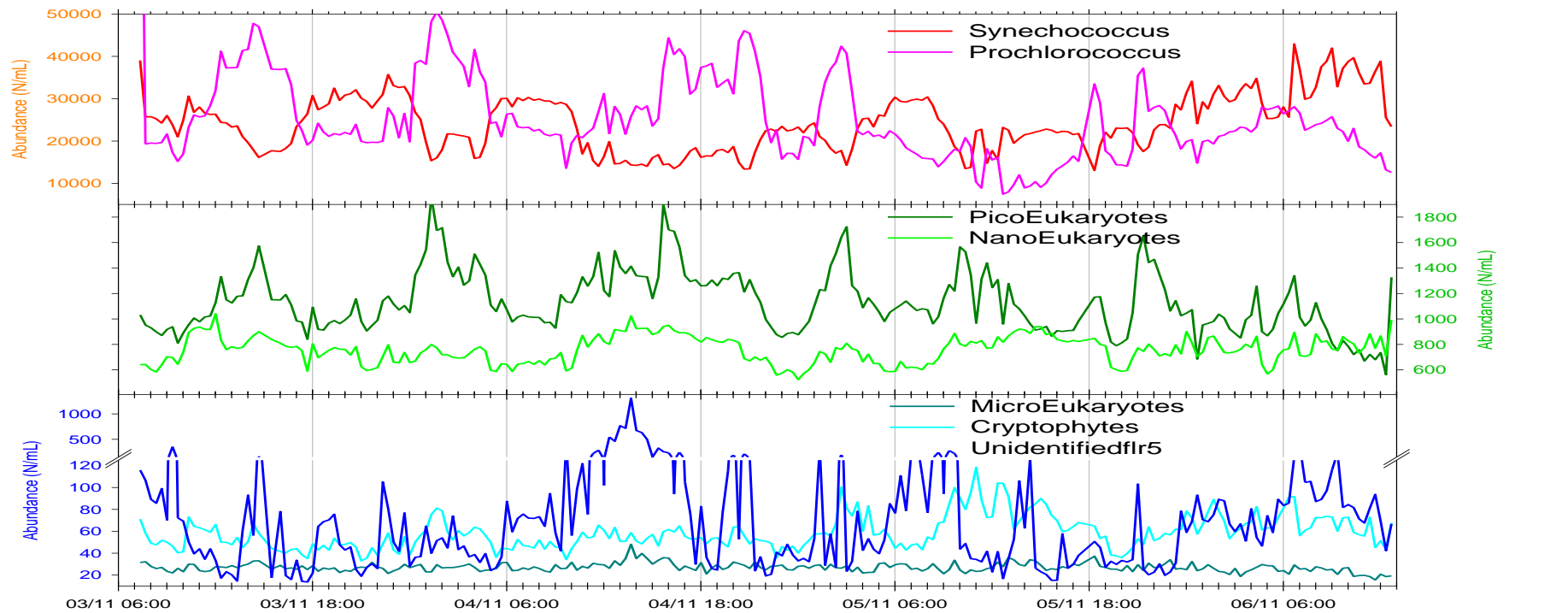


Total Fluorescence from the Cytometer (a.u.)

