



# SILICAMICS

Biogeochemistry and genomics  
of silicification and silicifiers

21-25 September 2015

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Brest, France

Centre de la Mer de l'Aber-Wrac'h, Landéda

<http://silicamics.sciencesconf.org/>

# SILICAMICS

The conference aims to develop a transdisciplinary approach (including chemistry, biogeochemistry, biochemistry, physiology, genomics) to better understand and model silicification and silicifiers.

Silicifiers are among the most important living organisms of planet Earth. They are able to take advantage of the abundance of silicon (silicon is the second-most-abundant element in the Earth's crust) to build silicified architectures, which in particular can help for protection against predators, for motility, or for facilitating the penetration of light and nutrients to the cells.

This transdisciplinary conference focuses on the marine realm, for which numerous unknowns remain regarding the global marine silica cycle. Marine diatoms vs. siliceous sponges and radiolarians rose to prominence about 100 Myears ago. Today diatoms play a key role in the trophic networks of the most productive coastal or open-ocean ecosystems, as well as in the biology-mediated transfer of CO<sub>2</sub> from the surface to the ocean interior (the so-called biological pump of carbon). The physiology and biochemistry of pelagic diatoms have been extensively studied but many gaps remain regarding the silicification mechanisms and their variations due to environmental changes. The link between Si and C cycles also needs to be re-evaluated using newly developed genomic and geochemical tools. Moreover, benthic diatoms and their role in coastal ecosystems have been largely overlooked despite their 40% contribution to coastal primary production. Along the same vein, the key role of other siliceous organisms, such as benthic sponges, in the silica cycle has only been demonstrated at regional scales and needs to be quantified at the global scale. Accumulation of silica by non siliceous picocyanobac-

teria has been evidenced but the reasons and mechanisms behind such accumulation are still unidentified. Similarly, the formation of siliceous compounds in some flagellates of the nanoplankton remains to be quantified. The understanding of all these processes closely associated with the metabolism of such a diversity of silicifiers should now benefit from the use of genomics.

Since 2000 the genomes of several diatom species have been sequenced and genomics can now be used to formulate new hypotheses and research strategies for explaining the role of different silicifiers in coastal and open-open ecosystems, and their control of C, N, P, and Si biogeochemical cycles. More knowledge must now be acquired about interactions between diatoms and other organisms, at the cell/species level, and their consequences for nutrient cycles and ecosystem functioning. Genomics can help understanding such processes, and the availability of new resources, (such as from the Tara Oceans expedition and from the Gordon and Betty Moore Foundation's MMETSP programme) make this a pivotal moment to exploit new opportunities to explore the biology of ocean silicification processes.

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## PROGRAM

### Monday 21 September 2015

17:00-19:00	arrival of participants and installation of the poster session for the week
19:00	cocktail

### Tuesday 22 September 2015

9:00 - 09:05	Welcome speech by <b>Aude Leynaert</b> (IUEM, Brest, France)
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#### General Introduction

09:10-10:10	<b>Chris Bowler</b> (ENS, Paris, France) - Genomics -Enabled exploration of the secrets of marine diatoms
10:10-10:40	Coffee break
10:40-11:40	<b>Paul Tréguer</b> (IUEM, Brest, France) - Silica cycle of the world ocean: knowns and unknowns
11:40-12:40	<b>Colomban de Vargas</b> (SBR, Roscoff, France) - Evolution of silicifiers (the great picture)
12:40 -14:00	lunch

#### Session 1: Silicification (processes and properties) in diatoms

14:00-14:40	<b>Pascal Jean Lopez / Jacques Livage</b> - General introduction Principe of silicon biomineralization and impacts of environmental factors
14:40-15:00	<b>Brivaëla Moriceau</b> (IUEM, Brest, France) - Impact of growth stress on diatom BSiO <sub>2</sub> recycling

15:00 – 15:20	<b>Jacques Livage/Sophie Cassaignon</b> (Collège de France, Paris, France) - Photonic cristal properties of diatoms frustule
15:20 – 15:40	<b>Paul Curnow</b> (Bristol University, UK) - Molecular studies of diatom silicic acid transporters
15:40 -16:10	coffee break

## Session 2: Silicification in other silicifiers

16:10 -16:50	<b>S. Baines</b> (Bigelow, USA) and <b>M. Maldonado</b> (CSIC, CEAB, Spain) - General introduction: The diversity of silicifiers in nature
16:50 -17:10	<b>Manuel Maldonado</b> (CSIC, CEAB, Spain) - The contribution of sponges to the marine silicon cycle
17:10 -17:30	<b>Taniel Danelian</b> (Université de Lille 1, France) - Silicifying radiolarians: a brief biological, oceanographic and taphonomic overview
17:30 - 17:50	<b>Stephen Baines</b> (Bigelow, USA) - A role for the cyanobacterium, <i>synechococcus</i> , in the marine silicon cycle
18:00- 19:00	Working Group for students
19:30	dinner

## Wednesday 23 September 2015

### Session 3: Genomics/proteomics as tools for silicifiers

08:30 - 09:10	<b>Chris Bowler</b> (ENS, Paris, France) - General introduction
09:10 - 09:30	<b>Peter Kroth</b> (University of Konstanz, Germany) - Molecular tools to study benthic diatom communities
09:30 - 09:50	<b>Mark Hildebrand, Sarah Lerch</b> (SIO, USA) - Identification and comparison of silicification genes in diatoms
09:50 - 10:10	<b>Kim Thametrakoln</b> (Rutgers University, NJ, USA) - Linking physiological and molecular aspects of diatom silicification
10:10 - 10:30	<b>Andrew Allen</b> (J. Craig Venter Institute, La Jolla, USA) - Phytoplankton- bacterial coupling mediates micronutrient limitation in the Southern Ocean
10:30 -11:00	coffee break

### Session 4: A biogeochemical/genomic approach for ecosystems modelling

11:00 - 11:40	<b>Marion Gehlen</b> (LSCE, Gif-sur-Yvette, France) - General introduction - Genomics: a new data stream to inform biogeochemical models?
11:40 - 12:00	<b>Olivier Aumont</b> (LOCEAN, Paris, France), <b>F. Diaz</b> , (MIO, Marseille, France) - Representing autotrophs silicifiers in Plankton Functional Types models – Application to the Northwestern Mediterranean Sea
12:00 - 12:20	<b>Markus Pahlow</b> (GEOMAR, Kiel, Germany) - Optimality-based view on phytoplankton and Si: how is silicification regulated and what is its role in phytoplankton succession?
12:20 - 12:40	<b>Christopher Algar</b> (Dalhousie University, Halifax, NS, Canada) - Approaches for integrating genomic data and biogeochemical models.
13:00 -14:30	Lunch



