

## Implementation of high-frequency approaches to characterize the phytoplankton community and the physico-chemical supporting parameters in the Eastern English Channel and the North Sea.

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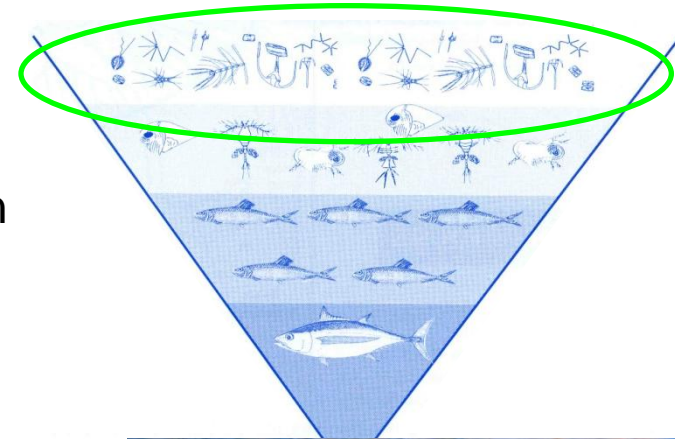
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# The DYMAPHY project

Development of a DYnamic observation system for the assessment of MARine water quality, based on PHYtoplankton analysis

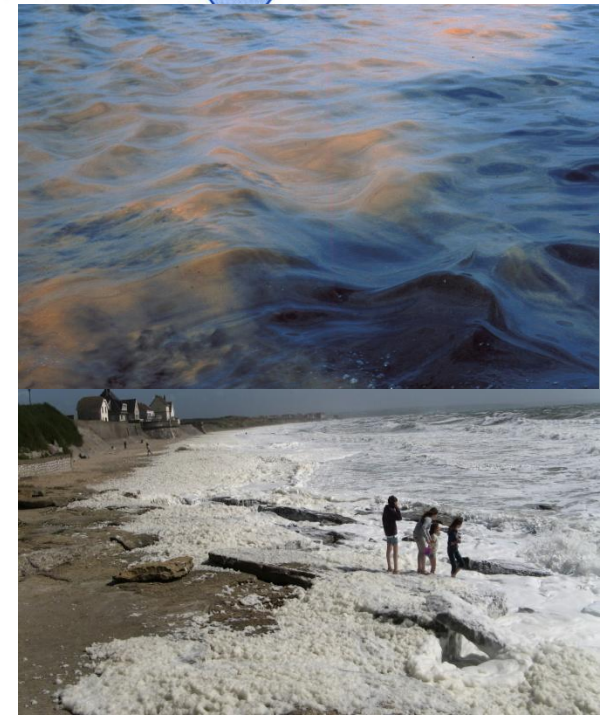
## Main objectives

- \* Development of standard operational procedures to monitor phytoplankton community structure *in situ* and in real-time.
- \* Better assessment of the quality of marine waters in the “2 Seas” Region.



**Focus** on phytoplankton as it represents the basis of all food chains in the sea and reflects the environmental status and water quality with consequences in socio-economic issues and human health (HAB).

Species composition and relative abundance of algal groups are fundamental determinants of aquatic ecosystem structure and function.



## Research Context

- Marine biodiversity as a key for goods and ecosystem services
- Fundamental knowledge (patrimony)
- Regime shifts in phytoplankton communities (natural vs anthropogenic)
- Phenology of phytoplankton blooms
- Occurrence of Harmful Algal Blooms
- Global change

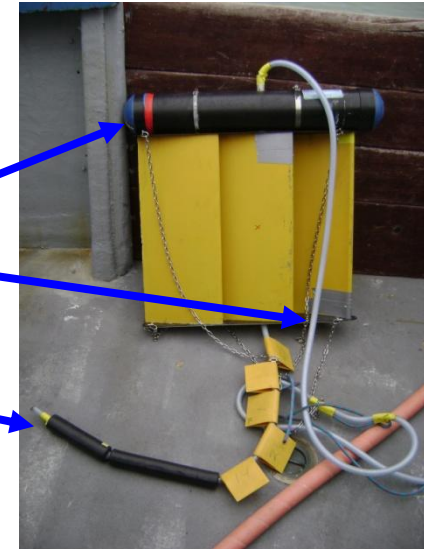
## Monitoring Context

- The OSPAR strategies (1992)  
(Common Procedure – Eutrophication Strategy)
- The Water Framework Directive (2000/60/EC)
- The Marine Strategy Framework Directive (2008/56/EC)

⇒ Reliable, cost-effective, (near) real-time information need for scientists, regional managers and policy-makers



# Development of the underwater sampling unit (The DymaPhin system = Dymaphy + Dolphin)



Float

Pipe connection to the pump

Water Intake  
(2 m depth)

*In situ* test with the RV "Sepia II" (cnrs/insu)



Test in the wave and current basin (Ifremer)



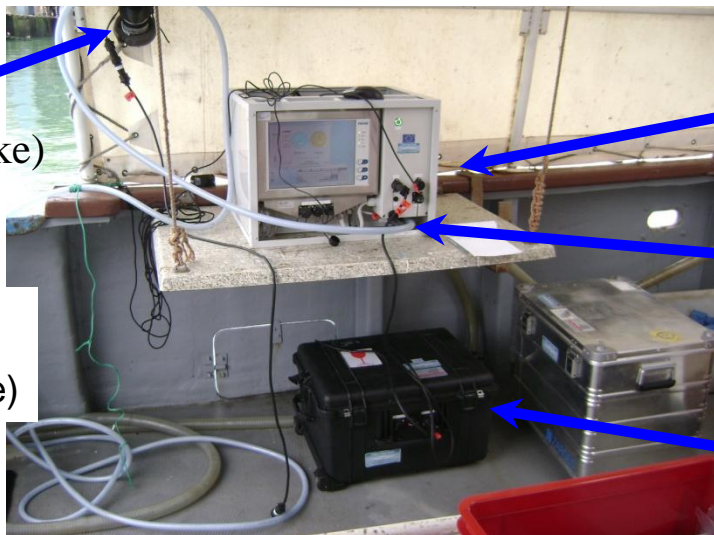


# DYMAPHY - Actions 1 & 3

Test and implementation of automated systems for water quality assessment

## Test of the Pocket Ferry Box + AOA on the RV " Sepia II " (cnrs/insu)

Pump  
(Water Intake)