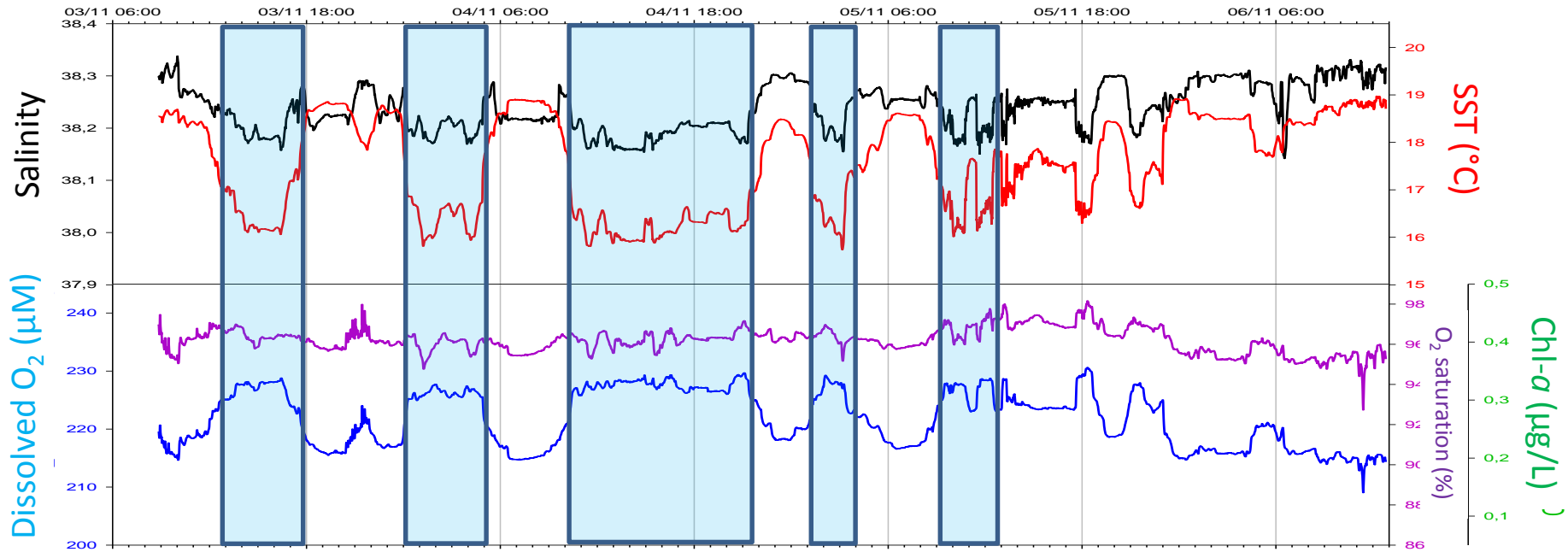
Leg2



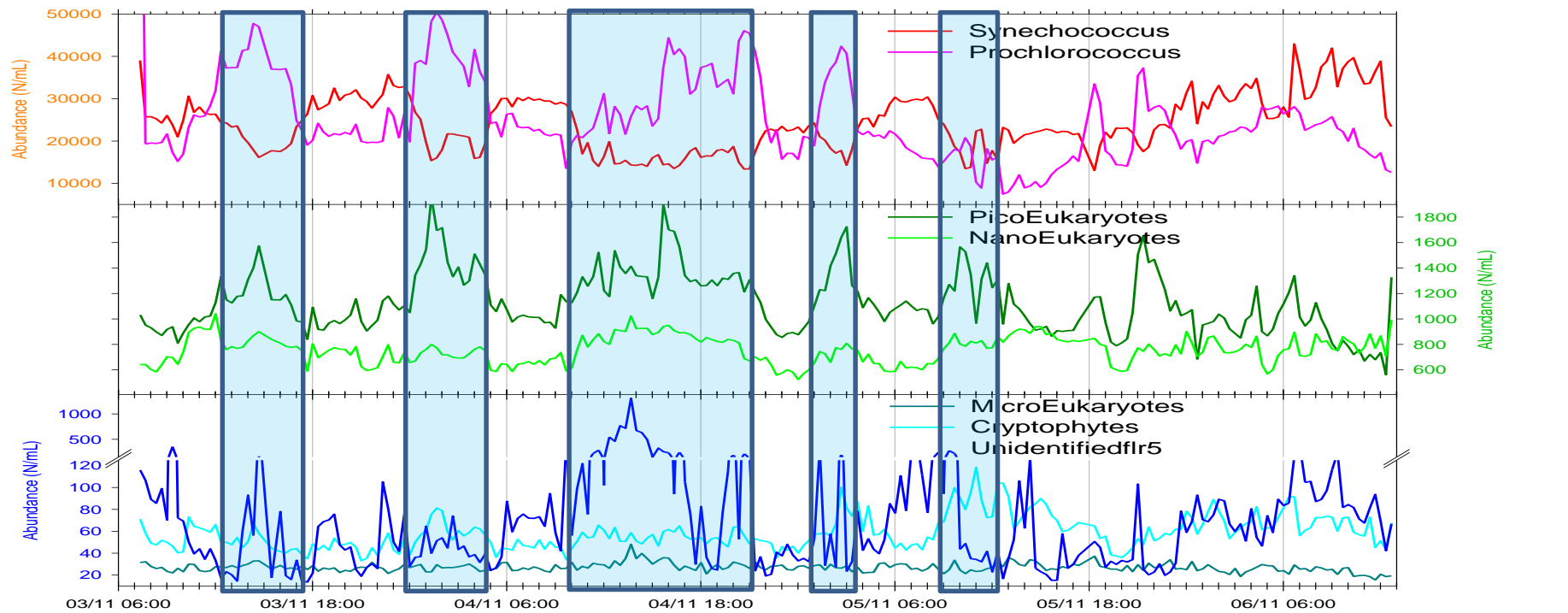
Abundance (N mL<sup>-1</sup>)

