ODATIS Données et Services pour l'Océan

1er Atelier technique

« Bonnes pratiques techniques au service de l'interopérabilité »

Soumaya LAHBIB – Institut Méditerranéen d'Océanologie (MIO)

Campus Luminy, Marseille

Brest, 11 et 12 septembre 2017

Institut Pythéas
Observatoire des Sciences de l'Univers
Aix*Marseille Université

mio Institut Méditerranéen d'Océanologie

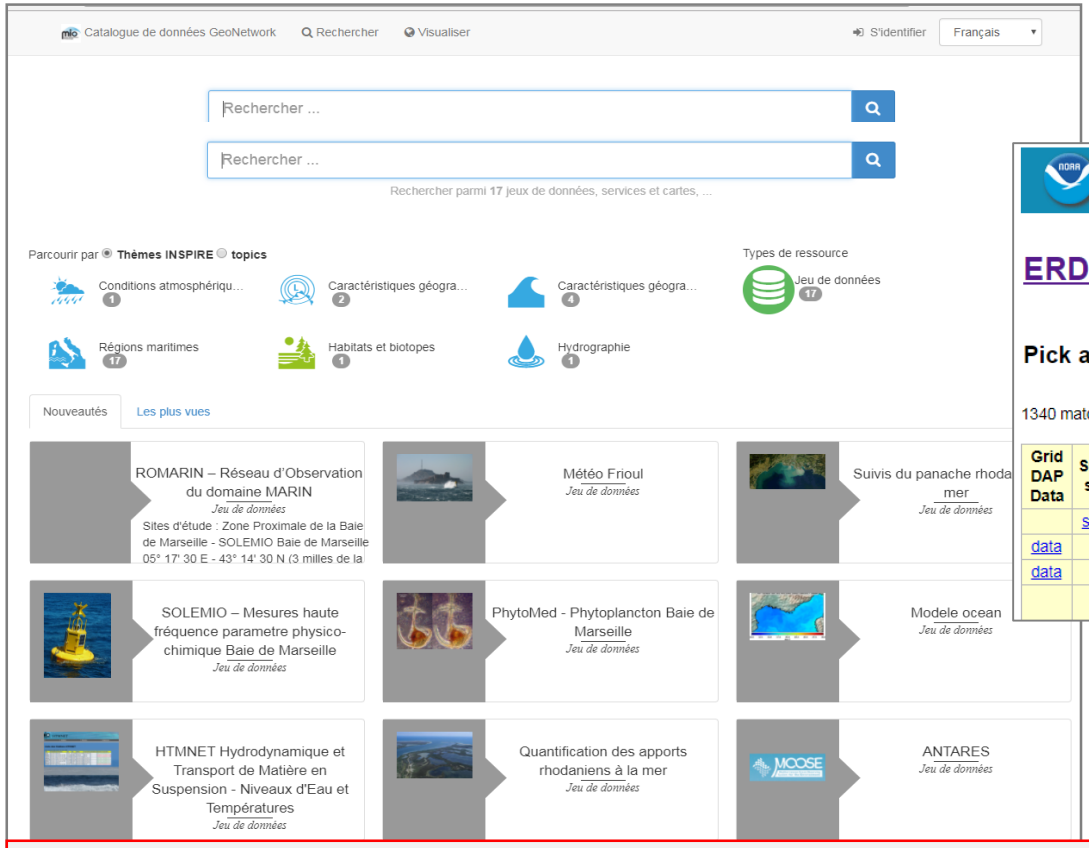
cnrs dépasser les frontières

Centralisation des jeux de données de l'OSU

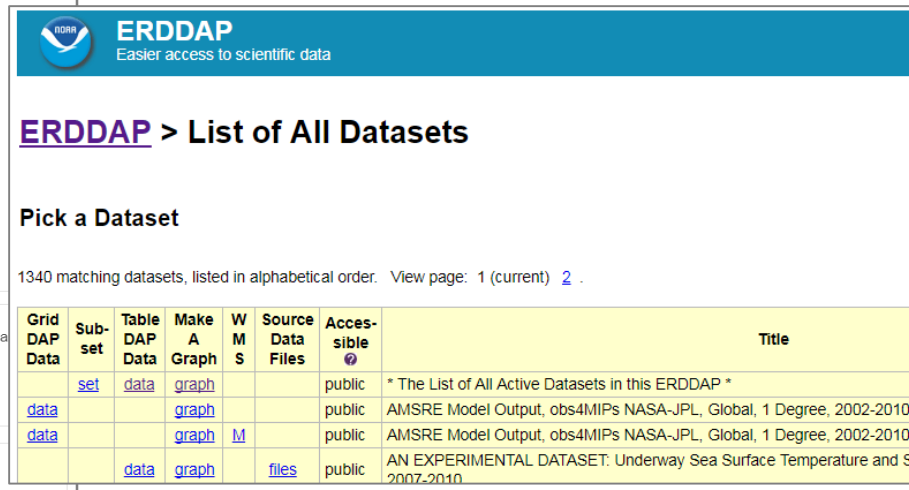
En cours par Maurice Libes

Géo-cataloguage

http://139.124.2.98/geonetwork/srv/fre/catalog.search#/home



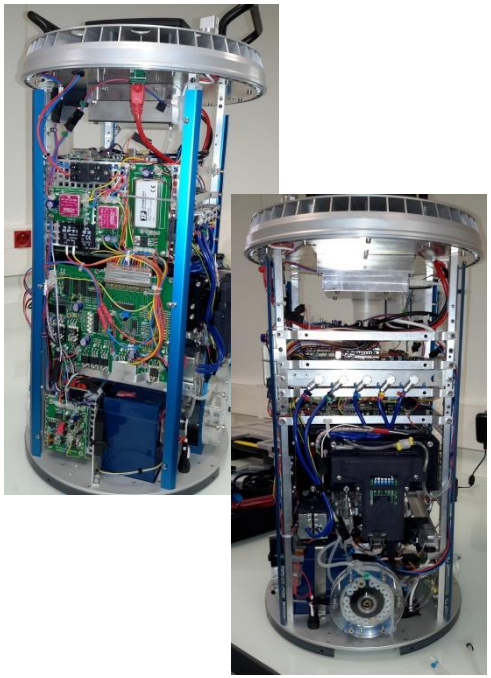
Visualisation



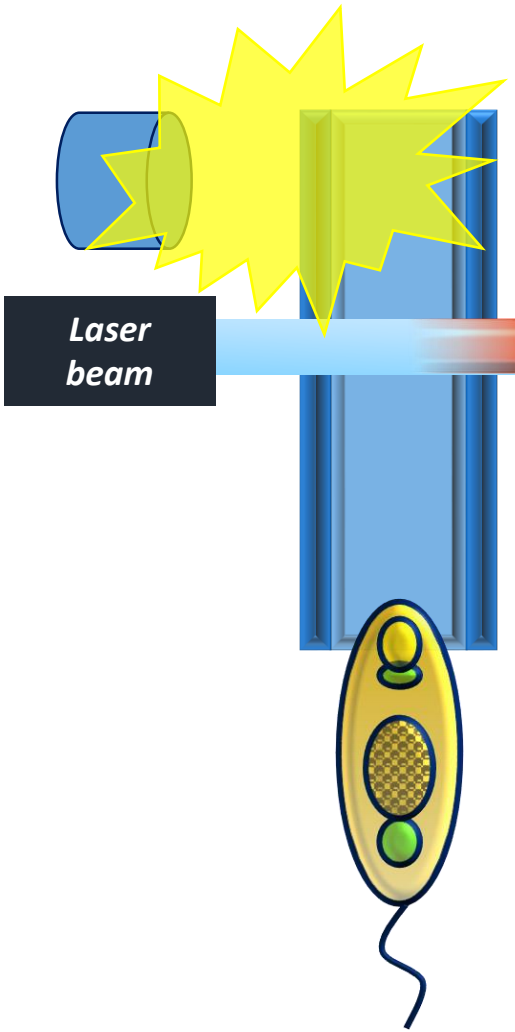
Frejus du 4 au 7 Décembre 2017 **ANF gestion des données scientifiques : recherche intervenant ERDDAP**

