BASICS

Bacterial single-cell approaches to the relationship between diversity and function in the Sea

Oslo-March 2003

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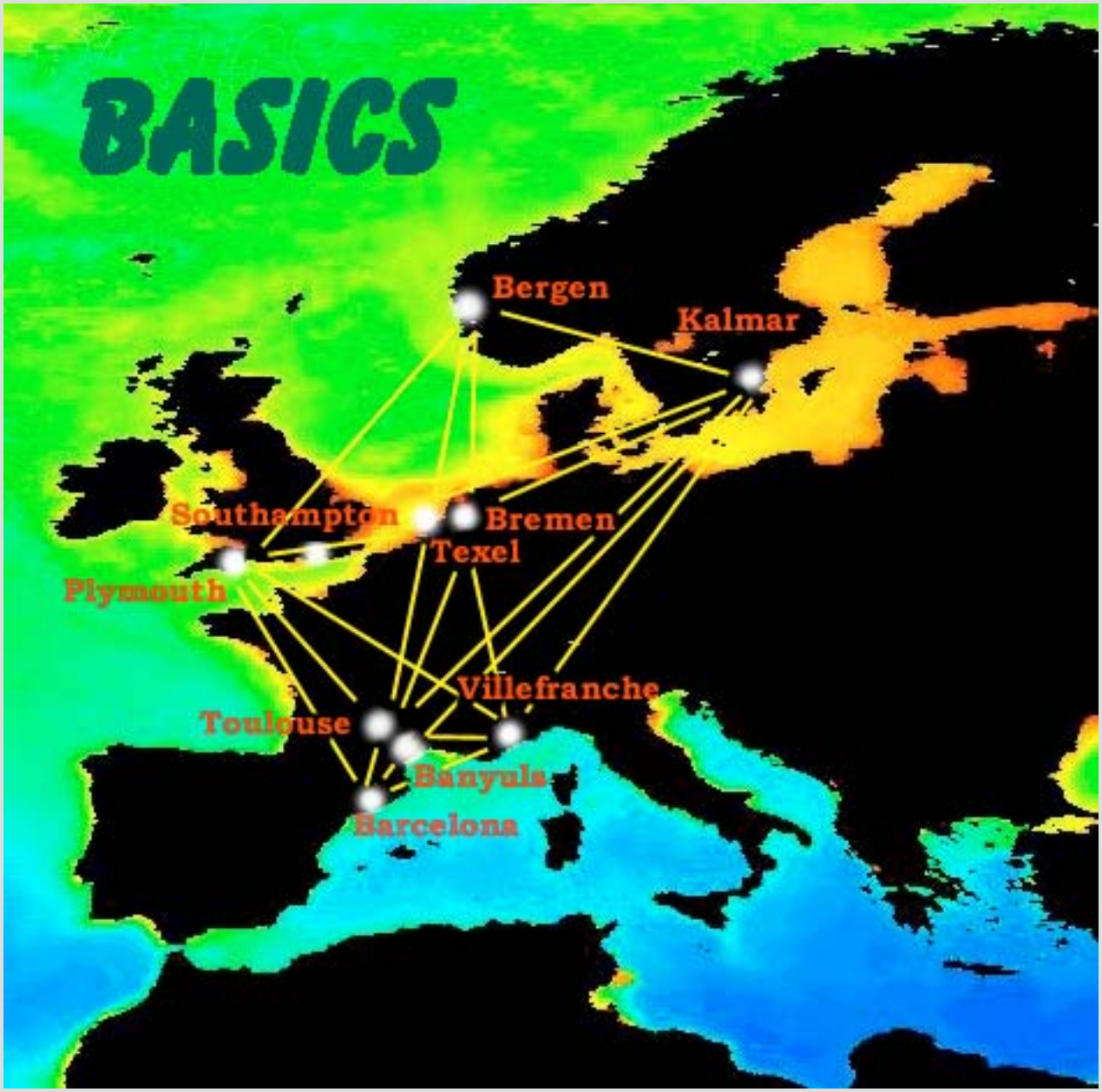
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Institut de Ciències del Mar-CMIMA