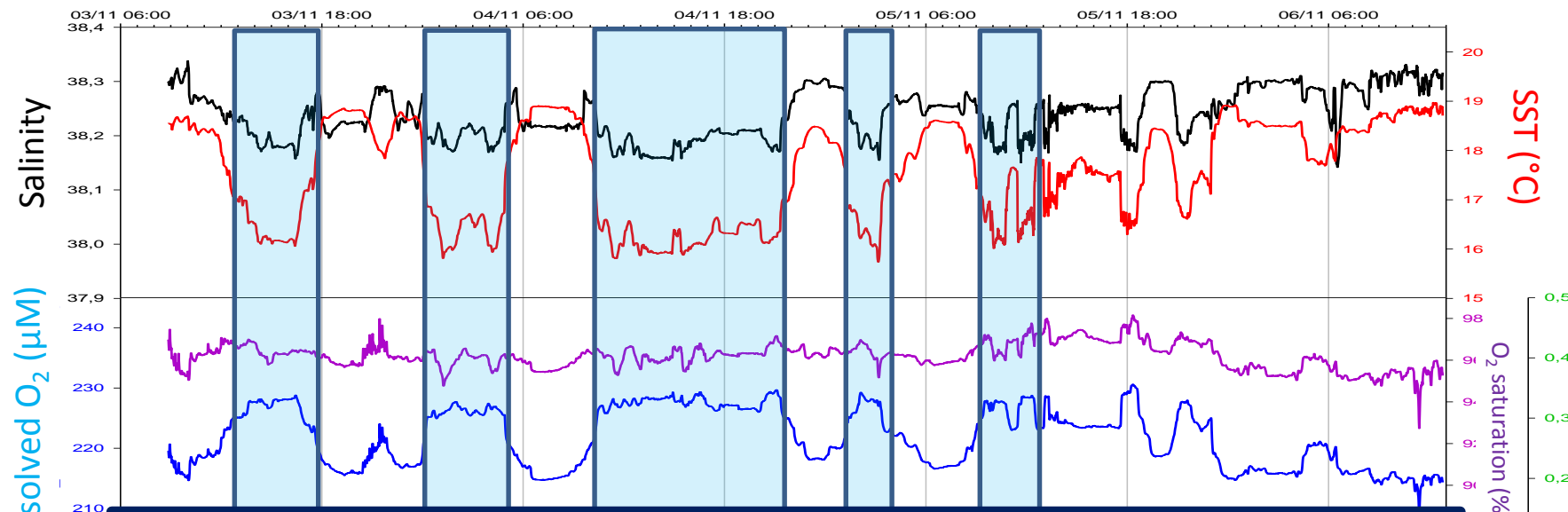


# Leg2

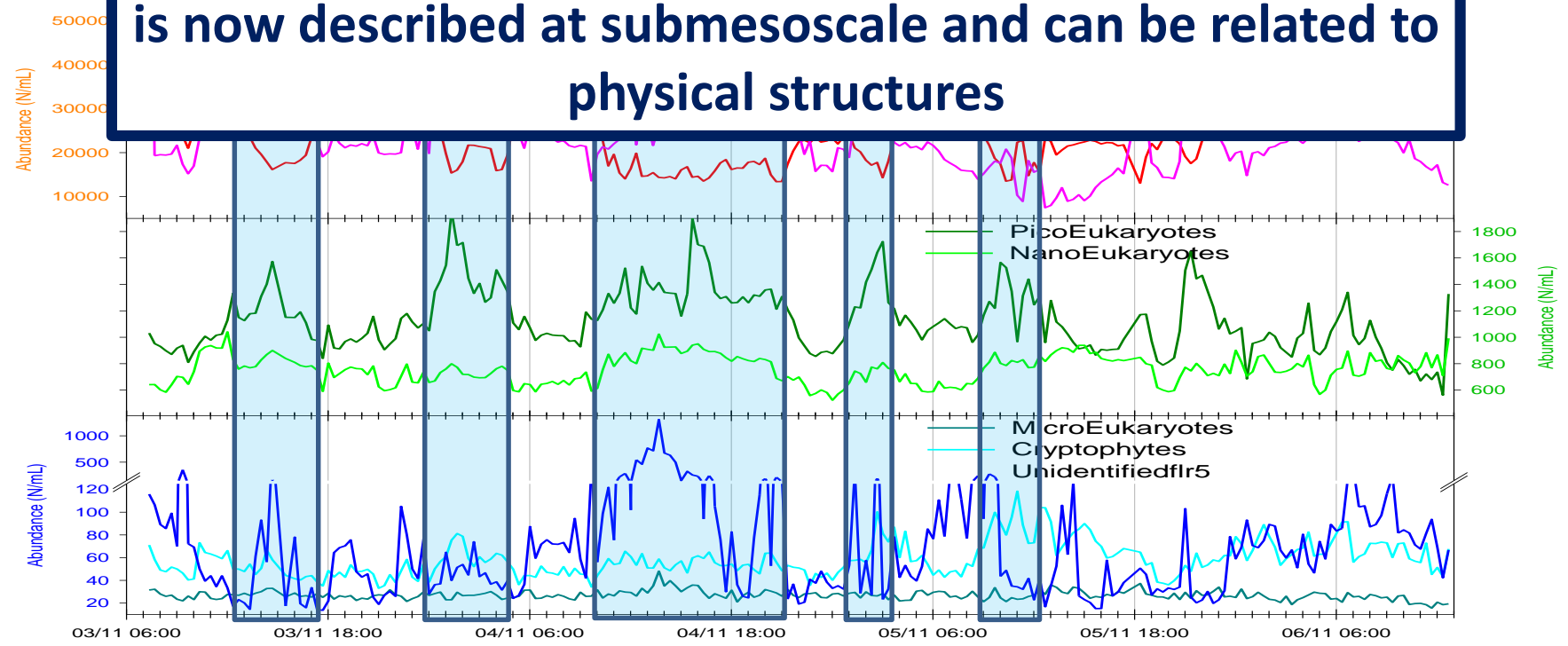


# Abundance (N mL<sup>-1</sup>)





**Phytoplankton functional and structural heterogeneity is now described at submesoscale and can be related to physical structures**