The Pocket Ferry Box  
on the RV « Sepia II »

Possibility to add a data  
sonde on the water output  
(complementary parameters)

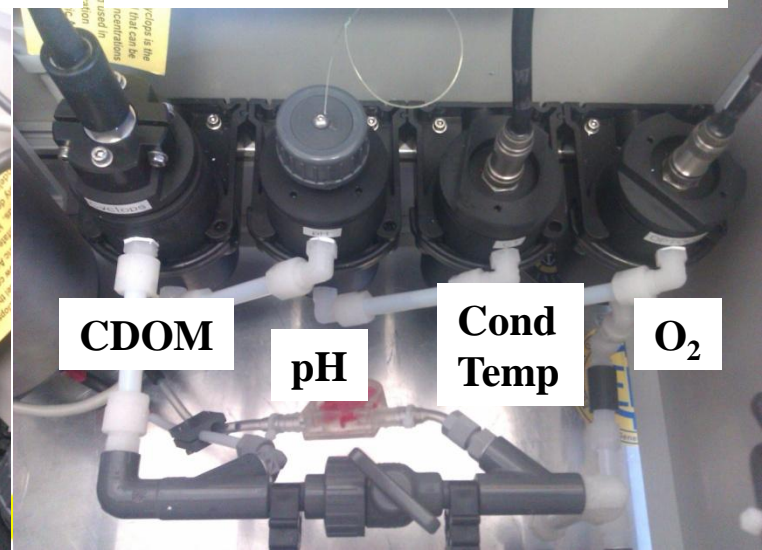
Battery

All « out of water » system  
=> Easy access (maintenance)

Top view of the Pocket Ferry Box

Algae Online Analyzer (AOA)

Top view of the Pocket Ferry Box



## The Algae Online Analyzer (bbe)

- Fixed-wavelength spectral fluorometer
- LED centered at 470, 525, 570, 590 and 610 nm (+ CDOM 370 nm)
- Emission measured at 680 nm.
- Built-in data-analysis and reporting
- Specification of the library of spectral fluorescence (algal fingerprints) => characterization of the phytoplankton community as spectral groups (as a preliminary taxonomic determination)
- Original fingerprints described in AOA parlance as Green, BlueGreen, Diatoms, Cryptophyceae



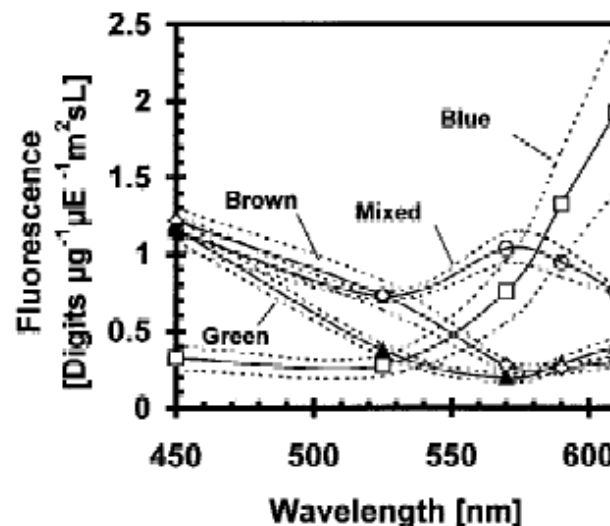
### Main principle

*Shape of the spectral fluo signature*

=> *taxa discrimination*

*Fluo intensity and the group-specific fluo/chl ratio*

=> *total phytopl biomass (chl<sub>a</sub>)*

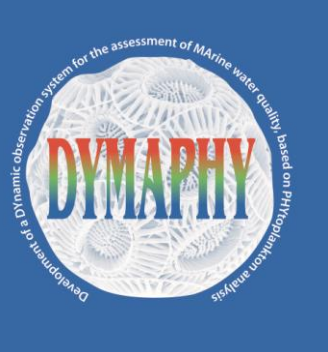


*Mean fluorescence-excitation probabilities for 4 spectral algal groups (norm spectra) (Source : Beutler et al., 2002)*

*Sampling frequency: 1 min. continuous sampling mode / spatial resolution approx. 0.1 nm*

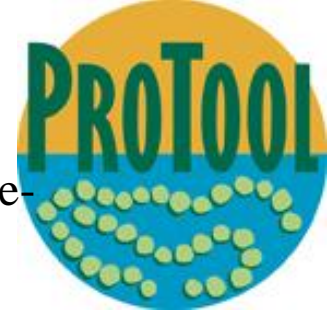
*System coupled with a YSI 6600 water-quality probe connected with a flow-through system (for comparison / added parameters) and with a flow cytometer (sampling frequency: 10 min.)*

*Discrete samples (water quality, phytoplankton, HPLC, spectro, fluo, flow cytometry)*