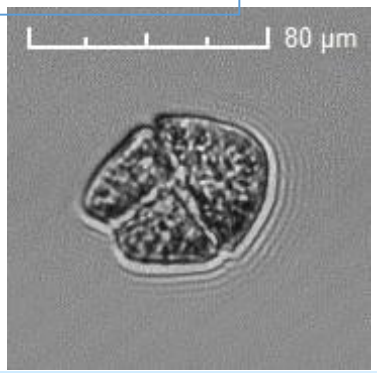
Equipe de Cytométrie en flux

New technology for the resolution of phytoplankton functional diversity at hourly and regional scales



CytoBuoy
flow cytometry solutions





Fluorescences
Scatters
Abundances

CHROME

Continuous High Resolution Observation of the Mediterranean Sea:

https://chrome.mio.univ-amu.fr/

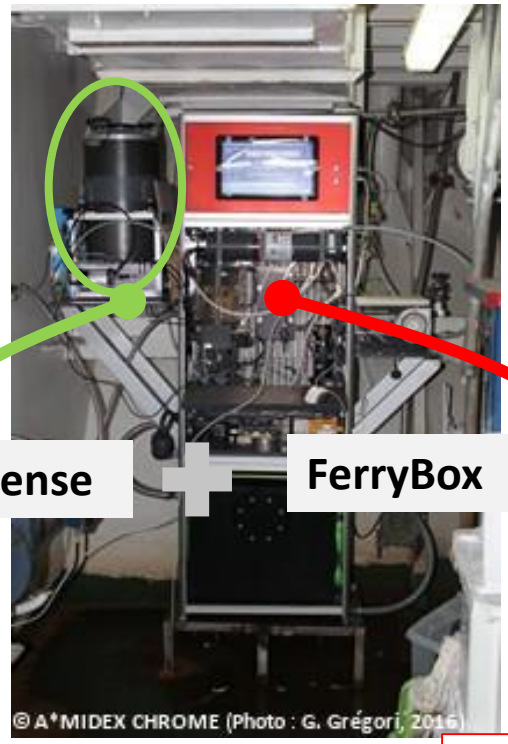
Understanding of the ecological and biogeochemical functioning in relation to meso-scale dynamics at the Mediterranean sub-basin scale and weekly scale.

A*MIDEX CHROME Project

Data Acquisition = one analysis every 30 min.

— C/F CARTHAGE trajectories

Pont 1 – C/F CARTHAGE



CytoSense + **FerryBox**

mio Institut Méditerranéen d'Océanologie

INSTM

./30min

./1min

- **Phytoplankton functional groups**
- **Phytoplankton abundance per group**
- **Fluorescences/scatter per cell**
- **Size estimation after calibration of scatter**
- **Phytoplankton images (taxonomical identification >20 µm)**

– **Temperature**
– **Salinity**
– **Fluoresence**
– **Turbidity**
– **pH**
– **pCO_2**
– **Oxygen**

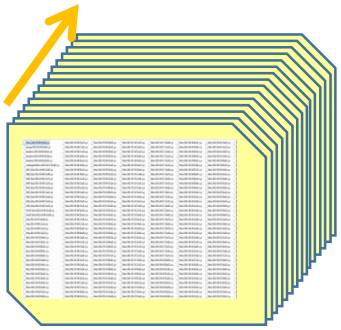
Data acquisition & analysis

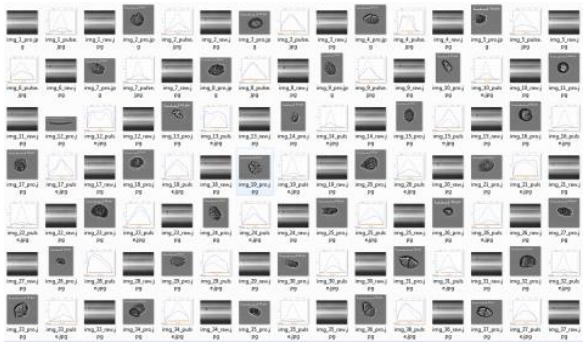
Measurements by CytoSense

CytoClus

Output

Manual clustering





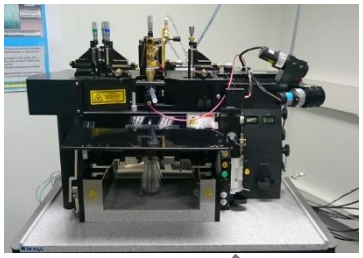
Separate statistical CSV files:
Average values of optical properties and Counts
+ Pictures

Backup

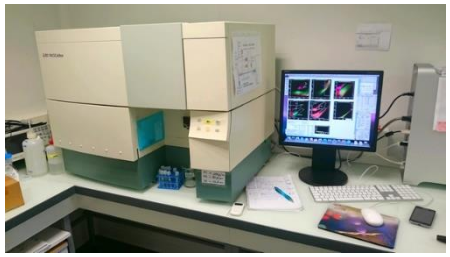
MIO FCM Data management
(*working on interoperability with SeaDataNet*)

In situ **automated cytometer**

Bentchtop flow cytometers



BD Influx



FACS Calibur

CytoSense

CytoClus

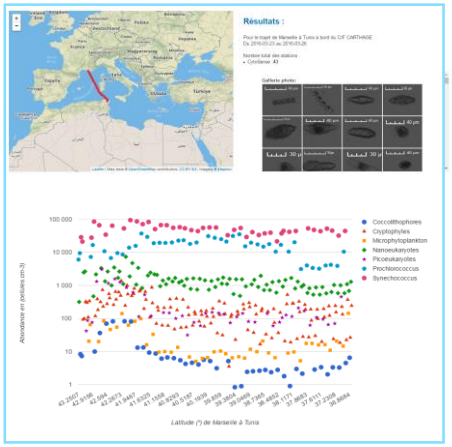
R

Conversion tool
+ Metadata entry

M. Dugenne, 2015

ASCII Data Table & Pictures

Acquisition | **Analyses** | **Consolidation** | **Visual QC**



CYTOBASE

Accessibility

Integration

→ Data Consolidation

Cytobase Input Processor (Mathilde Dugenne, 2015)

mathilde.dugenne@mio.osupytheas.fr

Parcourir... 11 fichiers sélectionnés.