- Pep Gasol
- Rafel Simó
- Carlos Pedrós-Alió
- Ramon Massana
- Marta Estrada
- Jarone Pinhassi
- Laura Alonso
- Maria Vila
- Vanessa Balagué
- Clara Cardelús

Departament de Biologia Marina i Oceanografia
Department of Marine Biology and Oceanography
Departamento de Biología Marina y Oceanografía

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Observatoire Océanologique de Banyuls

- Philippe Lebaron
- Jean-François Ghiglione
- Philippe Catala

OBSERVATOIRE OCEANOLOGIQUE de Banyuls-Sur-Mer

Netherlands Institute of Sea Research

- Gerhard Herndl
- Karen Stoderegger
- Eva Sintes

NIOZ

Department of Biology. Kalmar University

- Åke Hagström
- Karen Simu
- Lasse Riemann

HÖGSKOLAN KALMAR UNIVERSITY

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Pierre Fabre Dermo-Cosmétique

- Pascal Bordat
- Muriel Bourrain

Lab. d'Océanographie de Villefranche

- Markus Weinbauer

Plymouth Marine Lab.

- Steve Archer

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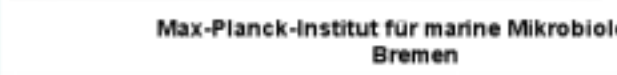
Max-Planck Institut for Marine Microbiology

- Jakob Pernthaler
- Bernard Fuchs
- Cecilia Alonso



Department of Microbiology. University of Bergen

- Frede Thingstad
- Ruth-Anne Sandaa
- Mikal Heldal

Southampton Oceanography Center

- Peter Burkill
- Mike Zubkov

Southampton Oceanography Centre
UNIVERSITY OF SOUTHAMPTON AND NATURAL ENVIRONMENT RESEARCH COUNCIL

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OBJECTIVE 1: To describe bacterioplankton diversity in the coastal seas of Europe

Describe bacterial diversity in the plankton of the coastal seas

The aims of the first objective are:

1.a Š To describe the seasonality in the presence of different bacterial phylotypes in the coastal Atlantic Ocean, coastal Mediterranean, Baltic Sea and North Sea.

1.b Š To compare the phylotypes present in these different seas and of their seasonal development.

1.c Š To compare the seasonal evolution of bacterial assemblages in nearby (< 500 km) sites in the Northwestern Mediterranean.

1.d Š To synthesize all the sequences of the 16S rRNA genes of cultured and uncultured marine bacteria currently stocked in the GeneBank, and to add as many new sequences as possible.

1.e Š To isolate as many marine bacteria as possible from different sites and characterize them physiologically and ecologically.

1.f Š To explore the biotechnological potential of marine planktonic bacterial isolates.

- Describe seasonality in “diversity” (what is there, who’s the most abundant)
- Usage of different techniques (fingerprinting &clon libraries & isolation...)
- Common framework
- Characterization of isolates

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OBJECTIVE 2: To describe the seasonal changes in the cycling of carbon and sulfur mediated by planktonic bacteria in surface waters of European coastal seas

Describe seasonal changes in the cycling of C and S mediated by bacteria

The aims of the second objective are:

2.a Š To describe the seasonality in the microbial composition and C and S species at selected sampling sites in the coastal Atlantic Ocean, coastal Mediterranean, Baltic Sea and North Sea.

2.b Š To describe the seasonal changes in the processes mediated by bacteria that contribute to the cycling of carbon in the surface coastal oceans.

2.c Š To describe the seasonal changes in the processes mediated by bacteria that contribute to the cycling of sulfur in the surface coastal oceans.

2.d Š To explore the link between sulfur and carbon cycles at the level of bacterial metabolism and transformation on a seasonal basis.