## Session 5: Isotope chemistry providing tools for processes and fluxes

14:30 - 15:10	<b>Kate Hendry</b> (University of Bristol, UK) and <b>Jill Sutton</b> (IUEM, Brest, France) - General introduction - The zinc isotopic composition of siliceous marine sponges: investigating nature's sediment traps
15:10 - 15:30	<b>Jill Sutton</b> (IUEM, Brest, France) - Silicon isotopes and diatoms in culture studies
15:30- 15:50	<b>Patricia Grasse</b> (GEOMAR, Kiel, Germany) - What controls the silicon isotope signature of biogenic silica?
15:50 - 16:10	<b>Luc André, Camille Delvigne</b> - (Royal Museum for Central Africa, Belgium) The Si-Mg isotopic compositions of the Juan de Fuca ridge low temperature hydrothermal fluids and the Si fluxes from the off-axis rock-water interactions
16:10-16:40	Coffee break
16:40 - 17:00	<b>Stefan Lalonde</b> (IUEM, Brest, France) - The Ge isotopic composition of seawater: First depth profiles (Southern Ocean) and estimated fractionation between seawater and biogenic opal
17:00 - 17:20	<b>Michael Ellwood</b> (Australian National University, AU) - Evidence of increased silicic acid leakage from the Southern Ocean during the glacial and de-glacial periods
17:20-17:40	<b>Morten Andersen</b> (University of Bristol, UK) - Zinc isotopes and diatoms in the southern ocean
18:00- 19:00	Working Group for students
19:30	Banquet

## Thursday 24 September 2015

### Session 6 : The silica cycle: key players and processes

8:30 - 9:10	<b>Bernard Quéguiner</b> (MIO, Marseille, France) - General introduction - Reflections on the role of silicifiers in oceanic biogeochemical cycles - conceptual approaches and research perspectives
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### Session 6a : Coastal ecosystems

09:10 - 09:30	<b>Aude Leynaert</b> (IUEM, France) - Benthic versus pelagic diatoms
09:30 - 09:50	<b>Patricia Glibert</b> (University of Maryland, USA) - The success of diatoms: the nitrogen perspective.
09:50 - 10:10	<b>Dan Conley</b> (University of Göteborg, Sweden) - The Global Si Cycle: Variable inputs into the oceans and possible changes in the Si cycle through geologic time.
10:10 - 10:40	Break
10:40 - 11:00	<b>Michalopoulos Panagiotis</b> (HCMR, Athens, Greece) - Early diagenesis of silica and reverse weathering in marine sediments: biological controls and interactions with organic carbon cycling and preservation.
11:00 - 11:20	<b>Olivier Ragueneau</b> (IUEM, Brest, France) - Si cycle and the coastal zone: what about man in this story?

### Session 6b: Open ocean ecosystems

11:20 - 11:40	<b>Séverine Alvain</b> (LOG, Lille, France) - Phytoplankton detection from space based on the synergy between ocean colour and <i>in situ</i> observations: principles and perspectives
11:40 - 12:00	<b>Dave Nelson</b> (IUEM, Brest, France) - Diatom strategies for persistence, growth and reproduction in the eastern equatorial Pacific: avoidance of nutrient limitation vs bloom-and-burst
12:30 -14:00	Lunch
14:00 - 14:20	<b>Akira Kuwata</b> (Japan) and <b>Daniel Vulot</b> (Station Biologique de Roscoff, France) - Parmales and Bolidophyceae, a picoplankton group closely related to diatoms

14:20 - 14:40	<b>Marine Lasbleiz</b> and <b>Bernard Quéguiner</b> (MIO, Marseille, France) - Interspecific variability of silicifying activity among dominant diatoms of the phytoplankton community in the Southern Ocean (KEOPS 2 project)
14:40 - 15:00	<b>Jeffrey Krause</b> (USA, Alabama, USA) - Quantitative determination of group-specific Si uptake in field diatom assemblages
15:00 - 15:20	<b>Soumaya Boussabat</b> (MIO, Marseille, France) - Different scales of variability of the elemental composition C/ N /P/ Si of diatoms across the Gulf of Lion, north-western Mediterranean
15:20 - 15:40	<b>Richard Dugdale</b> (Romberg Tiburon Center, San Francisco, USA) - Silicate pumping of excess carbon in coastal upwelling ecosystems
15:40 - 16:10	Coffee break
16:10 - 17:00	<b>Aude Leynaert</b> , <b>Brivaela Moriceau</b> and <b>Paul Tréguer</b> (IUEM, Brest, France) - general discussion ; new initiatives to help for the convergence of biogeochemistry and genomics on silicification and silicifiers.
17:00 - 19:00	Working Group for students
19:30	Dinner

## Friday 25 September 2015

8:30 - 10:30	Students working group report
10:45	End of the conference

# SILICAMICS

## ABSTRACTS

### GENOMICS: ENABLED EXPLORATION OF THE SECRETS OF MARINE DIATOMS

Chris Bowler

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Diatoms are thought to be the most successful group of eukaryotic phytoplankton in the modern ocean. Recently completed whole genome sequences have revealed a wealth of information about the evolutionary origins and metabolic adaptations that may have led to their ecological success. A major finding is that they have acquired genes both from their endosymbiotic ancestors and by horizontal gene transfer from marine bacteria. This unique melting pot of genes encodes novel capacities for metabolic management, for example allowing the integration of a urea cycle into a photosynthetic cell. We explore both the physiological functions of diatom gene products and the evolutionary mechanisms that have led to diatom success in contemporary oceans. Specific research topics that we are currently addressing are:

- How has diatom evolution enabled interactions between chloroplasts and mitochondria that have provided diatoms with physiological and metabolic innovations, and
- What are the relative contributions of DNA sequence variation and epigenetic processes in diatom adaptive dynamics? Additionally, the abundance, diversity, and distribution of diatoms in the global ocean is being explored using data from the Tara Oceans expedition, a 3.5 year global sampling of marine planktonic ecosystems that has collected more than 35,000 biological samples from all major oceanic basins, together with extensive environmental data.

### THE WORLD OCEAN SILICA CYCLE: KNOWN AND UNKNOWN

Paul Tréguer

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The silica cycle in the world modern ocean has been recently revisited by Tréguer and De La Rocha (2013). In this study the importance of new pathways for providing silicic acid to the oceans are highlighted, like the submarine groundwaters, or the in situ dissolution of siliceous material transported from the land to continental margins. The steady state of this cycle in the modern ocean is questioned. Indeed, the net silicic acid annual inputs to the world ocean can be balanced by the annual sink flux of opal, only if the flux due to siliceous sponges is taken aboard. However, as regards to sponges, both the uptake rates of silicic acid and dissolution rates of biogenic silica are not well documented. The silica fluxes due to radiolarians, silicoflagellates, and to benthic diatoms in the coastal zone, have not been quantified at world ocean scale. In addition, so far the role of picocyanobacteria or that of picophytoplankton in the silica cycle has been neglected. However, on the one hand accumulation of biogenic silica by picocyanobacteria in the open ocean has been demonstrated, although the processes that control this accumulation are not yet understood. On the other hand, evidence has been given that siliceous deposits can occur on extracellular polymeric substance associated with decomposing picophytoplankton and that Si is enriched in a previously unexplored group of marine particles (called micro - blebs) from the deep - water column.