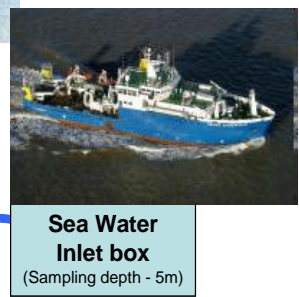
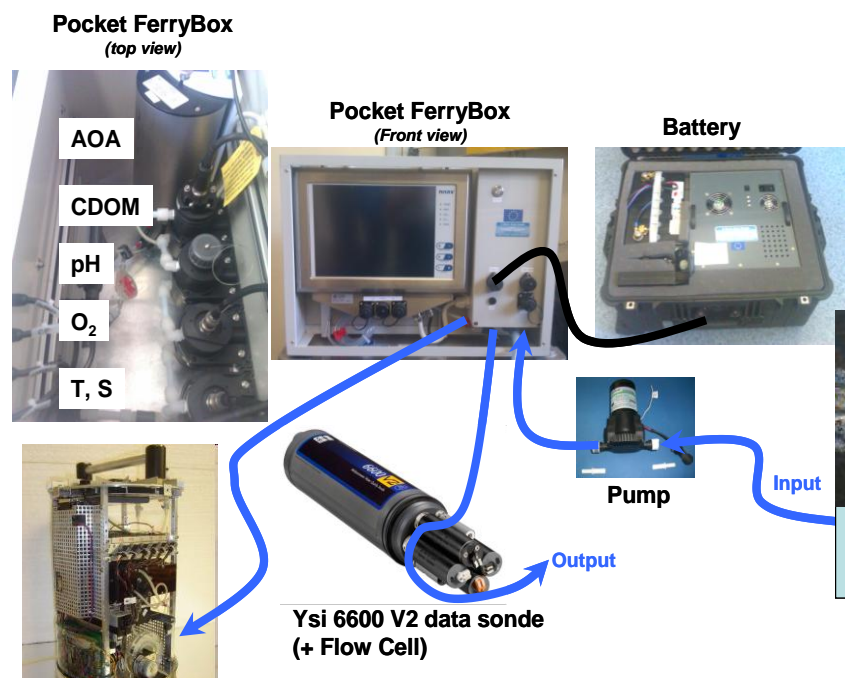
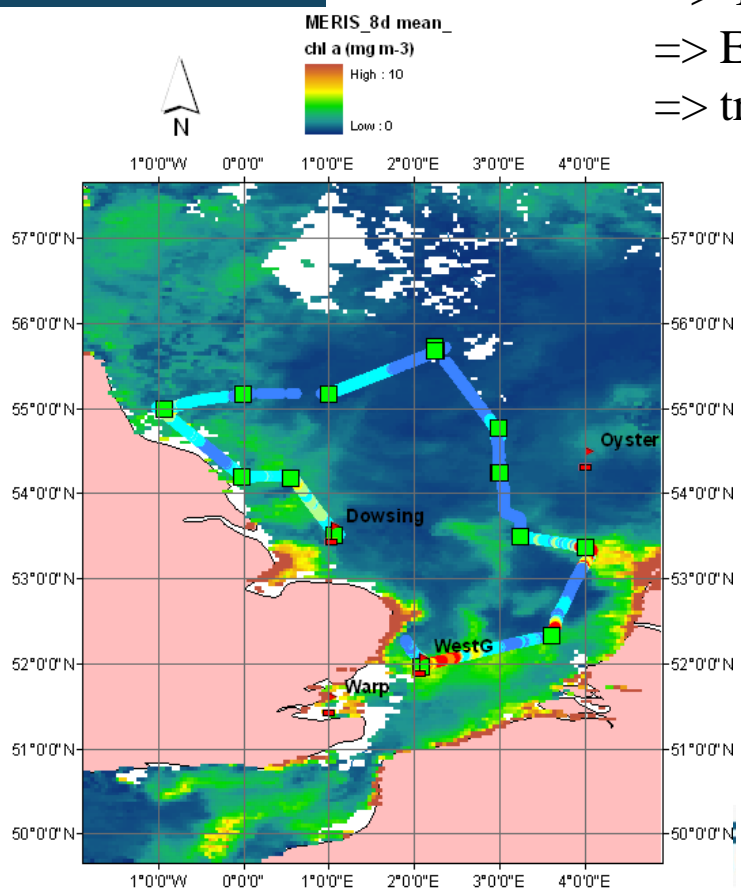
# The Protocol / Dymaphy campaign

## 7 – 13<sup>th</sup> May 2011



The **Pocket Ferry Box** (4H-JENA©) coupled with a multiple-fixed wavelength **spectral fluorometer** (AOA, bbe©), an **YSI Data Sonde** and a **Cytosense flow cytometer** (Cytobuoy ©) was implemented during the scientific cruise PROTOOL (+ PAM, Fastracka, PSI fluorometers, radiocarbon) on the RV « Cefas Endeavour ».

- = > Test of the reliability of the system
- => Exchanges on « problems » or update needs
- => training sessions with 4H-JENA, BBE





# The Protocol / Dymaphy campaign

7 – 13<sup>th</sup> May 2011

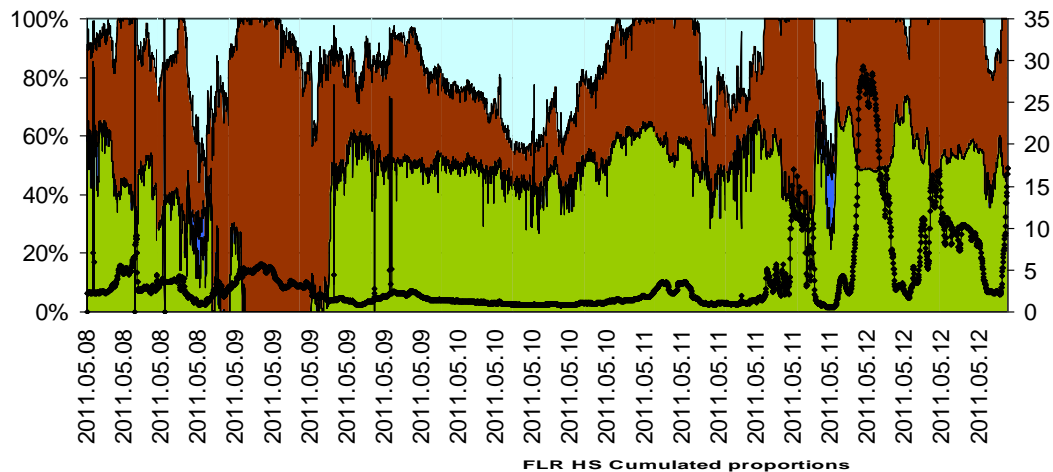
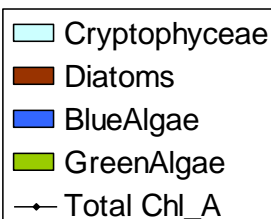
Dutch coastal