- Seasonal studies in C and S cycling
- Key biogeochemical steps little studied

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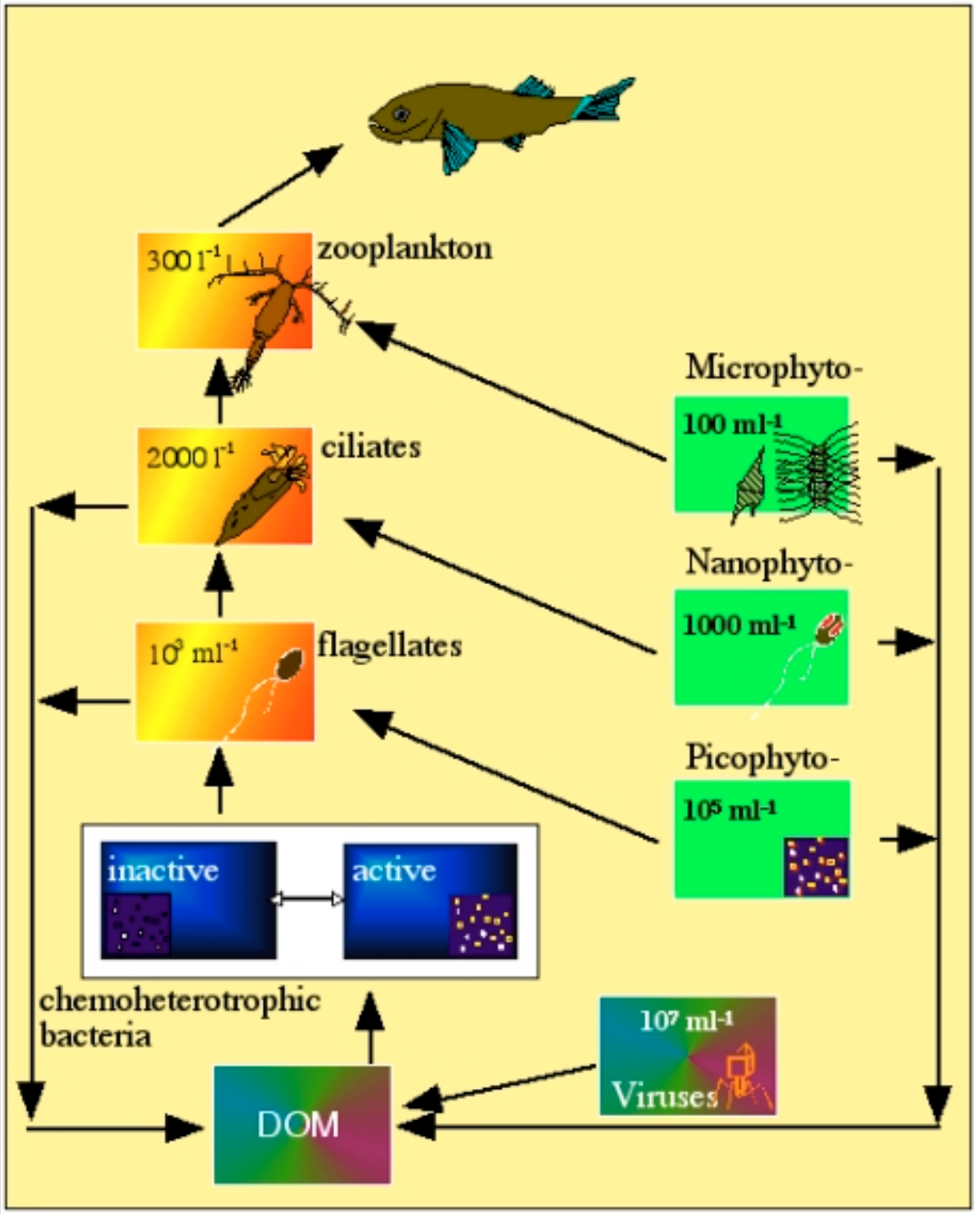

Fig. 2.1: The microbial food web in the oceanic plankton. Only pathways of organic carbon are presented, and the arrows connecting all organisms to viruses are omitted for clarity