## SESSION 1: SILICIFICATION (PROCESSES AND PROPERTIES) IN DIATOMS

### GENERAL INTRODUCTION

#### PRINCIPE OF SILICON BIOMINERALIZATION AND IMPACTS OF ENVIRONMENTAL FACTORS

Jacques Livage<sup>1</sup> and Pascal Jean Lopez<sup>2</sup>

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Silicon and oxygen are the two most abundant elements on earth. About 80% of the earth crust and mantle is made of silicates. It is therefore rather surprising that most solid biomaterials such as shells, bones or teeth are made of calcium carbonate or phosphates rather than silica. Actually, silica glasses are very common materials. They are widely used for making bottles, windows or fibers. However, since their discovery about 5.000 years ago, silica glasses have always been made from molten sand at temperatures well above 1000°C. Even natural glasses such as obsidian are formed at high temperature from volcanic lava. Despite the fact that such high temperatures are not compatible with life, silicon is involved in the metabolic activity of many living organisms. Biogenic glasses have therefore to be made at room temperature, from soluble silica species dissolved in the aqueous environments. They can be found in many aquatic and terrestrial organisms such as planktonic species, sponges and land plants. Biogenic silica structures with precisely controlled morphologies are produced on a huge scale by single-cell organisms such as diatoms. However, the morphology of these structures is sensitive to environmental factors, which might impact the formation process. Nevertheless they exhibit sophisticated nanostructures and open new possibilities for green chemistry and nanotechnologies.

#### IMPACT OF GROWTH STRESS ON DIATOM BSIO<sub>2</sub> RECYCLING

Brivaëla Moriceau\*, Julia Bouthorh, Charlotte Soler, Morgane Gallinari

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The silicic acid availability determines the occurrence of diatom bloom, and as diatoms are main vessels of the carbon to the deep ocean, influences also the biological pump of carbon and its variability. Remineralization of diatom frustules in the surface layer is an important source of silicic acid. Despite the importance of this process, dissolution is still poorly represented: while it has long been recognized that dissolution is a two-steps process, the community generally uses a simple exponential equation. To ameliorate this representation, we tried to understand the mechanisms involved by using long term experiments on fresh diatom. Impact of external factors on bSiO<sub>2</sub> dissolution has been studied for many years but silicification is overall a biological process. Diatom silicification is sensitive to stresses encountered during growth in most oceanic areas like nutrients or light limitations, and like grazing. By monitoring the dissolution of diatom frustules formed under different nutrient conditions, we had 2 objectives. We expected to better understand the mechanisms involved in the 2-steps dissolution and secondly, we expected to address the impact of nutrient limitation on bSiO<sub>2</sub> dissolution. Our results suggested that two different biogenic silica phases coexist in the frustule. These two phases have different dissolution rate constants, and both are affected by nutrient limitation. Moreover the proportion of each phase that constitutes the frustule also changes when diatoms have grown under different nutrient limitations. Overall, nutrient limitations tested in this study, tend to increase the preservation of the biogenic silica formed.

## PHOTONIC CRISTAL PROPERTIES OF DIATOMS FRUSTULE

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Single-celled algae called diatoms are enclosed in a nanostructured amorphous silica skeleton which is composed of two valves. For each species of diatoms, valve surface exhibits specific pattern of regular arrays of holes called areolae. Their dimensions are varying from a few hundreds of nanometers to a few micrometers. The structure of these pores allows the frustule playing a role of an optical lens, whose focal length is specific to the species of diatom considered. Indeed, according to the depth where they live, they filter differently the sun wavelengths. When the structure of the holes is periodic, the frustules acquire photonic crystal properties, and then selective specific wavelengths can be selected. Artificial photonic crystals have already been widely used in solar cells to guide the light and to increase the photon absorption. In the present work, we study the optical properties of different diatoms species, in order to realize the coupling with a photovoltaic device like luminescent sheet concentrator (LSC) solar cells for example. We study several diatoms species with various diameters from 20  $\mu\text{m}$  to 500  $\mu\text{m}$ . They are cultivated in specific solutions before the organic material is removed and morphologic characterizations are realized by MET, MEB. To achieve the association between diatoms and solar cell, it is necessary to realize deposition of thin films containing diatoms. Different methods are investigated, especially dip-coating and electrophoresis. In parallel, an optical set-up is developed to study the properties of the light transmitted or reflected by a frustule thin layer.

## MOLECULAR STUDIES OF DIATOM SILICIC ACID TRANSPORTERS

Paul Curnow

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Diatom silicification depends upon the uptake and accumulation of silicic acid within the diatom cell. This process is thought to be mediated by specific proteins that act as silicic acid transporters (SITs). The molecular basis for silicic acid binding and transport by the SITs remains unclear, and we are now attempting to understand the structure and function of these proteins using biochemical and biophysical methods. I will present our recent work in which we have used a yeast expression system to recombinantly express twelve SIT homologues. Six of these homologues were expressed at high levels, and five could be successfully purified. Of these, SITs from *P. tricornutum* and *N. alba* were found to be particularly amenable to in vitro studies. We have developed a novel fluorescence assay in order to study silicic acid transport by these proteins, and we have now used this assay to determine the transport kinetics and binding affinities of purified recombinant SITs in synthetic proteoliposomes. We are now exploring the impact of specific amino acid mutations on protein structure and function, and whether SIT proteoliposomes can be used as a simple reductionist model of diatom silicification.

## SESSION 2: SILICIFICATION IN OTHER SILICIFIERS

## GENERAL INTRODUCTION

## THE DIVERSITY OF SILICIFIERS IN NATURE

S. Baines<sup>1</sup> and M. Maldonado<sup>2</sup><sup>1</sup> Bigelow, USA<sup>2</sup> CSIC, CEAB, Spain

The ability to deposit polymerized silica is a trait shared by organisms from many disparate parts of the tree of life, suggesting either the trait is ancestral or has evolved multiple times. Silica sheaths are well known to form around filamentous colonial cyanobacteria in hot springs, a process that requires supersaturating silicic concentrations. Organic surface compounds are thought to regulate formation of these sheaths, and could be ancestors of those used by eukaryotes to guide silicification within the cell. Silicon has also been observed in association with manganese in decaying marine cyanobacteria. Some bacteria have also been seen to incorporate Si within endospore walls at subsaturating silicic acid concentrations. This impregnated wall can protect the spore against acidic conditions. Recently a number of reports suggest that free-living cyanobacteria concentrate Si against strong concentration gradients. Eukaryotes who engage in silicification generally restrict formation of silica to vacuoles, after which mineral silica can be exported extracellularly in some way. We will discuss in some detail how sponges deposit mineral silica. Then will discuss ways in which other eukaryotes organisms may differ in the mechanisms of silicification. These differences will be considered in light of extant phylogenies of the major group.