England  
East coast

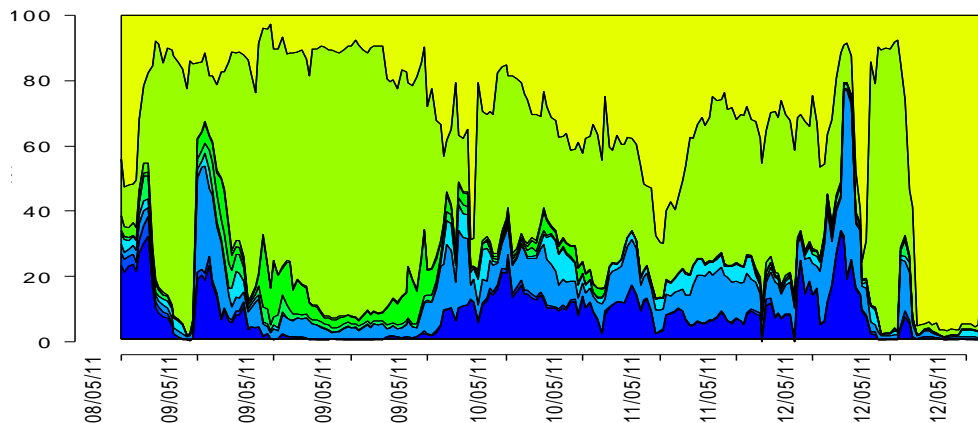
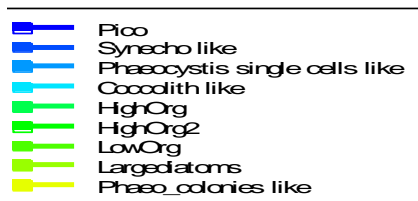
Central North Sea

Outer Thames

## Algae Online Analyser



## Flow cytometer



A total of 9 groups were distinguished by flow cytometry based on their optical properties.

We show **high spatial temporal changes in the phytoplankton community** which could hardly be detected by employing discrete sampling strategies. High frequency measurements (HFM) allows to identify precisely, in time and space, the **contrasted environment** sampled. Moreover within a given environment, HFM highlight **changes in the composition of the phytoplankton community**.



**Main problem :**  
Before / after cruise control of  
the parameters of fit  
⇒ Settings of the fingerprints  
according to references  
(discrete samples)

Original Fingerprints

Parameters of fit (2011-03-24)

Offsets LEDs:  filtrated water  distilled water

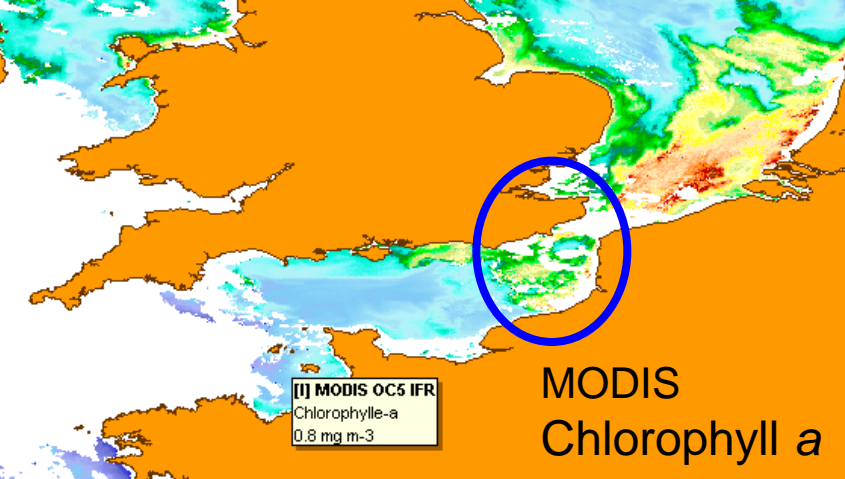
|    |   |        |   |        |   |        |   |        |   |        |    |        |
|----|---|--------|---|--------|---|--------|---|--------|---|--------|----|--------|
| F  | 1 | 11.093 | 2 | 3.3564 | 3 | 3.4712 | 4 | 3.3331 | 5 | 9.3363 | UV | 16.953 |
| F0 | 1 |        | 2 |        | 3 |        | 4 |        | 5 |        | UV | —      |
| FM | 1 |        | 2 |        | 3 |        | 4 |        | 5 |        | UV | —      |

global corr. factor 1

| fit_type                            | name              | factor LEDs |         |         |         |         |         | standard deviation of LEDs |     |     |     |     |     |
|-------------------------------------|-------------------|-------------|---------|---------|---------|---------|---------|----------------------------|-----|-----|-----|-----|-----|
|                                     |                   | 1           | 2       | 3       | 4       | 5       | UV      | 1                          | 2   | 3   | 4   | 5   | UV  |
| <input checked="" type="checkbox"/> | Green Algae       | 1.313       | 0.249   | 0.417   | 0.308   | 2.417   | 2.793   | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input checked="" type="checkbox"/> | Bluegreen         | 0.955       | 1.041   | 2.217   | 1.431   | 0.237   | 1.259   | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input checked="" type="checkbox"/> | Diatoms           | 7.149       | 0.794   | 0.746   | 0.633   | 6.972   | 6.647   | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input checked="" type="checkbox"/> | Cryptophyta       | 4.115       | 1.578   | 1.056   | 1.202   | 3.129   | 3.149   | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input type="checkbox"/>            | Phaeocystis       | 1           | 1       | 1       | 1       | 1       | 1       | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input type="checkbox"/>            | Pseudonitzschia   | 4.49748     | 0.50878 | 0.48567 | 0.45056 | 4.50818 | 1.87941 | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input type="checkbox"/>            | #6                | 1           | 1       | 1       | 1       | 1       | 1       | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |
| <input checked="" type="checkbox"/> | Yellow substances | 3.165       | 0.298   | 0.13    | 0.201   | 4.574   | 15.185  | 0.1                        | 0.1 | 0.1 | 0.1 | 0.1 | 0.1 |