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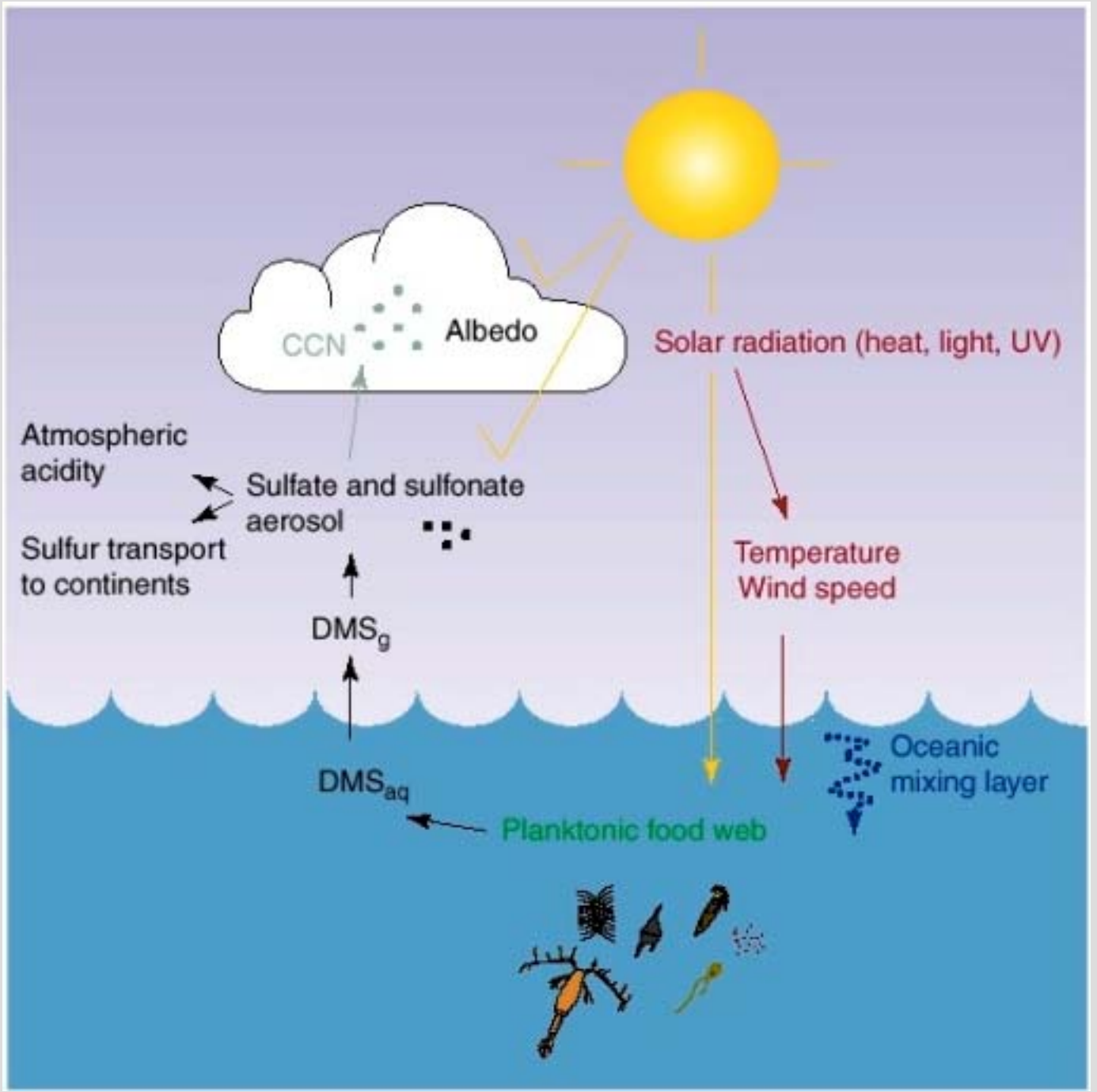

Fig. 2.2. The feedback linking oceanic plankton and climate through the production of atmospheric sulfur. The original hypothesis postulated that production of dimethylsulfide (DMS) by phytoplankton, and its subsequent ventilation and oxidation in the atmosphere feeds cloud condensation nuclei in marine stratus, thereby increasing cloud albedo. If the consequent reduction in solar irradiance forced phytoplankton toproduce less DMS, then a negative feedback would operate, thus stabilizing climate. Recent advances suggest that it is not only phytoplankton but the whole food web (with bacteria playing a crucial role) that releases DMS.