Upload complete

Warning:

Upload successful

Create Inputs folder

Project and samples context | Raw data | Size conversion

Metadata

Project	Project date	PI	Cytometer ID
Enter project name	2015-08-18	Enter PI name	Enter cytometer ID
Station	**Depth**	**Latitude**	**Longitude**
Filename	**Filename model**		
	BERRE_082013_3F_FLR9 2013-12-17 13(		
Samples operator	**Standards reference**	**Clustering method**	**Observation type**
Enter name of operator	Enter standards beads ref	Automated	In situ

Data

2013-12-17T14:17:00Z	2013-12-17T14:17:00Z	16,056.60		Synechococcus	BERRE_082013_12S_FLR9 2013-12-17 14u17.cyz	1.99	FL Red	10
2013-12-17T14:17:00Z	2013-12-17T14:17:00Z	16,056.60		Cryptophytes	BERRE_082013_12S_FLR9 2013-12-17 14u17.cyz	1.99	FL Red	10
2013-12-17T14:43:00Z	2013-12-17T14:43:00Z	16,056.61		Beads 2 mu	BERRE_082013_16F_FLR9 2013-12-17 14u43.cyz	2.04	FL Red	10
2013-12-17T14:43:00Z	2013-12-17T14:43:00Z	16,056.61		Microphytoplankton	BERRE_082013_16F_FLR9 2013-12-17 14u43.cyz	2.04	FL Red	10
2013-12-17T14:43:00Z	2013-12-17T14:43:00Z	16,056.61		Picooeukaryotes 2	BERRE_082013_16F_FLR9 2013-12-17 14u43.cyz	2.04	FL Red	10
2013-12-17T14:43:00Z	2013-12-17T14:43:00Z	16,056.61		Picoeukaryotes 1	BERRE_082013_16F_FLR9 2013-12-17 14u43.cyz	2.04	FL Red	10

Please associate each selection set to trigger, PMT's amplification and standardized phytoplankton category

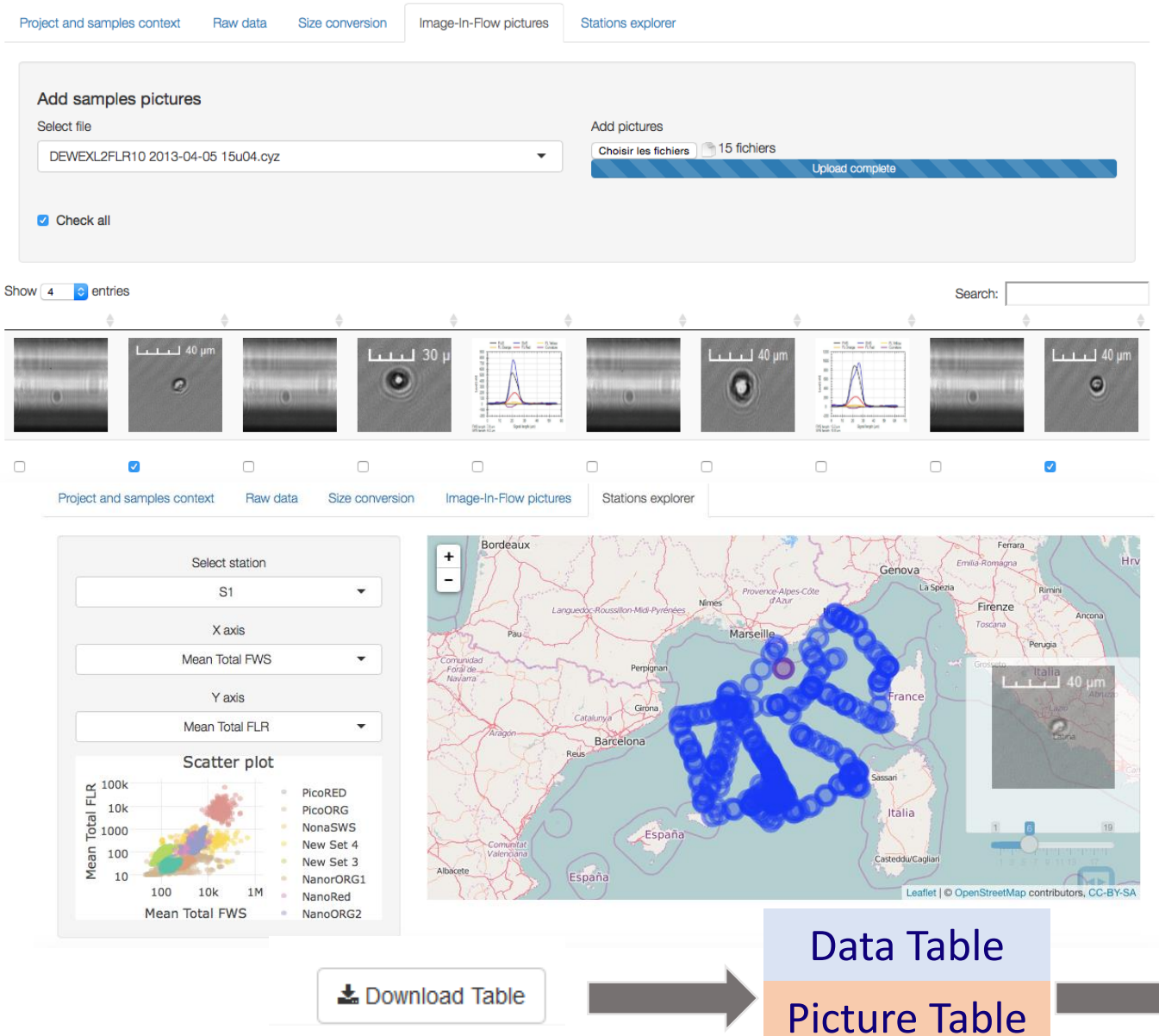
NB: All incompatible entries will be removed

Expert name	Trigger	PMT's amplification	Standardized name	
Cluster	Channel/Level	SWS	Cluster	
Beads 2 mu	FL Red 10	0 50 100	Standard beads	Associate
		FLO 0 50 100		

→ Data Consolidation

Picture selection

Stations Explorer



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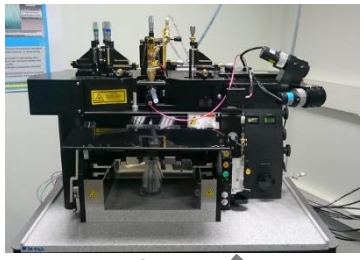
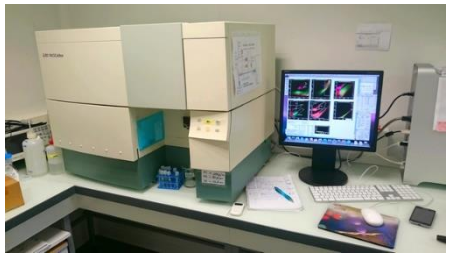
In situ **automated cytometer**