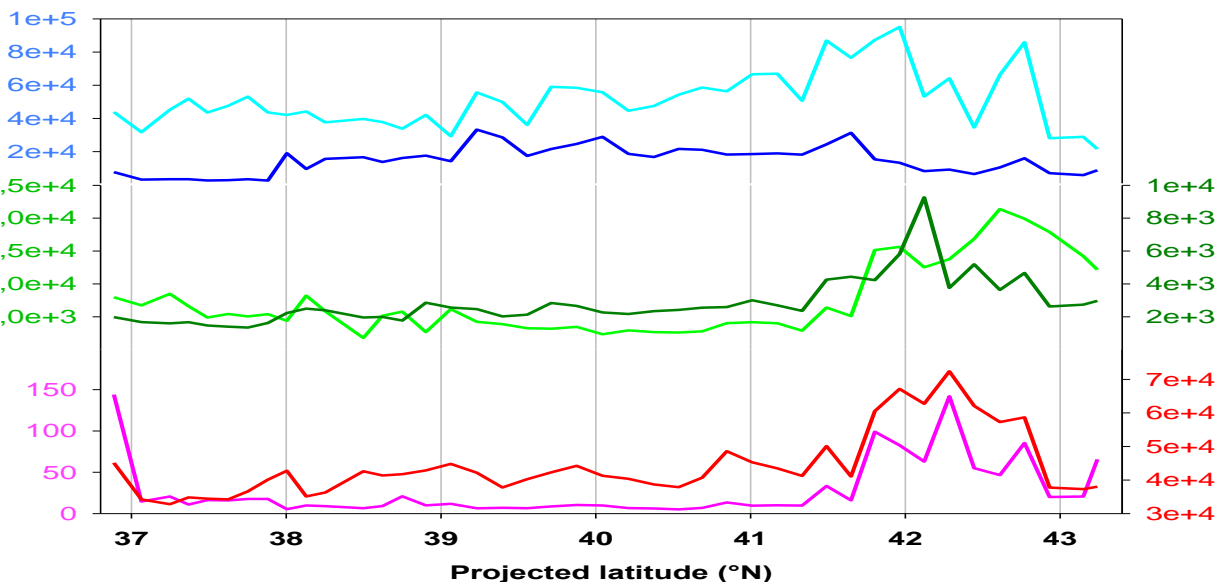
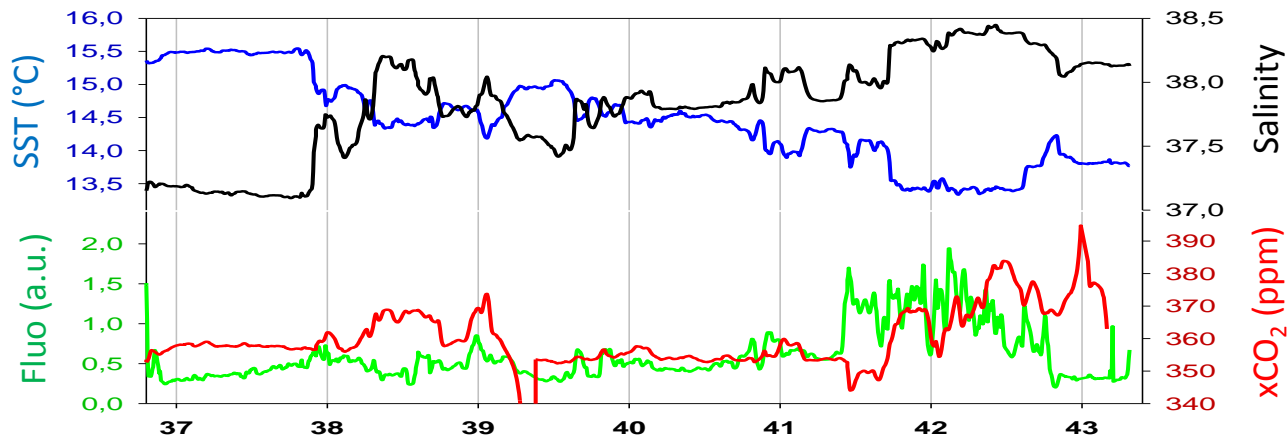
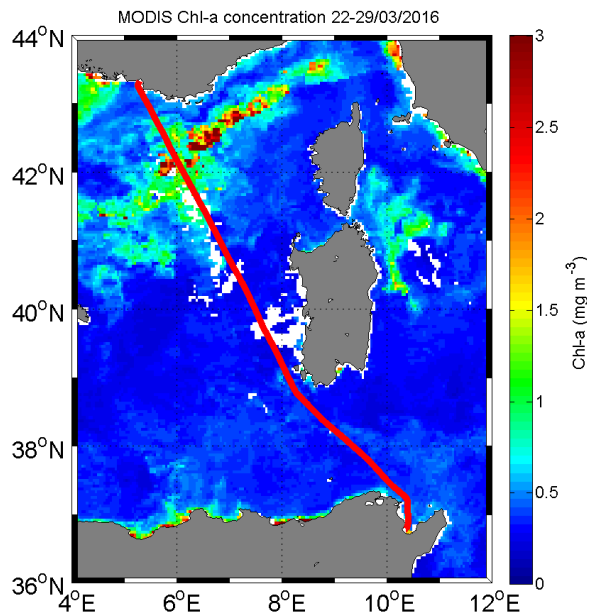