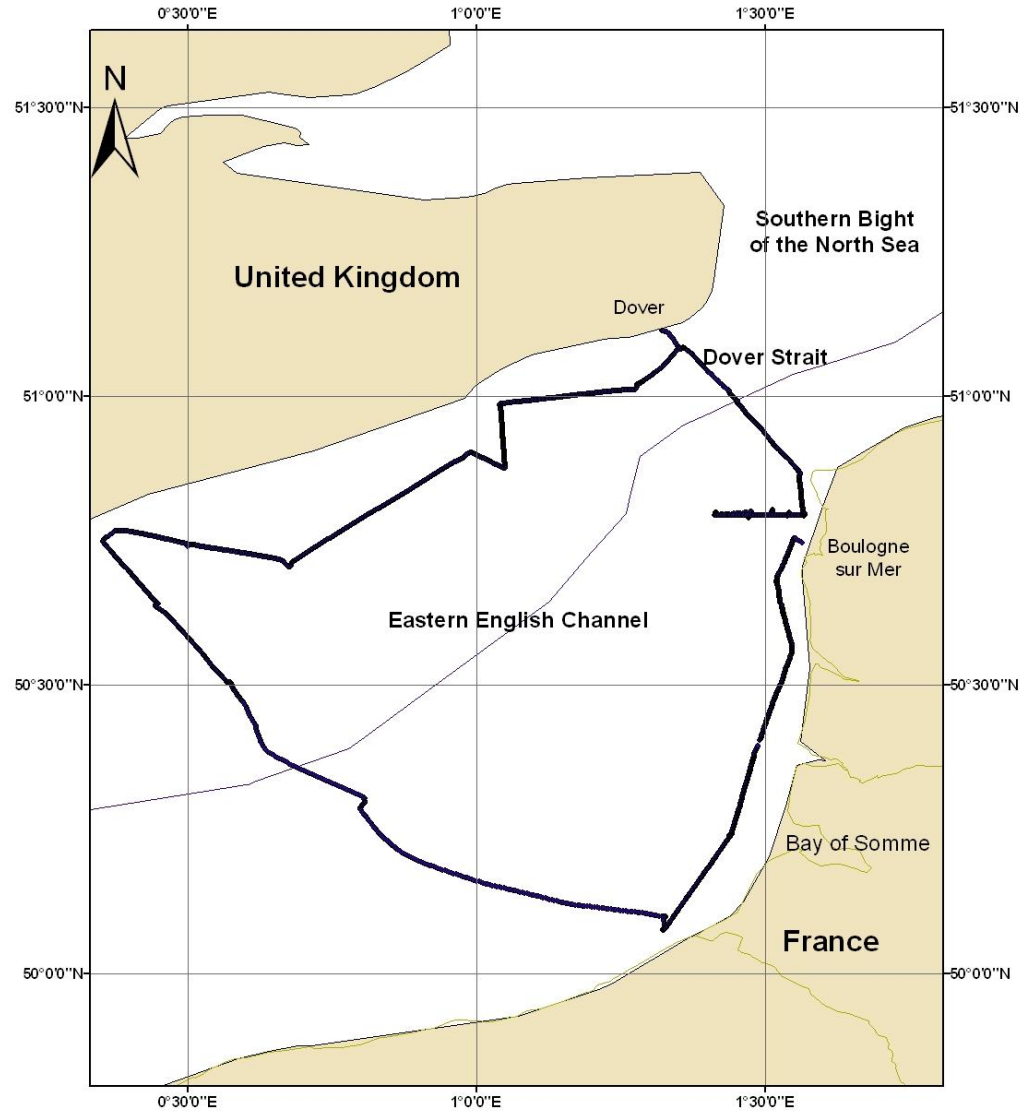
Implementation of new fingerprints: (Limited numbers)  
*Pseudonitzschia, Phaeocystis globosa, Isochrysis*



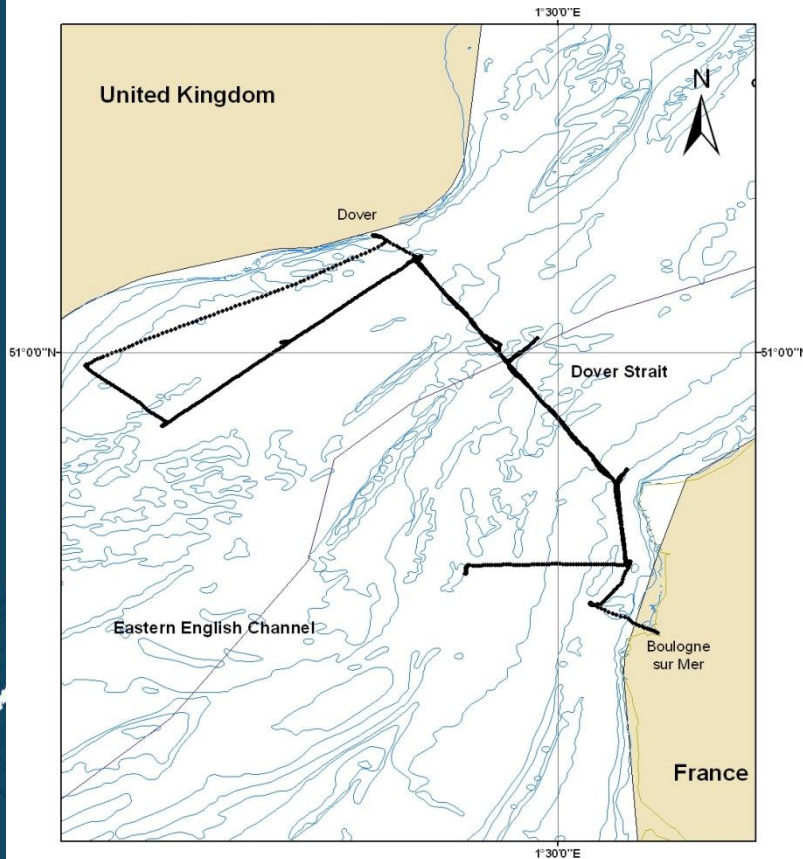
# The Dymaphy campaign on the RV "Côtes de la Manche" (cnrs/insu)