CytoSense

Bentchtop flow cytometers



BD Influx



FACS Calibur

CytoClus

Conversion tool
+ Metadata entry

M. Dugenne, 2015

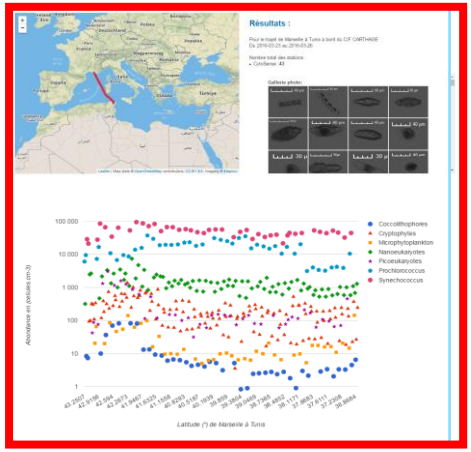
ASCII Data Table & Pictures

Acquisition

Analyses

Consolidation

Visual QC



Accessibility

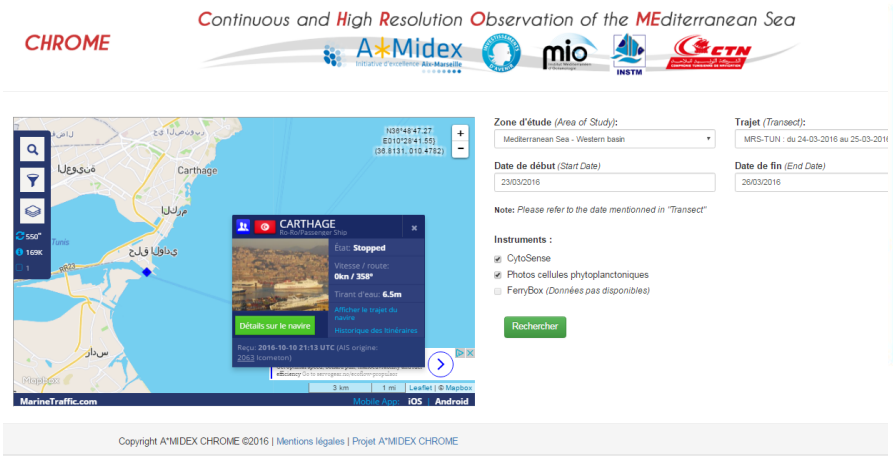
CYTOBASE

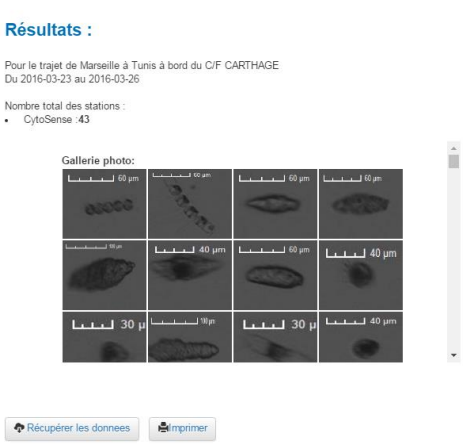
Integration

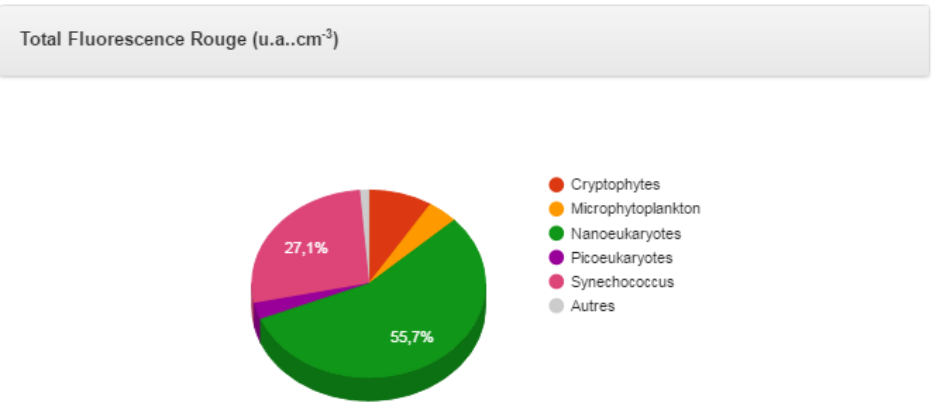
Visualisation

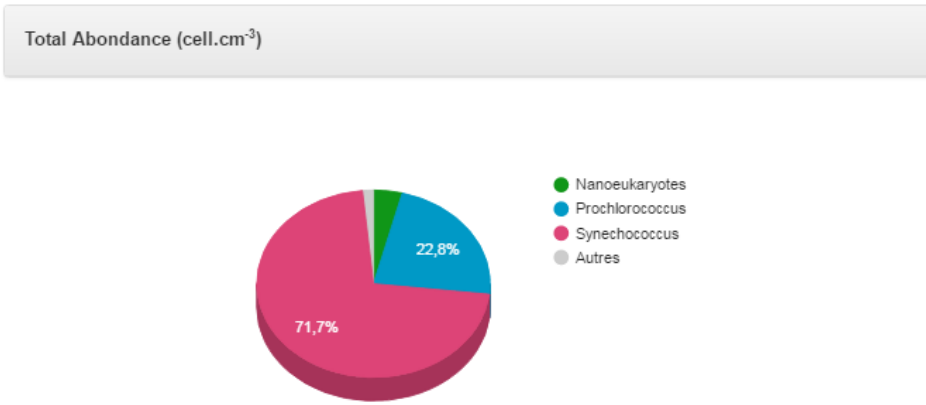
CYTOBASE FLOW CYTOMETRY DATABASE

https://chrome.mio.univ-amu.fr/chrome-cytobase/







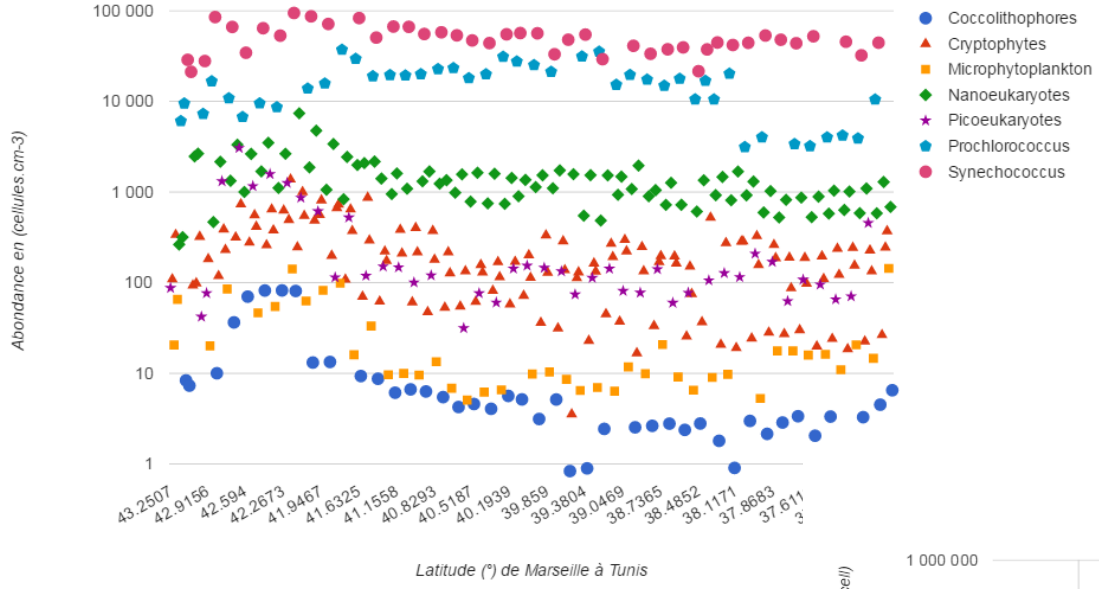


php Google Developers Leaflet dygraphs HTML CSS JavaScript MySQL AJAX jQuery

Visualisation

CYTOBASE

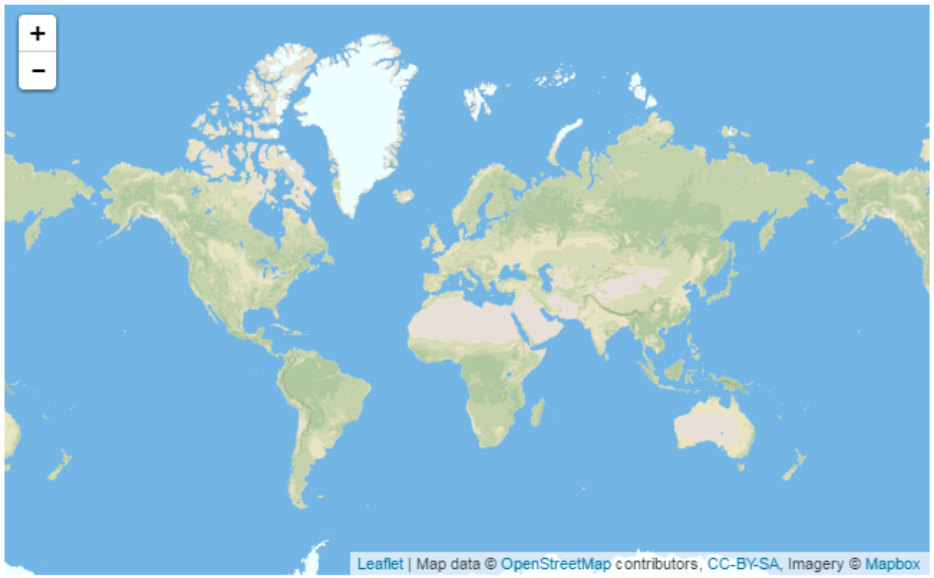