# First Results:

## 1 Marseille-Tunis transect

### 24-25/03/2016



— Prochlorococcus     — PicoEukaryote     — MicroEukaryotes  
— Synechococcus     — NanoEukaryotes     Cryptophytes Like

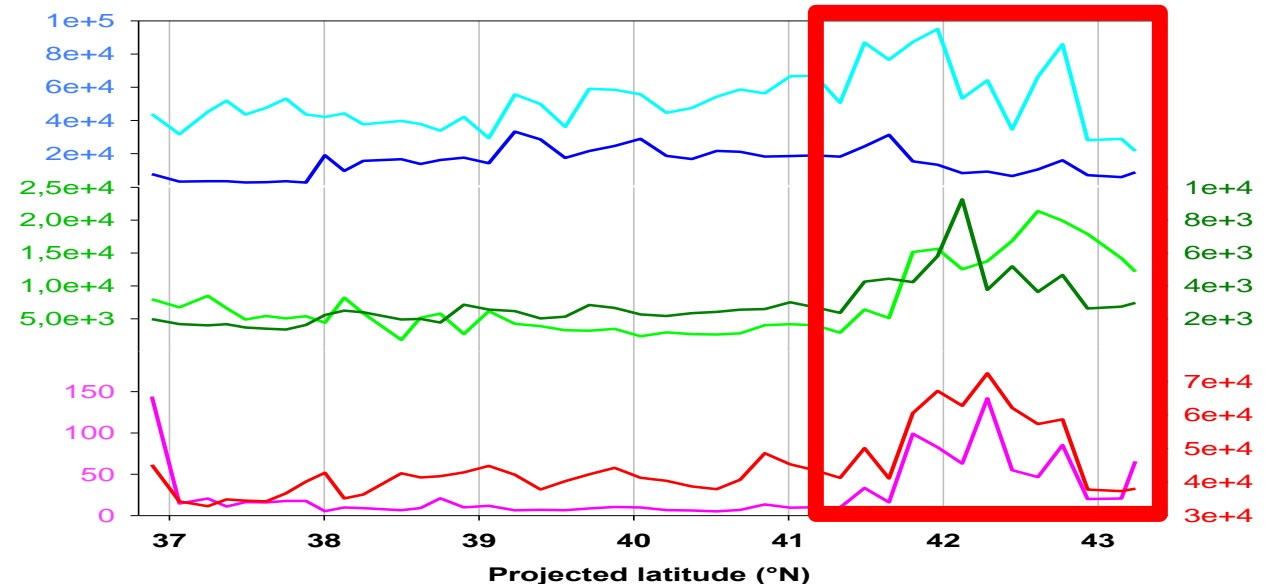
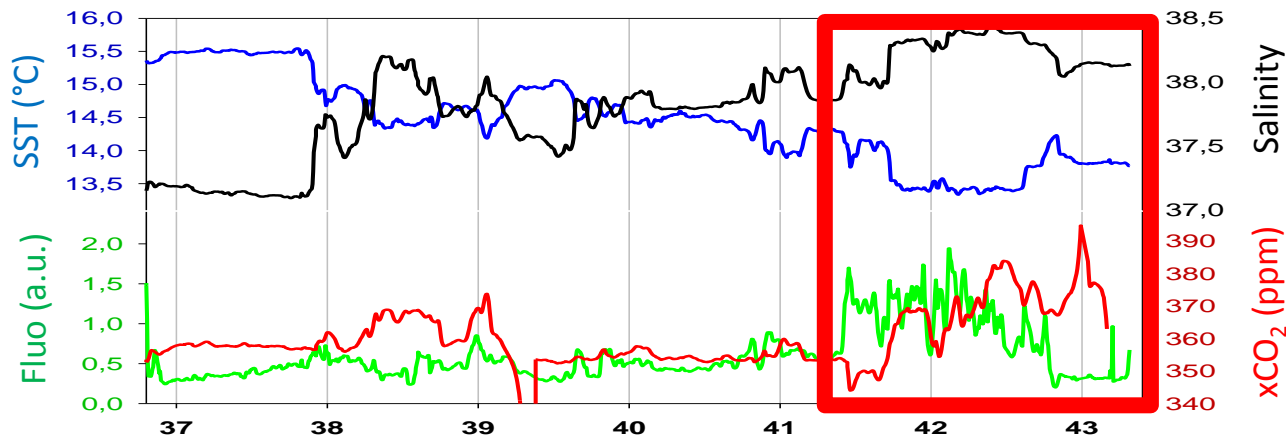
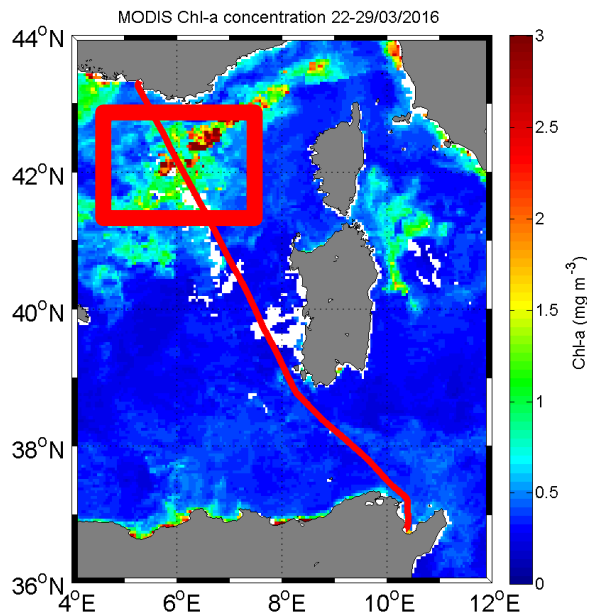
### Phytoplankton Group Abundance