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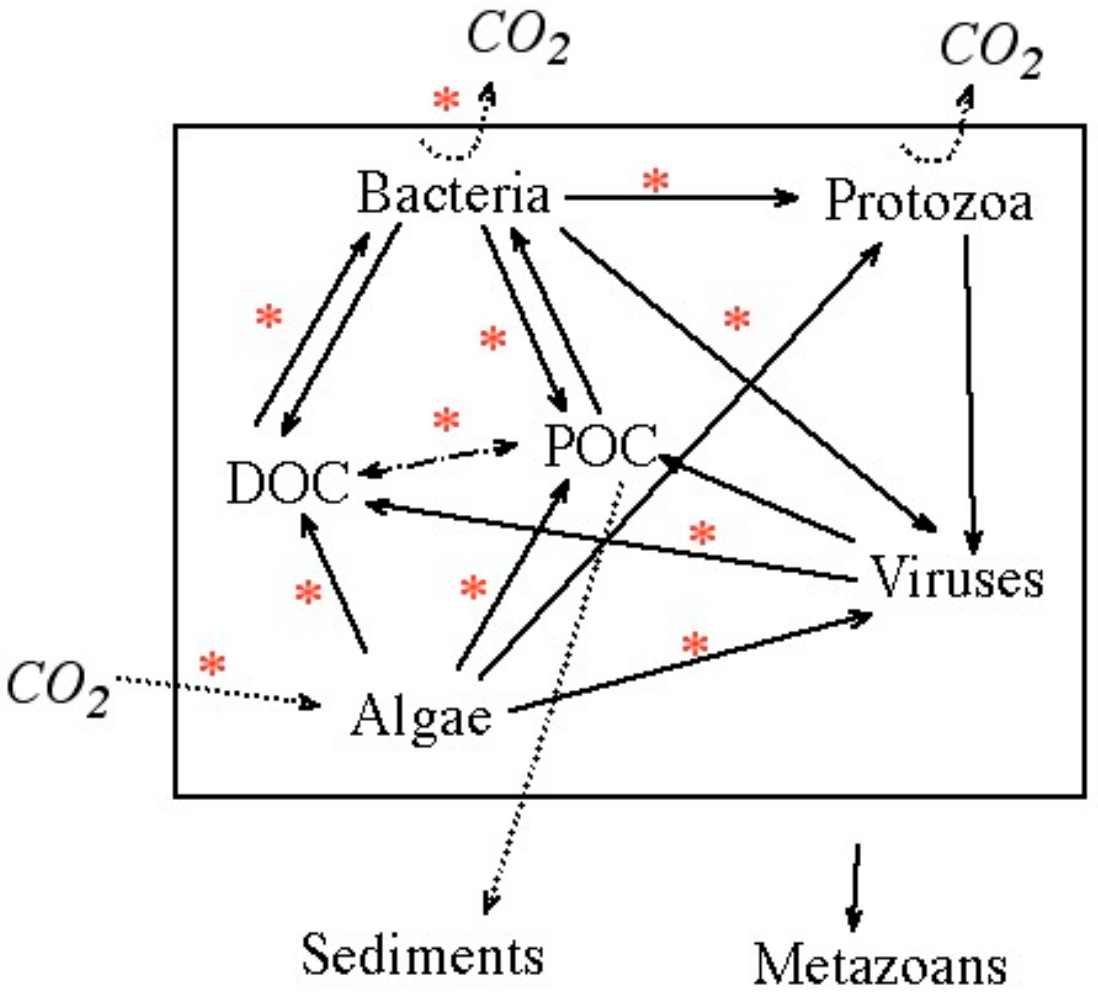
C-cycling in marine plankton