FLOW CYTOMETRY DATABASE



php Google Developers Leaflet dygraphs

CYTOBASE data accessibility (Access within the MIO)

FLOW CYTOMETRY DATABASE



Zone d'étude: Mediterranean Sea - Western basin

Nom et date du Projet: DEWEX LEG2 : du 01-04-2013 au 30-04-2013

Date de début: 31/03/2013

Date de fin: 30/04/2013

Flow cytometer :

- ○ CytoSense
- ◉ FACS Calibur
- ○ BD Influx
- ○ All instruments

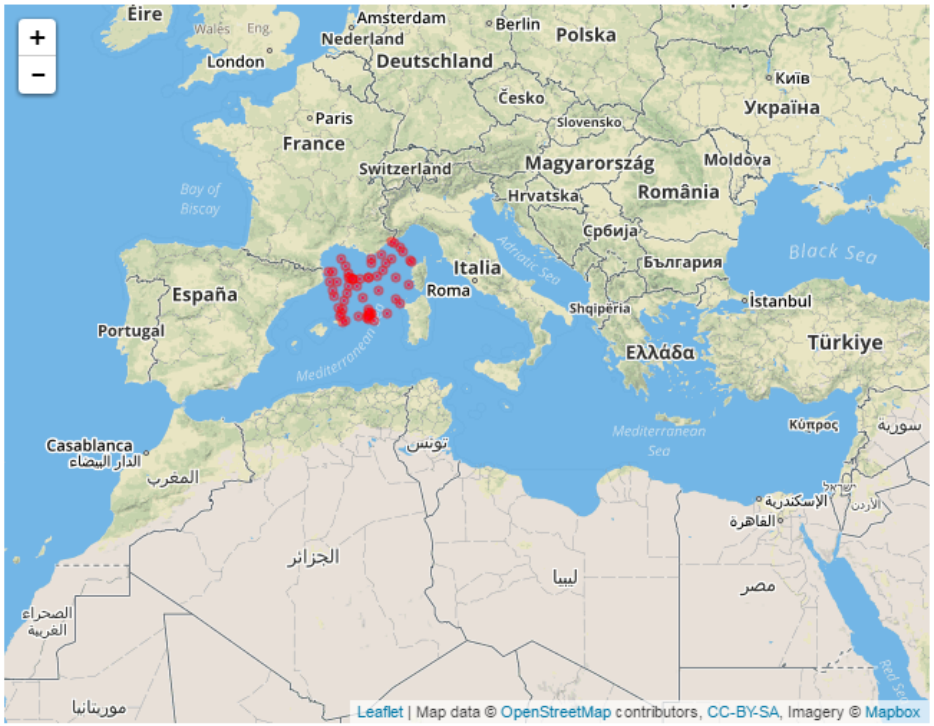
- ○ Phytoplankton groups
- ◉ Heterotrophic Bacteria

Search

http://www.mio.univ-amu.fr/cytobase/

CYTOBASE

FLOW CYTOMETRY DATABASE



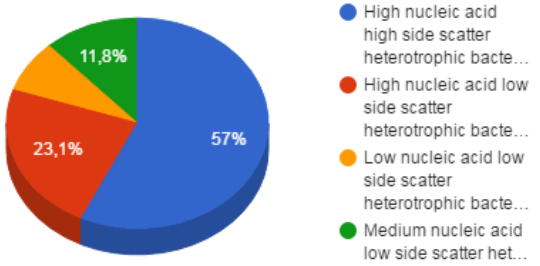
Results :

For the period from **2013-03-31 to 2013-04-30**
Project name **DEWEX LEG2**
Flow Cytometer : **FACS Calibur**
Station number : **59**