# First Results:

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### 24-25/03/2016



— Prochlorococcus     — PicoEukaryote     — MicroEukaryotes  
— Synechococcus     — NanoEukaryotes     Cryptophytes Like

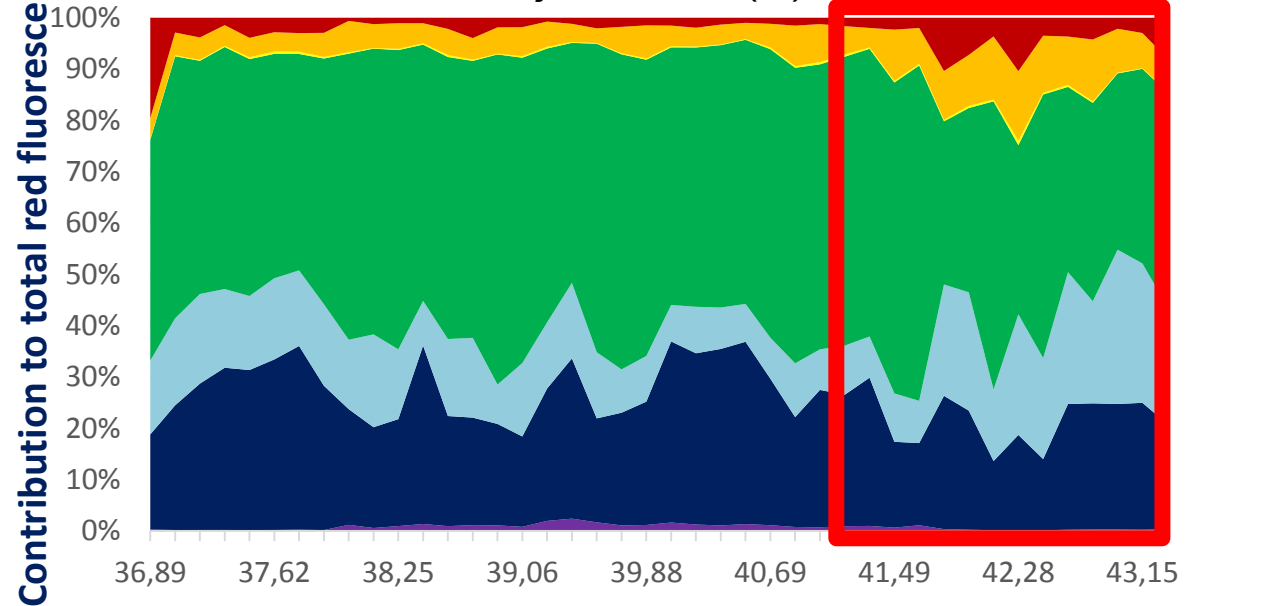
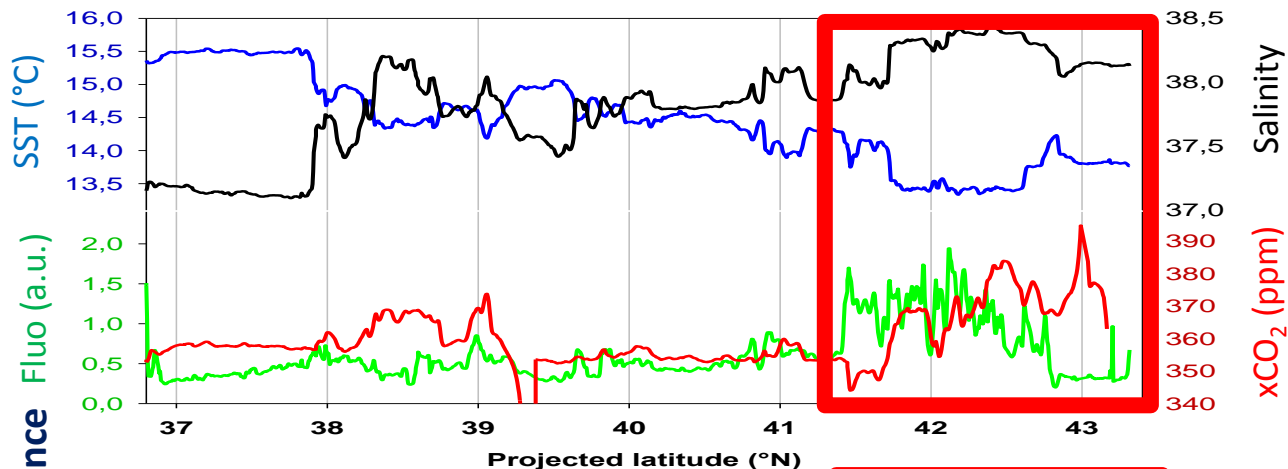
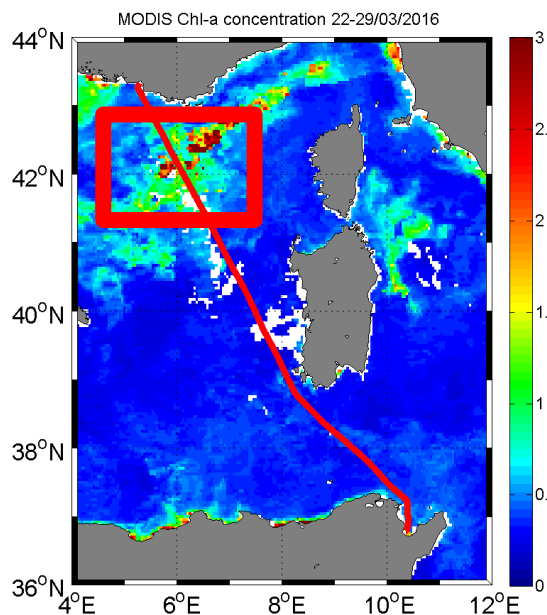
### Phytoplankton Group Abundance



# First Results:

## 1 Marseille-Tunis transect

### 24-25/03/2016



- Prochlorococcus
- Synechococcus
- PicoEukaryotes
- NanoEukaryotes
- Cryptophytes Like
- MicroEukaryotes





- Reinstallation of the sensors as soon and as long as possible.
- High-resolution phytoplankton structure heterogeneity.
- Essential information of phytoplankton contribution to  $p\text{CO}_2$  variability and biogeochemical processes.
- Expectation of a huge dataset to get new insights about surface Mediterranean ecosystems.



Continuous and High Resolution Observation of the Mediterranean Sea



7<sup>th</sup> FerryBox Workshop

# Thanks for your attention