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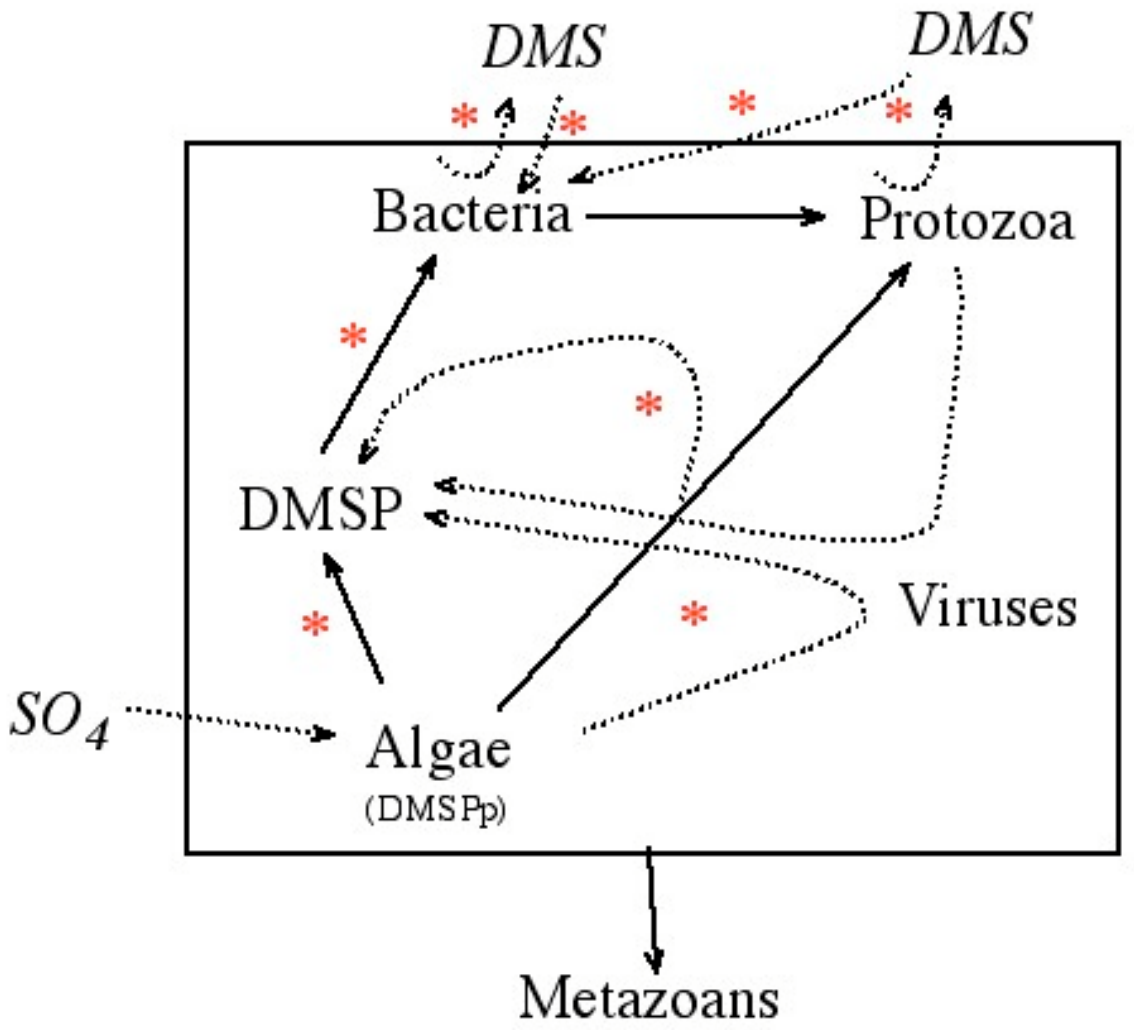
Volatile S-cycling in marine plankton



* processes under scrutiny in BASICS

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Sampling sites

NW Mediterranean
- Blanes Bay (ICM)
- Banyuls - MOLA & MILA station (OOB)
- Villefranche point “B” (LOV)

North Sea
- North Sea Texel site (NIOZ)
- Helgoland site G (MPIMM)

Other
- L4 English Channel station (PML & SOC)
- Baltic proper landsort (UNIK)

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OBJECTIVE 3: To design, test and fine-tune different methods and research strategies for the single-cell analysis of natural bacterioplankton organisms

Develop, adapt and fine-tune methods of SCA

This objective will be subdivided into two more explicit enunciates. The aims of the third objective are:

3.a Š To fine-tune, compare and standardize existing methods of Single-Cell Anlaysis (SCA).

3.b Š To develop new methods of SCA.