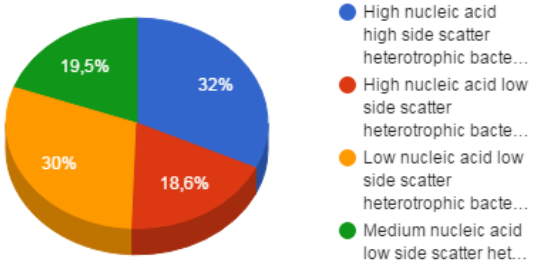
No pictures

Download Data | Print

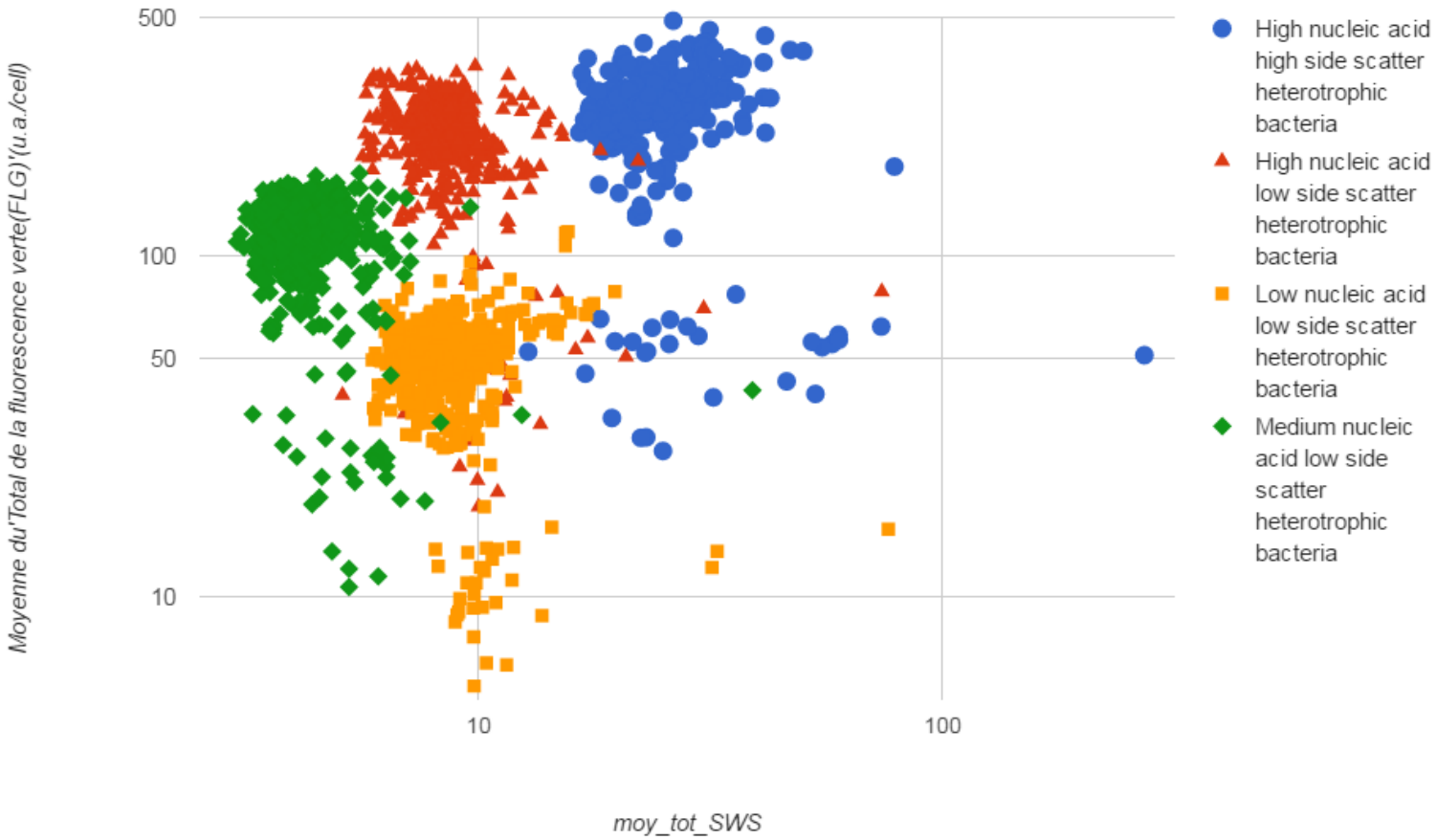
Contribution to Green Fluorescence (u.a..cm^{-3})



Relative Abundance (cell.cm^{-3})



Cytometric distribution parameters per group



Toward EU standardisation of FCM data

PI: Patrick FARCY (IFREMER)

PI: Michèle FICHAUT (SISMER/IFREMER)

Task 3.1: Automated platform for the observation of Phytoplankton diversity in relation to ecosystem services

WP9.5.2: Ingesting, validating, long-term storage and access of Flow Cytometer data

Leader: Felipe ARTIGAS (CNRS-ULCO)

Leaders: VLIZ, CNRS-MIO, NERC-BODC and ICES

Vocabulaire commun FCM

Toward EU standardisation of FCM data

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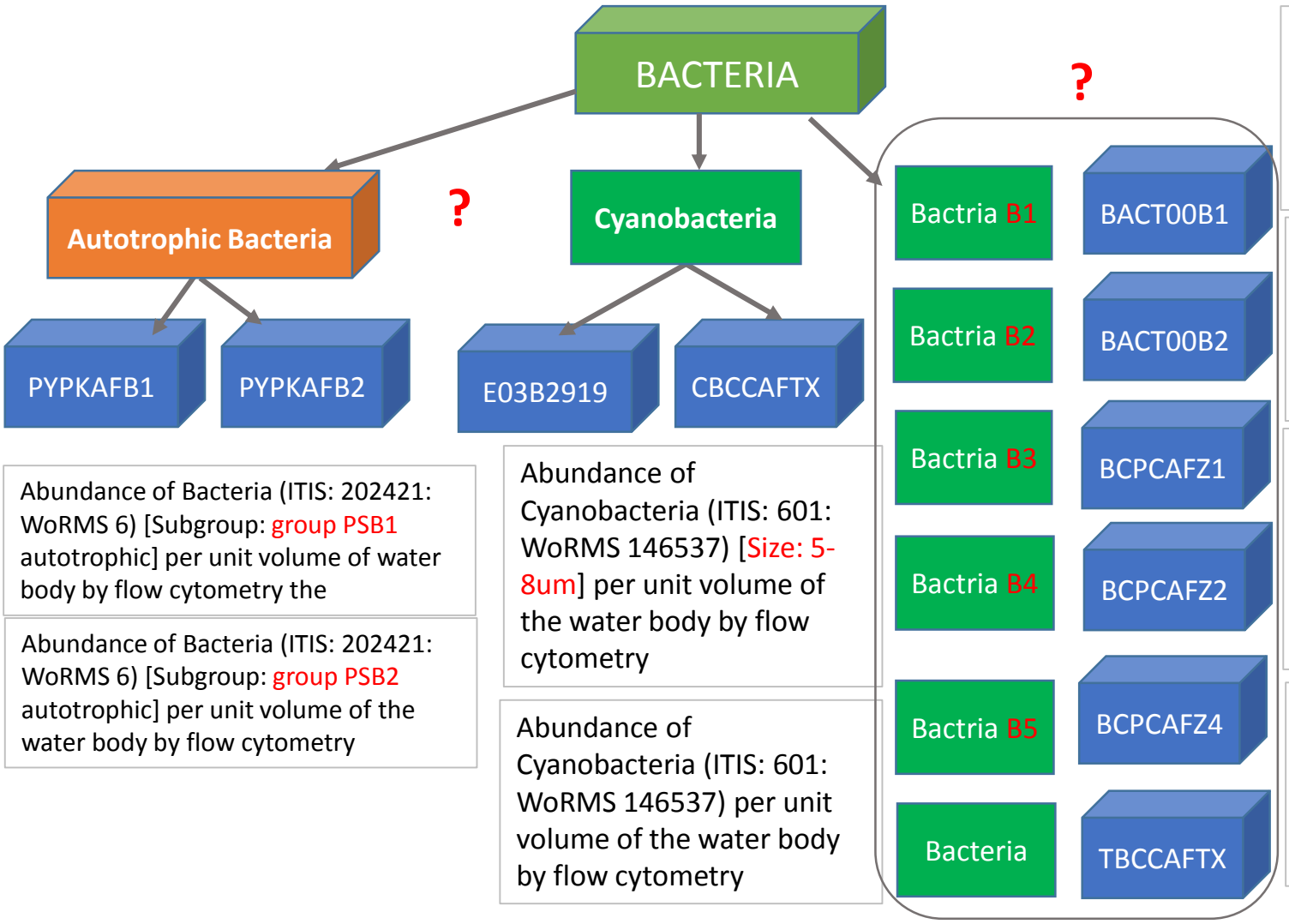
Analyse critique des 34 codes existants au NERC-BODC