## THE CONTRIBUTION OF SPONGES TO THE MARINE SILICON CYCLE

Manuel Maldonado

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The understanding of how Si is cycling in the ocean has grown in association with the idea that diatoms are the only relevant biological players, being other Si users, such as sponges, choanoflagellates, radiolarians and silicoflagellates, unimportant. As diatoms account for about 40% of ocean primary production and help to recycle the anthropogenic CO<sub>2</sub> excess, the coupled use of carbon (C) and silicon (Si) by these organisms is receiving notable attention. By investigating Si budgets associated with siliceous sponges, we show this group to have also a substantial, though overlooked, impact on the Si cycle. On continental shelves and slopes, Si standing stocks per bottom area in sponge communities may equal and even surpass those in the diatom communities of the overlying water column. Sponge longevity and resistance of sponge spicules to dissolution in seawater cause sponges to lock DSi into biogenic silica (BSi) for periods that are orders of magnitude longer than those estimated for diatoms. Consequently, sponge populations are net sinks, favoring massive export of Si to the geological cycle. More importantly, the fact that most sponge DSi demands, BSi standing stocks, and Si exports are concentrated on continental margins has important implications for modeling local and regional budgets. In summary, there is a relevant, sponge-driven Si sub-cycle that is strongly decoupled from the C cycle and shows extremely slow turnover rates. This sponge role challenges several aspects of the traditional model describing the biogeochemical cycling of Si in the marine environment.

## SILICIFYING RADIOLARIANS: A BRIEF BIOLOGICAL, OCEANOGRAPHIC AND TAPHONOMIC OVERVIEW

Taniel Danelian

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Radiolarians are part of the marine protistan microzooplankton. Till recently, three different lineages were included in radiolarians; these are the Polycystines, with an aesthetically pleasing siliceous skeleton known since the Cambrian, Acantharea secreting a skeleton in celestite (strontium sulfate), not preserved in sediments and Phaeodarea with a highly porous siliceous test, known since the Cretaceous. Their single cell is subdivided in two parts by an organic membrane, the central capsula. The outer cytoplasm contains numerous bubble-like inclusions and an elaborate network of cytoplasmic extensions. The siliceous skeleton is secreted in intracellular vesicles; on living individuals it is covered by a thin layer of protoplasm. There are several hundreds of living polycystine species, known from all depths of the water column, although each is specialized in its trophic strategy. Symbiont-bearing species are restricted in the surface waters; others prefer to live near the nutricline, feeding on picoplankton, while others live in the abyssal parts, feeding on suspended organic particles. By grazing on the phytoplankton and thus controlling their population size, radiolaria play an important role in modern oceanic food webs. They are locally abundant, depending on the water mass conditions, especially with respect to nutrients. Thus, polycystines are known to be abundant in upwelling areas, in both coastal and equatorial upwellings. Few studies have attempted to provide quantitative data on radiolarian assemblages with depth for some parts of the world ocean. Estimates in radiolarian flux of biogenic silica are much influenced by dissolution processes in the water column and surface sediments.

## A ROLE FOR THE CYANOBACTERIUM, SYNECHOCOCCUS, IN THE MARINE SILICON CYCLE

Stephen B. Baines\*, Benjamin S. Twining, Jefferey Krause, Jackie Collier, Mark A. Brzezinski

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The biogeochemical cycling of silicon in the ocean is generally presumed to be dominated by diatoms, which constitute a large fraction of photosynthetic biomass in regions where pulses of nutrients into surface waters occur regularly. Over most of the remaining ocean that is permanently nutrient-poor, relatively little silicon is generally thought to be incorporated by organisms. Using synchrotron x-ray fluorescence microscopy (SXRF), we have shown that single cells of the cyanobacterium, *Synechococcus*, collected from three regions of the ocean often contain substantial amounts of silicon. Surprisingly, cellular concentrations sometimes approach those observed in co-occurring diatoms. As these organisms dominate the biomass in the large fraction of the ocean that is nutrient poor, this finding suggests that the silicon cycle in such regions may be more active than previously thought. Further work has revealed that there are systematic differences in silicon content among *Synechococcus* strains, negative effects of ambient phosphate concentrations on Si uptake, and order of magnitude variation in silicon contents within populations in the field and cultures in the lab. Furthermore, silicon does not appear to be required for growth. Field studies show that the inventory of silicon in *Synechococcus* sometimes rivals that in diatoms at some field sites, although most biogenic silica in the water column is detritus and of unknown origin. I will discuss possible mechanisms of Si accumulation by *Synechococcus*, and possible biogeochemical implications of silicon uptake by these organisms.

## SESSION 3: GENOMICS/PROTEOMICS AS TOOLS FOR SILICIFIERS

### GENERAL INTRODUCTION

Chris Bowler

ENS, Paris, France

## MOLECULAR TOOLS TO STUDY BENTHIC DIATOM COMMUNITIES

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Phototrophic, epilithic biofilms are a typical feature of aquatic ecosystems. To study interactions between diatoms and bacteria we isolated and identified different diatoms and associated satellite bacteria. Interestingly, purified diatom cultures showed significant differences with respect to biofilm formation when compared to the corresponding xenic cultures. The diatom *Achnanthes minutissimum* forms capsule - like structures in the presence of an isolated bacterial strain, but not in axenic state. SEM analyses show that capsule material that was mechanically stressed by being stretched between or around cells displayed fibrillar substructures. Fibrils were also found on the frustules of non- encapsulated cells, implicating that *A. minutissimum* capsules may develop from fibrillar precursors. We furthermore have screened the genome of the marine diatom *Phaeodactylum tricornutum* for gene models encoding proteins exhibiting leucine- rich repeat (LRR) structures. We were able to identify several transmembrane LRR- proteins, which are likely to function as receptor -like molecules. Moreover, *P. tricornutum* encodes a family of secreted LRR proteins likely to function as adhesion or binding proteins as part of the extracellular matrix. We furthermore have analyzed the extracellular proteome of *P. tricornutum* the presence or absence of *Roseovarius* bacteria.

## IDENTIFICATION AND COMPARISON OF SILICIFICATION GENES IN DIATOMS

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The patterns and assembly processes of diatom silica cell walls are an outstanding example of how genetic information is translated into structural design. Characterization and manipulation of silicification genes is a viable approach to understanding cell wall formation processes. However, because an analogous process has not been described elsewhere in biology, many genes involved in diatom cell wall synthesis do not have homologs in other organisms, and their functions must be described by fundamental characterizations. Simply identifying such genes is challenging, but techniques such as synchronized culture growth coupled with transcriptomic analysis provide a viable option. Our lab identified 485 genes potentially involved in cell wall formation of *Thalassiosira pseudonana*. Analysis of these genes highlights diverse aspects of the silicification and cell wall formation process. In particular, genes encoding proteins associated with the silica deposition vesicle membrane were identified, and knockdowns of these genes altered the surface structure of the valves, supporting a long-proposed model for silicification. These genes, and others shown to be involved in silicification, form the basis of a dataset to explore the conservation of such genes in diverse diatom species. The fundamental concept behind this approach is that diatom species that make similar silica structures may share genes with conserved characteristics, and species that make distinct structures may have unique genes related to formation of those structures. A cross-species comparison of demonstrated and potential cell wall synthesis genes, using available genomic and transcriptomic datasets suggests that this concept is valid.