- Flow cytometry sorting standarization & controls
- FISH improvements
- MicroFISH, MicroACT
- Capillary electrophoresis
- X-Ray microanalysis

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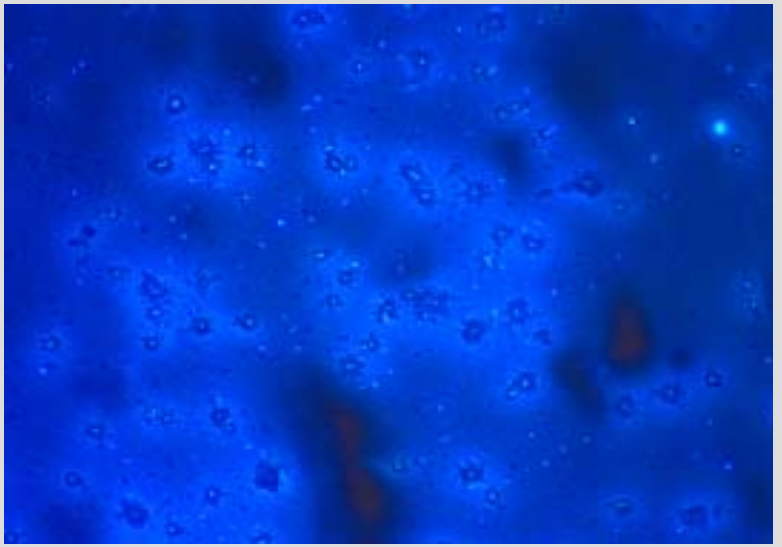
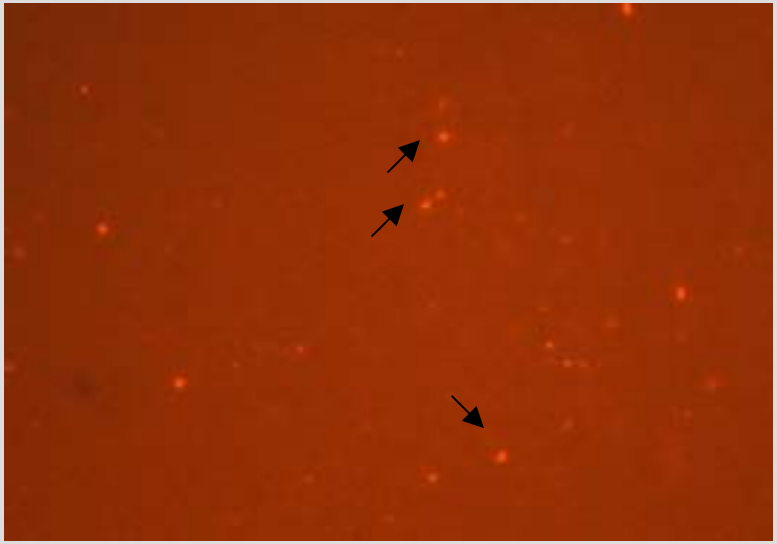
MicroFISH

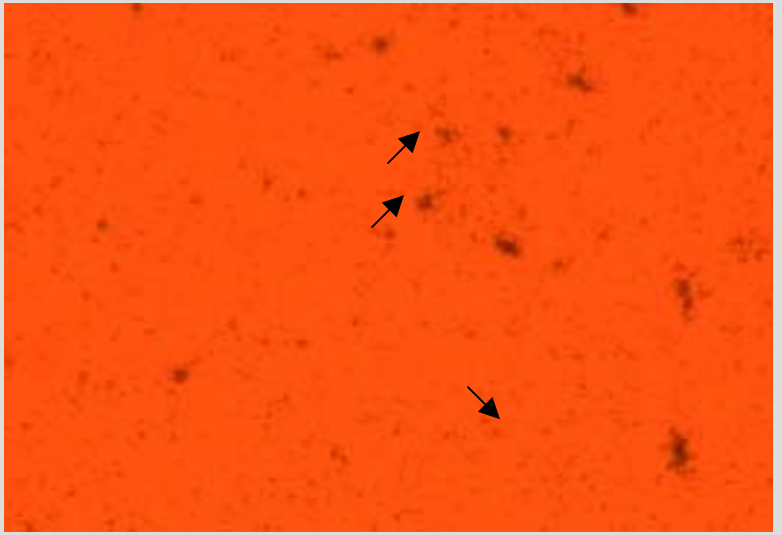
^{35}S DMSP

DAPI + AU



***Roseobacter* + AU**





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- CTC (5-cyano-2,3-ditolyl tetrazolium chloride)
  - ↕ → bacterial respiratory activity
- Autorradiography (glucose)
  - ↕ → Organic substrate uptake
- Eub338
  - → r RNA content
    Pos. correlated : AU *vs* EUB338 r^2=0.97