Towards a synoptic *in situ* view thanks to HFM

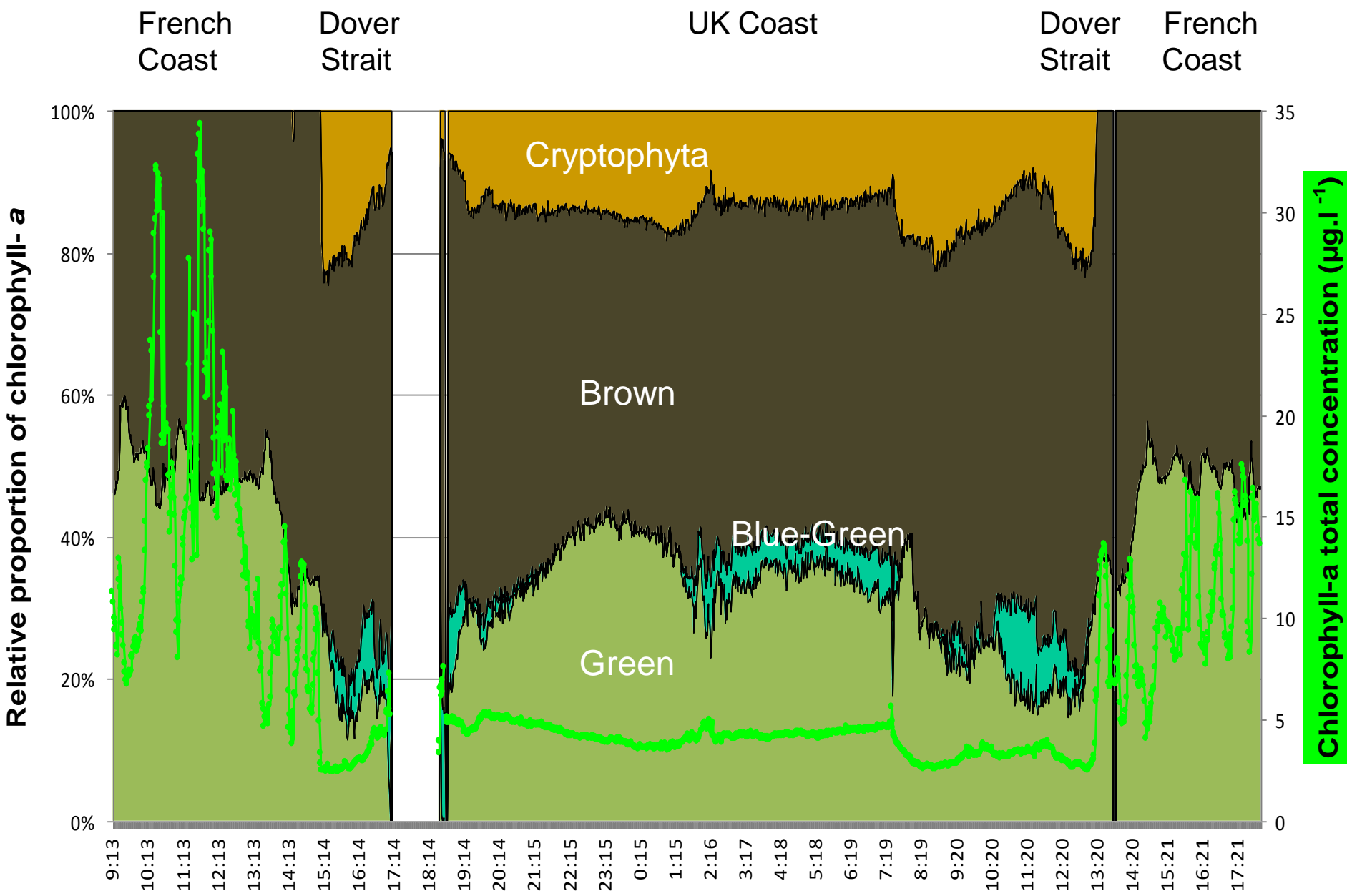
## LEG 2 - 27-29/4/2012



## LEG 1 - 20-21/4/2012



Original Fingerprints



Fingerprints **Blue Green** / **Brown** / **Cryptophyceae** / *Phaeocystis*

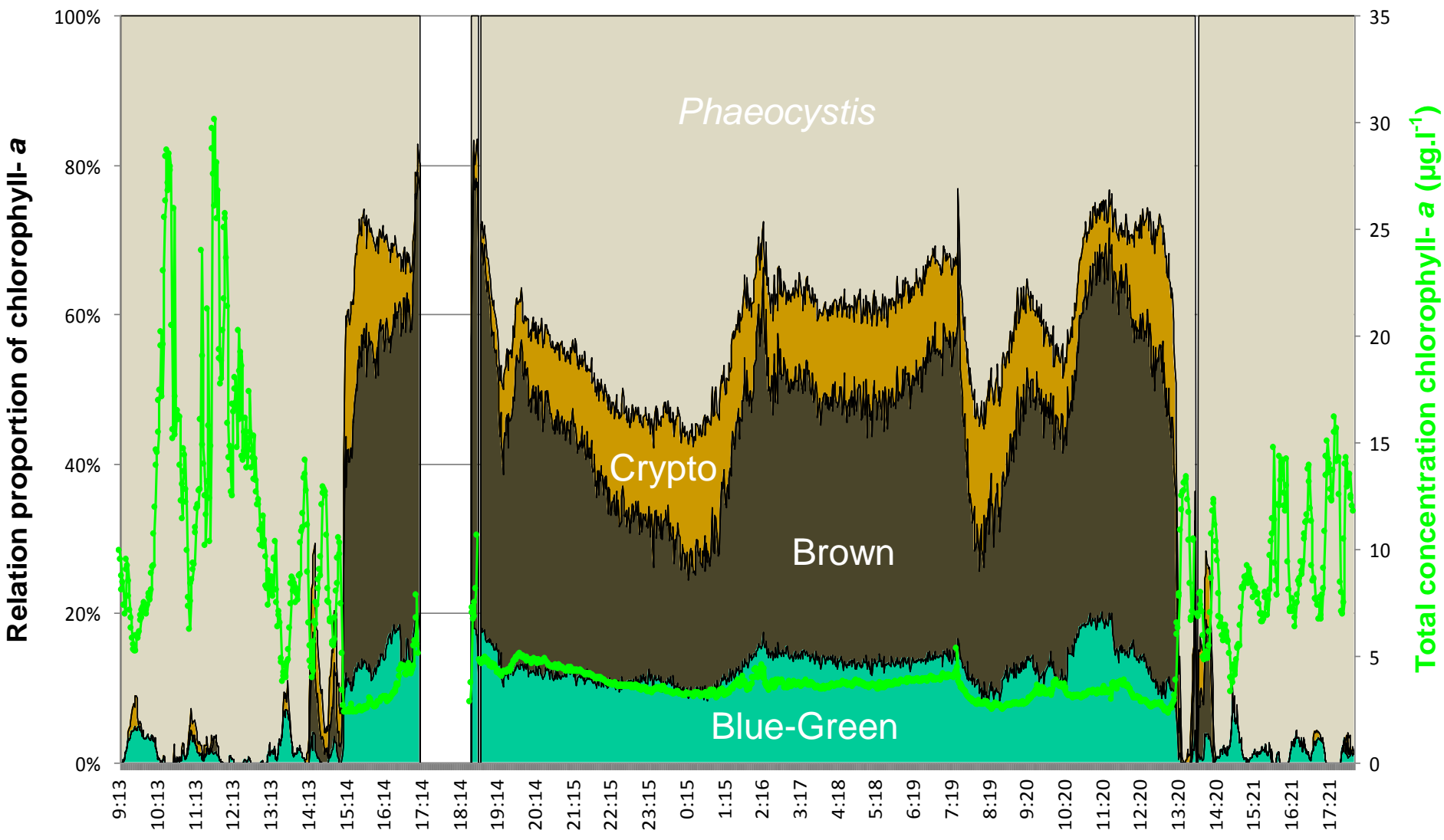
French Coast

Dover Strait

UK Coast

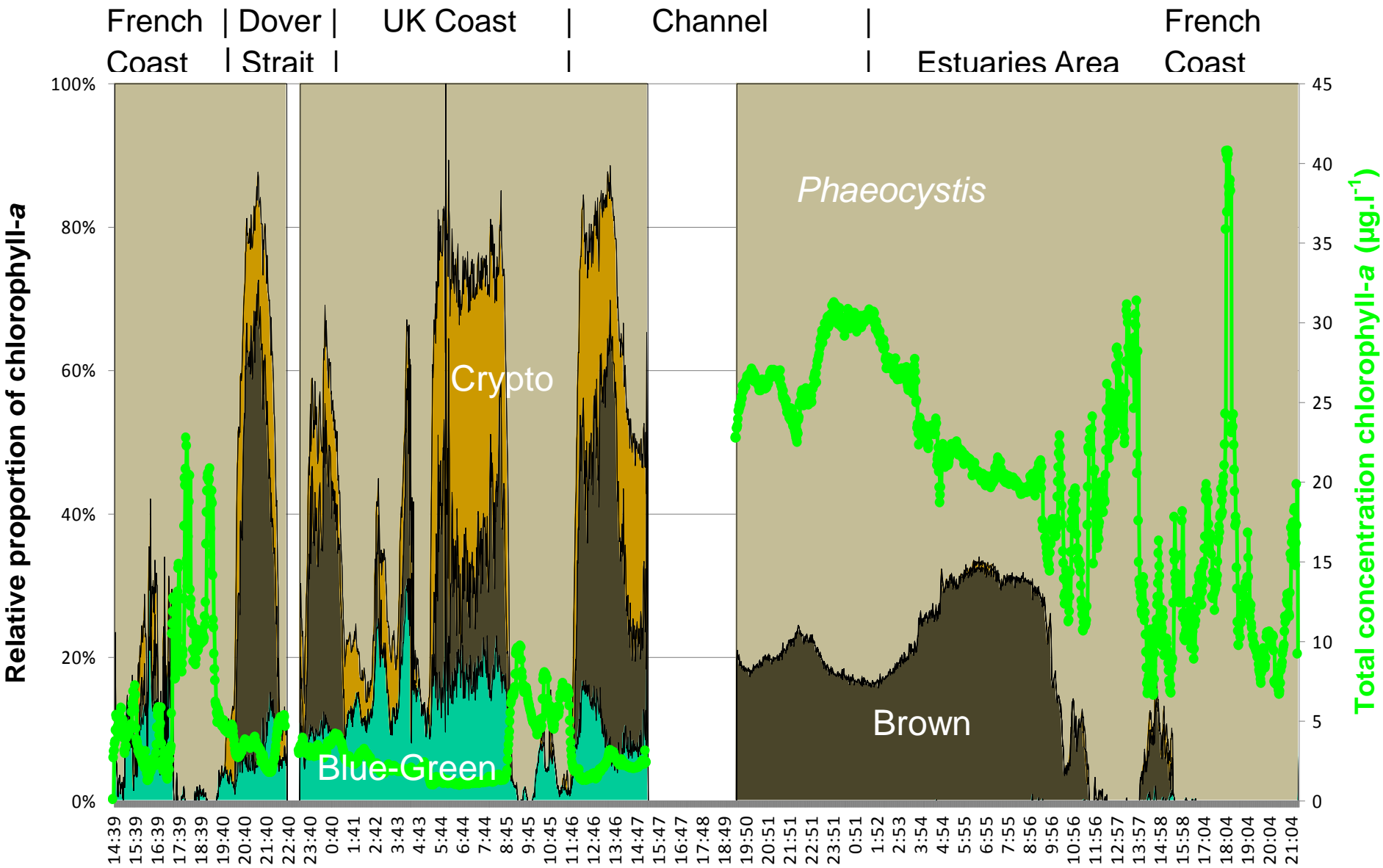
Dover Strait

French Coast





Fingerprints **Blue Green** / **Brown** / **Cryptophyceae** / *Phaeocystis*



## Next steps (*ongoing analysis*)

- Verification of phytoplankton community composition with those from microscopy (Utermöhl 1958) and flow cytometry
- Compare estimates of biomass with HPLC values and variations (effect of light intensity and nutrient availability on fluo quenching)
- Compare estimates of taxonomic structures with those from HPLC-derived marker pigment by ChemTax