Abundance of Bacteria (ITIS: 202421: WoRMS 6) [Subgroup: group PSB1 autotrophic] per unit volume of water body by flow cytometry the

Abundance of Bacteria (ITIS: 202421: WoRMS 6) [Subgroup: group PSB2 autotrophic] per unit volume of the water body by flow cytometry

Abundance of Cyanobacteria (ITIS: 601: WoRMS 146537) [Size: 5-8um] per unit volume of the water body by flow cytometry

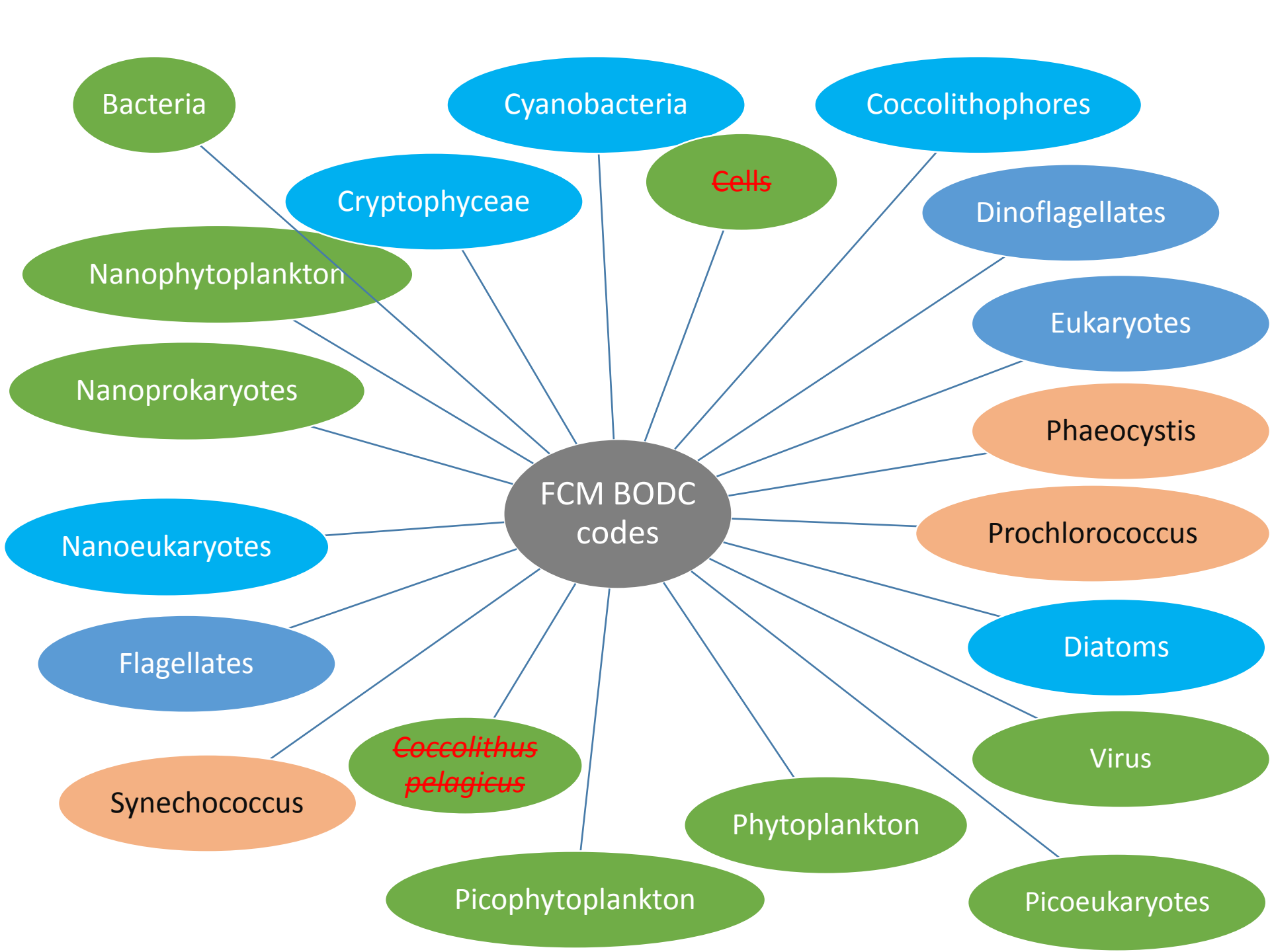
Abundance of Cyanobacteria (ITIS: 601: WoRMS 146537) per unit volume of the water body by flow cytometry

Abundance of Bacteria (ITIS: 202421: WoRMS 6) [Subgroup: group B1] per unit volume of the water body by flow cytometry

Abundance of Bacteria (ITIS: 202421: WoRMS 6) [Subgroup: group B2] per unit volume of the water body by flow cytometry

Abundance of Bacteria (ITIS: 202421: WoRMS 6) [Subgroup: Zubkov population 1] per unit volume of the water body by flow cytometry

Abundance of Bacteria (ITIS: 202421: WoRMS 6) per unit volume of the water body by flow cytometry


FCM BODC codes
Bacteria
Cyanobacteria
Coccolithophores
Cryptophyceae
~~Cells~~
Dinoflagellates
Nanophytoplankton
Eukaryotes
Nanoprokaryotes
Phaeocystis
Nanoeukaryotes
Prochlorococcus
Flagellates
Diatoms
~~Coccolithus pelagicus~~
Virus
Synechococcus
Phytoplankton
Picophytoplankton
Picoeukaryotes

Etude bibliographique de 137 articles (1983 – 2017)