## LINKING PHYSIOLOGICAL AND MOLECULAR ASPECTS OF DIATOM SILICIFICATION

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Diatoms form a critical link between carbon and silicon biogeochemistry due to their contribution to primary productivity and their obligate requirement for silicon (Si) for growth. Decades of oceanographic research has provided insight into the dynamics of silicon uptake and silica production in natural populations and into the role of silicic acid concentration as a limiting nutrient for diatom productivity. However, these studies, based on bulk biomass and rate measurements, do not adequately capture the underlying genetic and physiological mechanisms that regulate the observed rates. To bridge this gap we are linking measurements of silicon uptake using the radioisotope <sup>32</sup>Si with molecular and biochemical analysis of silicon transporters (SITs), diatom-specific proteins responsible for the uptake of silicic acid into the cell, and other silicification-related genes (SiRGs). For example, natural diatom populations in the California Coastal Upwelling system demonstrate elevated SIT expression at low external silicic acid concentrations that decreases upon Si addition, consistent with culture-based studies that suggest diatoms switch from SIT-mediated Si transport at low Si concentrations to diffusion-mediated uptake at high Si concentrations. We are currently working on methods to relate <sup>32</sup>Si-production rates to the expression of SITs and other SiRGs by natural diatom assemblages over diel cycles and with changing silicic acid and iron concentrations to help provide a mechanistic understanding of the genetic and biochemical basis of diatom silicification in the ocean.



## PHYTOPLANKTON - BACTERIAL COUPLING MEDIATES MICRONUTRIENT LIMITATION IN THE SOUTHERN OCEAN

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Primary productivity in the coastal Southern Ocean has a disproportionately large impact on ocean biogeochemistry and responses to global change. Here we show, using transcriptome sequencing of experimentally manipulated water column communities, that McMurdo Sound is interactively co-limited by iron and cobalamin availability. Diatoms, the dominant primary producers, displayed gene expression patterns consistent with simultaneous, independent limitation by both micronutrients. The major producer of cobalamin present in this location, an *Oceanospirillae* gamma-proteobacterial strain, highly expresses adhesion and attachment related functions, supporting the notion that cobalamin supply to phytoplankton may be mediated by direct interactions with bacteria. *Methylophaga*, another dominant bacterial group present, is central to processing of the key compounds dimethylsulfide and methanol and displayed molecular signatures of simultaneous cobalamin and iron deprivation. These experiments suggest multiple layers of interactive micronutrient co-limitation. Primary producers appear to be simultaneously limited by iron and cobalamin availability, and data suggest that availability of cobalamin is controlled by gamma-proteobacterial response to those phytoplankton. Therefore, iron limitation, widely documented in the ocean, may be better understood as micronutrient co-limitation that is mediated by phytoplankton-bacterial interactions.

## SESSION 4: A BIOGEOCHEMICAL/GENOMIC APPROACH FOR ECOSYSTEMS MODELLING

### GENERAL INTRODUCTION

## GENOMICS: A NEW DATA STREAM TO INFORM BIOGEOCHEMICAL MODELS?

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The rapid expansion of (meta) genomics opens novel opportunities for mapping the microbial and viral diversity of the world ocean. The combined collection of metagenomic and environmental data during observational programs allows for the identification of biodiversity patterns and their possible controls. Linking metagenomics and models will allow to identify controls of microbial community structure, dynamics and biogeochemical functioning in the present day and – by means of projections of physico-chemical conditions – in the future ocean. The combination of these novel data streams and biogeochemical models is still in its infancy. At present biogeochemical or lower trophic level models are mostly variants of the plankton functional type approach. A certain degree of plasticity is added to these models, with some allowing for the emergence of ecotypes. Most recently, a ‘gene functional type’ approach has been proposed, which allows for the first time to bridge the gap between biogeochemical function and gene activity. First analysis of extensive data sets provided by e.g. *Tara Ocean* reveal, however, a structure in microbial communities at a level superior to the organism and its genetic make-up. It appears that identified biogeochemical functions (f.i. export versus recycling) are linked to distinct states of the microbial community and associated gene activity. These novel data and insights not only challenge our understanding of the functioning of the marine

ecosystem, but also question the conceptual framework of dominant modelling approaches. Here we start from an accepted understanding of the role of diatoms in carbon export and explore how a modelling concept allowing for the emergence of microbial diversity and model ecotypes could be used to upscale observations both in time and space. We then confront the underlying principles to the results emerging from the combined genomic – environmental data set from Tara Ocean. Finally, we explore possible ways of integrating genomic information to novel modelling approaches.

## MODELING MARINE BIOGEOCHEMICAL CYCLES: CURRENT STATE AND FUTURE DIRECTIONS.

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Diatoms are known to be a major component of marine phytoplankton community as they probably constitute the most abundant and diversified group of eukaryotic phytoplankton. They generally dominate in eutrophic regimes and during phytoplankton blooms. Due to their large size and their siliceous frustules, they are thought to be key players in the transfer of energy to the upper trophic levels and in the export of carbon to the interior of the ocean. As a consequence of this pivotal role, most biogeochemical models, including those embedded in Earth System Models, now explicitly represent diatoms and the associated marine silicon cycle. In a first step, I will review how the silicon cycle, and in particular diatoms, are described in these biogeochemical models. Taking NEMO-PISCES as an example, the performance of the models will be briefly evaluated against observations at global scale. Most biogeochemical models currently rely on the plankton functional type or trait-based approaches. However, even the most sophisticated models fail to accurately represent the biodiversity of plankton. Furthermore, none of them properly consider adaptative responses, for instance to temperature changes. In a second step, I will discuss the future of microbial modeling. A key will be the representation of the organisms from the level of their macromolecular components. This will allow to combine traditional physiological studies with the new insights gained from genomics.

## REPRESENTING AUTOTROPHS SILICIFIERS IN PLANKTON FUNCTIONAL TYPES MODELS

### – APPLICATION TO THE NORTHWESTERN MEDITERRANEAN SEA

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While attention has been usually paid on the Nitrogen and Phosphorus biogeochemical cycles in the Mediterranean Sea the Silicon cycle remains poorly investigated and then not deeply understood in this oceanic area. Furthermore the abundance and role of autotrophs silicifiers have been generally considered as minor owing to the oligotrophic status of the Mediterranean Sea. Here we propose to show how ecosystem plankton model can help to better understand silicon dynamics and more accurately quantify the biogeochemical fluxes associated to the activity of autotrophs silicifiers. We first present an attempt for implementing them in Plankton Functional Types (PFT) model (Eco3M platform) and especially the great diversity of their ecology relative to nutrient uptake and ambient conditions of light. The alterations of some physiological parameters of diatoms-like PFT (as for example those of photosynthesis apparatus and affinity for nutrients) associated to special conditions of light and nutrient supplies enable to reproduce some of the main niches of autotrophs silicifiers usually observed in the sea. This way to represent diversity in autotroph silicifiers has been implemented in a 3D coupled physical-biogeochemical model (Eco3M-S-Symphonie). It is then possible to show and analyze the impacts of physical processes on their spatial and temporal distributions in the Northwestern Mediterranean Sea that is an area characterized by the presence of contrasted trophic water masses and strong (sub) mesoscale activity. The contributions of autotrophs silicifiers to primary productivity and export of biogenic silicon and particulate organic carbon below 200 m have been also assessed.