- Relations of the highlighted patterns with physico-chemical supporting factors (nutrients, turbidity, ...)

## Recommendations

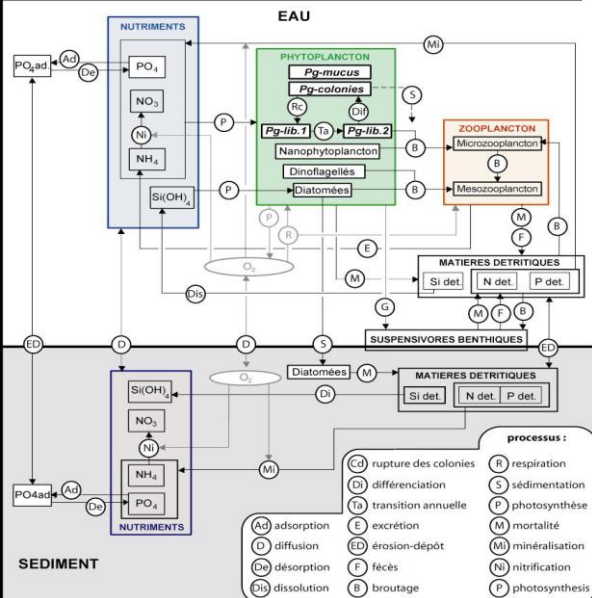
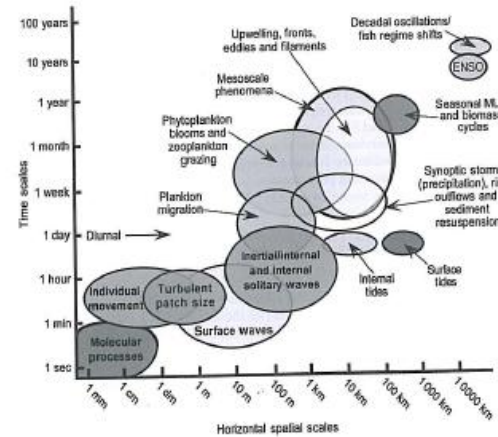
- Frequent calibration by discrete sampling collections
- Calibration/Fingerprint with mixture of species (not with a single sp.) representative of the eco-region of interest to avoid misclassification, then over-estimation of contribution

## Conclusions

- HFM useful, otherwise essential, for monitoring of phytoplankton community composition and supporting physico-chemical parameters (WFD, MSFD, OSPAR,...).
- Use as an Early Warning System (HAB, direct and indirect effects of eutrophication)  
=> ability to separate diatoms from dinoflagellates !?
- Microscopy still needed for species identification.
- Need to couple systems (PFB + HPLC + flow cytometry, satellite imagery ...) => *in situ* HF and synoptic view.