Titre	Créateur	Année
Flow cytometry and cell sorting: A technique for analysis and...	Yentsch et al.	1983
Marine phytoplankton distributions measured using shipboa...	Olson et al.	1985
Discrimination between types of pigments in marine <i>Syn...	Wood et al.	1985
Effects of Environmental Stresses on the Cell Cycle of Two M...	Olson et al.	1986
Light and dark control of the cell cycle in two marine phytopl...	Vaulot et al.	1986
Effect of light on the cell cycle of a marine Synechococcus st...	Armbrust et al.	1989
Kinetics of bacterial processes in natural aquatic systems bas...	Button et Robertson	1989
Using phytoplankton and flow cytometry to analyze grazing ...	Cucci et al.	1989
A flow cytometric approach to assessing the environmental a...	Demers et al.	1989
Rapid analytical technique for the assessment of cell metabol...	Dorsey et al.	1989
Optical plankton analyser: A flow cytometer for plankton ana...	Dubelaar et al.	1989
Use of a neural net computer system for analysis of flow cyto...	Frankel et al.	1989
Change in Photosynthetic Capacity over the Cell Cycle in Lig...	Gerath et Chisholm	1989
Photobiology of natural populations of zooxanthellae from t...	Lesser	1989
Discrimination of eukaryotic phytoplankton cell types from li...	Olson et al.	1989
Optical plankton analyser: A flow cytometer for plankton ana...	Peeters et al.	1989
Flow cytometry and phytoplankton	Phinney et Cucci	1989
Algorithm to estimate cell biovolume using image analyzed ...	Sieracki et al.	1989
Role of light and the cell cycle on the induction of spermato...	Armbrust et al.	1990
The Rapid Analysis of Single Marine Cells by Flow Cytometry ...	Burkill et al.	1990
DNA polymorphism within the WH7803 serogroup of marine...	wood et Townsend	1990
Flow cytometric determination of phytoplankton DNA in cult...	Boucher et al.	1991
High-density photoautotrophic algal cultures: Design, constr...	Javanmardian et Palsson	1991
Survival of Aeromonas salmonicida in lake water.	Morgan et al.	1991
Phycoerythrins of Marine Unicellular Cyanobacteria. I.Bilin ty...	Ong et Glazer	1991
Growth and cell cycle of two closely related red tide-forming ...	Partensky et al.	1991
The picoplankton in Antarctic lakes of northern Victoria Land...	Andreoli et al.	1992
VERTEX: biological implications of total attenuation and chlo...	Broenkow et al.	1992
Interactions between marine bacteria and dissolved-phase an...	Button et al.	1992
Prochlorococcus marinus nov. gen. nov. sp.: an oxyphototro...	Chisholm et al.	1992
Analyzing multivariate flow cytometric data in aquatic scienc...	Demers et al.	1992
Evaluation of photosynthetic capacity in phytoplankton by fl...	Furuya et Li	1992
Quantifying heterogeneity: flow cytometry of bacterial cultur...	Kell et al.	1992

1. Les groupes les plus identifiés
2. Les définitions des groupes
3. Les paramètres les plus mesurés
4. Les capteurs les plus utilisés

Flow Cytometry vocabulary standardization Questionnaire

Flow Cytometry vocabulary standardization Questionnaire (FCM VSQ) is dedicated to identify the most common Flow Cytometry (FCM) Metadata and Data vocabulary used within the FCM community mainly involved in JERICO-NEXT (FP7 project) under the Task 3.1 on Automated platform for the observation of Phytoplankton diversity in relation to ecosystem services.

The aim of this questionnaire is to build a common standardized FCM vocabulary in order to better serve the marine research by understanding each other's data, sharing them and making them accessible through SeaDataNet which is a pan-European standardized infrastructure for managing a large and diverse marine data sets.

Part I: Flow Cytometer Metadata

What model of Flow Cytometer(s) do you use?

(e.g.: CytoSense, FACS Calibur)

Does your instrument have an image in flow device?

Yes No

(e.g.: pictures)

What is your sample inlet internal diameter (in micron)?

(Separate multiple entries with commas)

Which lasers wavelengths do you use (in nanometer)?

((e.g.:488). In case of multiple entries, separate them with commas)

Laser beam powers (in mW)?

((e.g.:25). In case of multiple entries, please indicate the power of each of your laser separated by commas.)

Part II: Sample Metadata

• What Beads reference do you use ?

(e.g.: brand, size, fluorescence, marterial)

• What beads diameters do you use ?

(e.g.: 1, 2, 3, 6, 10 um, ...)

• What are your Beads Fluorescences ?

(e.g.: Yellow)

• For what purpose do you use this instrument?

Check any that apply

- [ ] Research
- [ ] Monitoring
- [ ] Biotechnology
- [ ] Other:

• Where is your area of study?

Part III: Flow Cytometer Data

Do you use a fluorescent Dye?

- ○ Yes
- ○ No

Which type of particles do you measure?

Check any that apply

- ☐ Phytoplankton
- ☐ Heterotrophic bacteria
- ☐ Virus
- ☐ Other:

What are the recurrent autotrophic functional groups of your area of study?

Check any that apply

- ☐ Synechococcus
- ☐ Prochlorococcus
- ☐ Picoeukaryotes
- ☐ Nanoeukaryotes
- ☐ Coccolitophore
- ☐ Cryptophytes
- ☐ Microphytoplankton
- ☐ Not concerned
- ☐ Other:

What are the recurrent Heterotrophic groups of your area of study?

Check any that apply

Part IV: Groups definition from the FCM point of view

Based on litterature from 1983 to 2017, do you agree on these group definitions:

- **Prochlorococcus**

Prochlorococcus are defined as the smallest cyanobacteria found in marine environment. No staining is required to distinguish them by flow cytometry. FWS and FLR signatures are the smallest recorded up to now and require sensitive PMT or high powered lasers. The cluster, when well defined (often deep water communities) is below or may overlap that of Synechococcus group, and is often partially masked by the instrument background noise.
In samples stained for Heterotrophic bacteria analysis, Prochlorococcus can be distinguished using Sideward Scatter (SWS) vs Chlorophyll Red Fluorescences (FLR) cytogram. They do not emit orange fluorescence because they lack phycoerythrin.

Check any that apply

- ☐ I agree
- ☐ I do not agree

- **Synechococcus**

They are unicellular photosynthetic Cyanobacteria with flow-cytometry forward-scatter (FWS) and sideward scatter (SWS) signatures that are larger than those of most of marine heterotrophic bacteria. No staining is required to distinguish them by flow cytometry. The related cluster has higher FWS and red fluorescence (FLR) signatures than Prochlorococcus **and a distinct orange fluorescence (FLO) signature from their phycoerythrin accessory pigment when excited by lasers whose wavelength is below 533 nm. Cyanobacteria may contain phycocyanin, excited by a red laser and emitting above the chl a emission wavelength. The** Synechococcus **cluster is well resolved in red vs green (FLR/FLG) and in red vs orange fluorescences (FLR/FLO) cytogrammes. Due to their small size (0.8-1.2 µm) as reported in the literature,** Synechococcus **cells exhibit low intensity FWS, SWS and FLR signals.**

Check any that apply

- ☐ I agree
- ☐ I do not agree

- **Eukaryotes Picophytoplankton**

The common definition of this group is size dependent; i/e <2-3 µm. No staining is required to distinguish them by flow cytometry. The smallest known eukaryotic picophytoplankton is Ostreococcus**. Eukaryotic picophytoplankton exhibits a well defined flow cytometry signature, with FWS and FLR signals larger than that of** Prochloroccocus **and** Synechococcus,

Merci pour votre intérêt