## OPTIMALITY-BASED VIEW ON PHYTOPLANKTON AND SI: HOW IS SILICIFICATION REGULATED AND WHAT IS ITS ROLE IN PHYTOPLANKTON SUCCESSION?

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An optimality-based phytoplankton model describes physiological processes based on the assumption that their regulation serves to maximise growth rate. It can be derived from the physiological roles of nutrient elements and associated trade-offs (costs and benefits) in terms of net growth. Recent optimality-based models of phytoplankton growth and C:N:P stoichiometry have demonstrated the usefulness of this approach. Derivation of N and P regulation has been facilitated by the detailed knowledge about their physiological roles and clearly defined relations between C:N:P stoichiometry and ambient conditions and growth. An optimality-based description for Si has so far been hampered by the lack of clearly-defined relations to cellular functions such as photosynthesis and nutrient uptake and assimilation. Silicification seems negatively related to growth rate, but diatom physiology appears to operate neutrally within a wide range in the degree of silicification, failing below some lower threshold. The surface area-normalised silicification rate appears to depend mostly on silicate concentration. Hence, a meaningful description of silicification requires a good grasp on cell size. The most important aspect of Si for phytoplankton modelling may be the advantages and disadvantages of the siliceous shells of diatoms relative to other phytoplankton groups. Siliceous frustules of diatoms are energetically cheap, appear to facilitate buoyancy regulation, and might deter grazers. However, the obligate Si requirement restricts the niche of diatoms, owing to slow Si remineralisation. An optimality-based description of phytoplankton composition might lead towards better understanding of primary production, particularly due to the importance of diatoms for export production.

## APPROACHES FOR INTEGRATING GENOMIC DATA AND BIOGEOCHEMICAL MODELS

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The advancement of high throughput sequencing techniques has greatly expanded our knowledge of the diversity and functional potential of marine microbial communities. However placing this information in the context of key biogeochemical nutrient cycles (e.g. C, N, P, Si) remains a challenge. Numerical modeling of biogeochemical processes has the potential to be an important tool for tackling this problem, but current biogeochemical models are seldom formulated in a way to take advantage of “-omic” data. Here, we present a functional gene-based framework for describing microbial communities in biogeochemical models. To demonstrate the approach in practice, we modeled nitrogen cycling in the Arabian Sea oxygen minimum zone and examined key questions about cryptic sulfur cycling and dinitrogen production pathways. However the approach is general and can be expanded to incorporate other biogeochemical processes such as the coupling of carbon and silica cycles. By directly linking geochemical dynamics to the genetic composition of microbial communities the method provides a framework for achieving mechanistic insights into patterns and biogeochemical consequences of marine microbes. Such an approach is critical for informing our understanding of the key role microbes play in modulating Earth’s biogeochemistry.

## SESSION 5: ISOTOPE CHEMISTRY PROVIDING TOOLS FOR PROCESSES AND FLUXES

### GENERAL INTRODUCTION

## THE ZINC ISOTOPIC COMPOSITION OF SILICEOUS MARINE SPONGES: INVESTIGATING NATURE'S SEDIMENT TRAPS

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The Zinc (Zn) isotopic composition of marine biogenic opal has the potential to yield information about the nutrient availability, utilisation and organic matter export from surface to deep waters. Here, we report the first measurements of Zn isotope composition of deep-sea benthic sponge spicules from the Southern Ocean. Our results highlight the different Zn uptake and fractionation behaviour between the two major siliceous sponge clades (hexactinellids and demosponges), which is most likely linked to sponge filter-feeding strategy and internal physiology. Hexactinellid spicule Zn isotopic compositions are not fractionated with respect to seawater, and so may be a potential proxy for past ocean Zn cycling. In contrast, demosponge spicules exhibit a wide range of Zn isotopic compositions that are related to the opal Zn concentration, most likely reflecting fractionation processes during feeding. As such, demosponge Zn isotope records may be able to shed light on past changes in photic zone organic matter formation in the ocean.

## SILICON ISOTOPES AND DIATOMS IN CULTURE STUDIES

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Silicon (Si) uptake and biomineralisation by marine organisms are of particular interest to oceanographers studying the Si cycle and its relationship to atmospheric CO<sub>2</sub>. Progress in these areas requires rate of silica production and dissolution to be quantified. However, the usefulness of geochemical tools (e.g. proxies) used to quantify these rates are hindered by our poor understanding of the mechanisms that control Si uptake and biomineralisation. Variations in the natural abundances of stable isotopes of Si (expressed as  $\delta^{30}\text{Si}$ , the parts-per-thousand variation in  $^{30}\text{Si}/^{28}\text{Si}$ ) is one key geochemical tool used to track relative differences in the extent of silicic acid removal in surface waters by diatoms. Quantitative use of this tool, requires precise understanding how Si isotopes behave during the phytoplankton growth season, for example, biological fractionation of Si isotopes by diatoms during silicic acid uptake and biogenic silica production greatly influences the  $\delta^{30}\text{Si}$  of surface waters because diatoms discriminate against the heavier Si isotope during uptake of silicic acid. However, several environmental and biological factors affect the magnitude of isotope fractionation, introducing uncertainty into the  $\delta^{30}\text{Si}$  based reconstructions of nutrient removal. While previous investigations have suggested variables such as temperature have minimal influence on Si isotope fractionation during biogenic silica formation, a thorough understanding of the mechanisms that control Si isotope fractionation is still lacking. It is important to confirm what mechanisms control Si isotope fraction by marine diatoms prior to completely asserting the usability of  $\delta^{30}\text{Si}$  as a proxy for monitoring changes in silicic acid utilization.

## WHAT CONTROLS THE SILICON ISOTOPE SIGNATURE OF BIOGENIC SILICA?

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The silicon isotope composition of biogenic silica ( $\delta^{30}\text{bSi}$ ), in the sea changes as a function of  $\text{Si}(\text{OH})_4$  utilization by siliceous organisms, and carries information about water mass mixing and silica dissolution. In upwelling areas characterized by high primary productivity, the bSi in surface waters mainly consists of diatoms. Preserved in the underlying sediment they can serve as archives for past nutrient conditions. In the present study we compare the  $\delta^{30}\text{bSi}$  of diatoms in the water column off Peru with that of diatoms preserved in the underlying surface sediments. We find that the diatoms from the water column have higher  $\delta^{30}\text{bSi}$  values compared to their sedimentary counterparts. This may be explained by the differences in the diatom assemblages found in the sediment and the seawater. Notably, the small (<20 $\mu\text{m}$ ) and fragile diatoms, e.g. *Cerataulina pelagica* and *Pseudo-nitzschia*, are abundant in seawater samples, but are not found in the sediments. Furthermore, the occurrence of *Chaetoceros* resting spores (RS) in sediments may have a significant influence on sedimentary  $\delta^{30}\text{bSi}$  signatures given that RS develop when Si in the surface waters is nearly completely consumed. Even though the timescales for surface sediment accumulation (years to decades) and surface blooms (days to weeks) are vastly different, the water column data, which was collected during main productivity season, should reflect the main fraction in the sediment. However, our findings reveal the complexity associated with the transfer of the isotopic signature established in surface waters to sediments via its potential alteration during export and burial.