- On-line measurements allows real-time adjustment in the sampling strategy (discrete samples), and for monitoring purposes it allows to take the maximum possible time to take the necessary actions (communication to the authorities, shellfish farming management, ...).



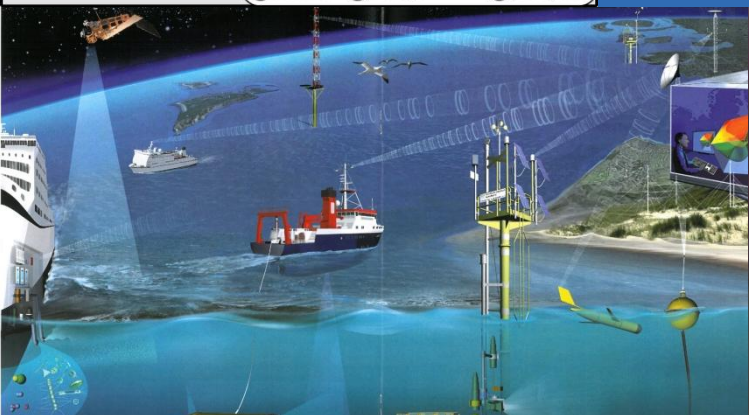
Thanks for your attention!  
 Merci pour votre attention !



**DYMAPHY**  
 Development of a Dynamic observation system for the assessment of MARine water quality, based on PHYtoplankton analysis

2 Mers Seas Zeeën  
 INTERREG IV A  
 FRANCE - ENGLAND - FLANDERS - NEDERLAND

Logo of the French Republic (Mars National de Galles) and the European Union flag.



[http://wwz.ifremer.fr/defimanche/projets/en\\_cours/dymaphy](http://wwz.ifremer.fr/defimanche/projets/en_cours/dymaphy)  
<http://www.dymaphy.eu>