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OBJECTIVE 4: To link bacterial diversity and biogeochemical function (in the cycles of C and S) and identify the bacterial phylotypes responsible for the crucial steps in oceanic biogeochemical cycling, and to refine recently developed conceptual frameworks for the links between species richness (number of dominant coexisting species) and biogeochemical cycling

Link bacterial diversity and C and S cycling

The aims of the fourth objective are:

4.a Š To identify biogeochemically important bacterial phylotypes through correlation analysis of the seasonal variations in bacterial diversity and biogeochemical function, and perform specific experiments using SCA methods.

4.b Š Design ribosomal RNA-targeted nucleic acid probes to label specific phylotypes and analyze their seasonal development.

4.c Š To describe the ecological characteristics of the identified phylotypes using the SCA approaches developed.

4.d Š To describe the factors regulating the abundance and growth rates of the identified phylotypes.

4.e Š To study the loss factors (predation and viruses) that regulate the abundance of the identified relevant phylotypes.

4.f Š To refine recently developed conceptual frameworks for the links between species richness (number of coexisting species) and biogeochemical cycling and compare the predictions of such a theoretical framework to the data obtained in this project.

Multistep strategy
All approaches are partial and risk failure

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OBJECTIVE 5: To estimate the effect of environmental changes affecting the oceanÕs bacterially-mediated biogeochemical function, global bacterial diversity and the link between bacterial diversity and C and S cycling

Response of bacterial diversity and biogeochemical function to environmental perturbations

We will restrict the study of environmental change to those changes affecting the surface ocean: increased temperatures, increased UV exposure, changed seawater pH and increased allochthonous DOM supply. The aims of the fifth objective are:

5.a Š To describe the effect of changed temperature, UV, pH and DOM loading on total bacterial-community diversity.

5.b Š To describe the effect of changed temperature, UV, pH and DOM loading on bacterialy-mediated C and S cycling.

5.c Š Study the resiliency of the link diversity-function to changed temperatures, UV, pHs and DOM loading.

- Functional redundancy, ecosystem stability...
  ... effects of env. change on
  - diversity
  - BGQ function
  - their linkage
- Environmental perturbations in microcosms

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WS0: *Coordination*

WP1: Seasonal studies of bacterial diversity

WP3: Seasonal studies of biogeochemical C & S cycling

WP 4: SCA method development

Phase 1

WP2: Explotaition of phylotypes and isolate information

WS1: *Synthesis WS on bacterial diversity in coastal european seas and C & S cycling*

WP5: Linking bacterial diversity with biogeochemical function

Phase 2

WP6: Experimental determination of the factors that regulate the link between bacterial diversity and function

WS2: *Mesocosm experiment*

WS3: *Synthesis WS on SCA*

WP7: Functional stability of the link facing global change

WS4: *Microcosm experiments*

Phase 3

WS5: *Summary*

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Workpackage number	Task	Year 1												Year 2												Year 3											
		1	2	3	4	5	6	7	8	9	10	11	12	1	2	3	4	5	6	7	8	9	10	11	12	1	2	3	4	5	6	7	8	9	10	11	12
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	Task 1.2																																				
	Task 1.3																																				
	Task 1.4																																				
WP2	Task 2.1																																				
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WP4	Task 4.1																																				
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WP5	Task 5.1																																				
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WP6	Task 6.1																																				
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	Task 6.3																																				
	Task 6.4																																				
	Task 6.5																																				
WP7	Task 7.1																																				
	Task 7.2																																				

Workshop number		Year 1												Year 2												Year 3											
		1	2	3	4	5	6	7	8	9	10	11	12	1	2	3	4	5	6	7	8	9	10	11	12	1	2	3	4	5	6	7	8	9	10	11	12
0	Startup																																				
1	Synthesis																																				
2	Experimental																																				
3	Experimental																																				
4	Synthesis																																				
5	Final																																				