## THE SI-MG ISOTOPIC COMPOSITIONS OF THE JUAN DE FUCA RIDGE LOW TEMPERATURE HYDROTHERMAL FLUIDS AND THE SI FLUXES FROM THE OFF-AXIS ROCK-WATER INTERACTIONS

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Low temperature fluids circulating in oceanic ridge-flanks play a major role in the heat and mass transfers between the crust and the ocean. To quantify the Mg and Si fluxes, we measured the dissolved Si and Mg isotopic compositions of pore water samples collected close to the sediment-basement interface of the eastern flank of the Juan de Fuca ridge (ODP Leg168). Starting from the proximity of the ridge axis,  $\delta^{26}\text{Mg}$  and  $\delta^{30}\text{Si}$  first change from -1.0‰ down to -2.6‰ and +1.7‰ up to +2.1‰, respectively, but becomes heavier (up to -1.5‰) and lighter (down to +1.4‰) eastwards.

These data fit with a sequence of two types of reaction of seawater with the igneous crust:

- uptake of Mg and Si by clays that preferentially incorporate heavy Mg and light Si;
- a Mg lost with no major change in Si content but lighter  $\delta^{30}\text{Si}$  indicative of slight clay replacements by carbonates. These isotopic shifts suggest that the coupling of  $\delta^{26}\text{Mg}$  with  $\delta^{30}\text{Si}$  could trace the Mg-Si gains and losses generated by the low temperature off-axis percolation of seawater. Considering the large differences of  $\delta^{30}\text{Si}$  and  $\text{DSi}$  ( $50\mu\text{mol} < \Delta\text{DSi} < 500\mu\text{mol}$ ) between these fluids and the bottom seawater, we expect that their discharge might impact the seawater compositions in a long term perspectives, especially for Si in regards of its short term oceanic residence time. Depending of the  $\delta T$  and  $\delta\text{DSi}$ , the net low-T hydrothermal Si input would be between 0.04 and 4 Tmol Si year<sup>-1</sup> potentially close to the continental inputs (5.8Tmol Si year<sup>-1</sup>).

## THE GE ISOTOPIC COMPOSITION OF SEAWATER: FIRST DEPTH PROFILES (SOUTHERN OCEAN) AND ESTIMATED FRACTIONATION BETWEEN SEAWATER AND BIOGENIC OPAL

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Germanium (Ge) has been long been considered a “geochemical twin” of silicon. Dissolved inorganic Ge displays a correlation with dissolved silica that is remarkably consistent across the world's oceans, underlying its potential as a complementary tracer for marine silicon cycling. Like silicon, it also possesses multiple stable isotopes whose natural variation may provide additional insights into marine silicon cycling. However, due to the low natural abundance of Ge, the Ge isotope composition of seawater, potential variations in isotopic composition with depth, and isotope fractionation between seawater and biogenic opal are all largely unconstrained. We have developed a new method for the pre-concentration of Ge from seawater and applied it to measure for the first time the Ge isotopic composition of seawater in three depth profiles from the Southern Ocean. Variations in  $^{74}\text{Ge}/^{70}\text{Ge}$  ratios are expressed as  $\delta^{74/70}\text{Ge}$  relative to NIST 3120a, and precision of the method is better than 0.2‰ (2SE). Our  $\delta^{74/70}\text{Ge}$  data demonstrate that surface ocean waters are isotopically heavy, becoming lighter until the oxygen minimum zone under which they appear stable. These data strongly suggest that biogenic opal preferentially takes up lighter Ge isotopes, resulting in seawater that is isotopically heavy ( $\delta^{74/70}\text{Ge} = 3.07 \pm 0.09\text{‰}$ , 2SE, average of all Southern Ocean samples) relative to the bulk silicate Earth ( $\delta^{74/70}\text{Ge} = 0.59 \pm 0.18\text{‰}$ , 2SE; Escoube, 2011). The Ge isotope fractionation factor  $\Delta^{74/70}\text{Ge}$  between seawater and siliceous phytoplankton is estimated at around 0.6‰. We also evaluated Ge isotope fractionation between sponges and coeval seawater, which we constrain to  $1.0 \pm 0.1\text{‰}$ . This study constitutes an important first step towards the application of marine Ge isotope systematics as a new tracer of global silicon cycling.



## EVIDENCE OF INCREASED SILICIC ACID LEAKAGE FROM THE SOUTHERN OCEAN DURING THE GLACIAL AND DE-GLACIAL PERIODS.

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The Southern Ocean plays a critical role in the exchange of carbon between the ocean and atmosphere. Hypotheses to explain late Quaternary glacial-interglacial atmospheric carbon dioxide (CO<sub>2</sub>) implicate changes in the nutrient dynamics and circulation of the Southern Ocean. Evidence from silicon isotopes ( $\delta^{30}\text{Si}$ ) from sponge and diatom opal preserved in sedimentary records from the Pacific sector of the Southern Ocean suggest that silicic acid concentrations in mode and intermediate water were higher during the Last Glacial Maximum (LGM) and late deglacial periods compared to the late Holocene (last 4 ka). Our results also suggest that the silicic acid concentration gradient between mode and intermediate waters was higher between 30 ka and the LGM. Finally these records provide evidence of an increased contribution of deeper nutrient-rich water to the surface Southern Ocean during these two periods challenging the current hypothesis of reduced ventilation of deep waters.

## ZINC ISOTOPES AND DIATOMS IN THE SOUTHERN OCEAN

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Trace metal micronutrients (e.g. iron, zinc, and cadmium) play a fundamental role in the biology of the oceans, due to their importance as co-factors in many enzymes. Dissolved Zn concentration depth profiles look like those of the major nutrients (phosphate, nitrate, and silica), with deep to surface concentration ratios as high as several hundreds. Culturing studies have shown that, though the majority of Zn in diatoms is hosted in organic material, the Zn/Si ratio in the opal of diatom frustules is positively correlated with the availability of free Zn(2<sup>+</sup>) in the growth media. To extend on these observations, Zn abundance and isotope compositions were measured in cleaned diatom opal obtained from core-top transects in the Southern Ocean. The measured  $\delta^{66}\text{Zn}$  (reported relative to the JMC Lyons standard) and Zn/Si ratios range from 0.7 to 1.5‰, and from 14 to 0.9  $\mu\text{mol/mol}$ , respectively. The systematic changes in the Zn/Si ratio are interpreted to be linked to the variability of bioavailable Zn in the ambient surface seawater where the diatom opal is formed. This variability is likely to be primarily controlled by the degree to which Zn is taken up into phytoplankton biomass. The systematic relationship observed in the core-top diatom opal suggests a potential tool for investigating the biogeochemical cycling of Zn in the past surface ocean for down-core diatom opal material. These observations will be discussed in relation to (1) zinc isotopes data in seawater and (2) Zn isotopes and abundances diatom opal extending back to the last glacial maximum.

## SESSION 6: THE SILICA CYCLE: KEY PLAYERS AND PROCESSES

### GENERAL INTRODUCTION

#### REFLECTIONS ON THE ROLE OF SILICIFIERS IN OCEANIC BIOGEOCHEMICAL CYCLES - CONCEPTUAL APPROACHES AND RESEARCH PROSPECTIVES

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In the marine environment, silicification is a process that occurs in several unicellular and multicellular organisms. Long regarded as the monopoly of diatoms, radiolarians and sponges, silicification appears today much more widespread in the plankton, whatever their size class, including silicoflagellates, parmales and cyanobacteria. From a geological point of view these different players have been modifying the Global Ocean biogeochemistry of silicon over some 500 my. In coastal ecosystems, diatoms appear as major players of the primary production, a position they tend also to occupy at the global scale. For this reason they are a major link between C and Si biogeochemical cycles hence influencing the global CO<sub>2</sub> biological pump. Regarding the silica cycle, coastal ecosystems are much complex than open ocean ecosystems from different points of view. In this context, the study of the dynamics of diatom blooms remains a challenge which can be tackled in the near future by a combined approach between classical biogeochemistry, quantitative ecology, and omics. Some conceptual models recently elaborated (Assmy *et al.*, 2013; Boyd, 2013; Quéguiner, 2013) need now to be assessed. Among the hot questions still under debate are the understanding of the initiation of blooms, and of the control of seasonal species succession, including the causality of the growth termination of individual diatom species. The deciphering of the mechanistic aspects of diatom developments is

essential to predict the evolution of both C and Si cycles in a warmer ocean. Among others, these aspects include identification of all the silica cycle players, identification of their pathogens, study of the emergence of sexual stages, biogeochemistry at the single cell level (imagery and cell quotas), as well as examination of specific processes, such as mixotrophy, hitherto overlooked for silicifiers.

## SESSION 6A: COASTAL ECOSYSTEMS

**BENTHIC VERSUS PELAGIC DIATOMS**

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Diatoms are found in almost all aquatic environments as long as light is available for their photosynthesis. In coastal ecosystems, pelagic diatoms are extensively studied, but their benthic counterparts, more discreet and more difficult to access, are often ignored. Yet living at much lower light intensities, they are a key compartment of coastal ecosystems functioning, being able to produce as much biomass as pelagic diatoms that live in the well-lit surface waters. Benthic diatoms differ considerably from pelagic forms on several aspects: First, pelagic and benthic diatoms do not follow the same seasonal dynamics: In the bay of Brest, we showed that benthic diatoms are the first to bloom in the season, providing food and energy to the ecosystem right from the beginning of spring (with 60% of the total biomass until April). The system then moves from a system dominated by benthic biomass in early spring to a system where the pelagic biomass dominates. Regarding biodiversity, most benthic diatoms species are different from those found in the water column, they are mostly pennate forms and are far more diverse. On the physiological side, benthic diatoms are incredibly well adapted to low light intensities. However, the mechanisms which result in this adaptation remain unclear. We search for the role of the nanostructure of the frustule in the capture of light, but no difference between pelagic and benthic diatoms could be highlighted so far.

Of particular interest is that benthic diatoms able to migrate into the sediment, are adapted to the high  $\text{Si(OH)}_4$  concentrations found in their surrounding and evidence a multiphasic Si uptake with very high half saturation constant (KS). Their capacity to reverse benthic  $\text{Si(OH)}_4$  fluxes at the interface indicate the important role they also play in the coastal silica cycle and particularly in controlling the pelagic diatom production.

It remains to examine the coupling between carbon and silica at the water/ sediment interface and to determine which factors control the benthic production. Further studies are also needed to understand which evolution has allowed pennate diatoms to colonize so successfully the benthic habitat.

## THE SUCCESS OF DIATOMS: THE NITROGEN PERSPECTIVE

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Diatoms live in dynamic environments. Cells in dynamically changing environments are more likely to experience superimposed stresses, including fluctuating light availability, fluctuating availability of different N substrates, and changing temperatures, all of which can create conditions affecting the balance or coupling of carbon and nitrogen assimilation. Balancing energy-generating and energy-using processes with those of carbon and nitrogen acquisition is a delicate task for the cell, requiring signaling molecules to facilitate cross-talk and various release valves to rebalance the flow of electrons or materials when imbalances develop. For carbon, some of the processes that aid in the regulation of cellular energy and redox balance include non-photochemical quenching, xanthophyll cycling, and the Mehler reaction. For nitrogen, an important pathway regulating overall cellular energy balance is the reduction of  $\text{NO}_3$  and  $\text{NO}_2$  via nitrate reductase (NR) in a nonassimilatory mode that complements such reduction in  $\text{NO}_3$  assimilation. This pathway is favored under cool temperatures. When  $\text{NO}_3$  is comparatively unavailable, photorespiration can increase significantly. Diatom dependence on nonassimilatory  $\text{NO}_3$  reduction as an energy balance mechanism is one of the reasons they are  $\text{NO}_3$  specialists. It also makes them especially susceptible to inhibition and repression by  $\text{NH}_4$ . Such repression may be a function of failure of NR-related dissipatory pathways and enhanced photorespiration at the expense of assimilation pathways when imbalances develop. These physiological pathways and their ecological implications, in a world where anthropogenic loads of  $\text{NH}_4$  are increasing, will be reviewed.

## THE GLOBAL SI CYCLE: VARIABLE INPUTS INTO THE OCEANS AND POSSIBLE CHANGES IN THE SI CYCLE THROUGH GEOLOGIC TIME

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Recently, a new paradigm has emerged regarding the importance of the terrestrial Si cycle. We now understand that it is strongly mediated by biological action, which creates and recycles a pool of reactive Si in soils and sediments that ultimately buffers the export of DSi to the oceans and probably has done so for at least the last 250 million years. Although a common assumption is that the global Si cycle is at steady-state, our recent results show that imbalances in the continental Si pool on millennial scales may have caused systematic mis-estimation of weathering rates, particularly when using river chemistry at large spatial scales. Quantifying this imbalance is essential to understand ocean-terrestrial biogeochemical interactions. Over long time scales significant changes in Si fluxes to the oceans have probably occurred. For example, periods of glacial-interglacial transitions provide new, highly weatherable material on the continents, extensive changes in sea-level that modify the extent of estuarine area and accumulation, with subsequent impacts on the oceanic Si budget. On yet longer timescales large-scale changes in chemical weathering due to *e.g.* orogenesis, the emplacement of large igneous provinces, or changes in volcanic activity can similarly impact the land-to-ocean delivery of Si. Using modeling approaches, our results demonstrate that changes in the continental Si cycle alone can be sufficient to drive changes in whole ocean  $\delta^{30}\text{Si}$  at comparable rates and magnitudes to that recorded in marine BSi, without the need to invoke within-ocean processes. This implies we may need to revisit aspects of our understanding of the coupled Si and C cycles in the ocean.

## EARLY DIAGENESIS OF SILICA AND REVERSE WEATHERING IN MARINE SEDIMENTS: BIOLOGICAL CONTROLS AND INTERACTIONS WITH ORGANIC CARBON CYCLING AND PRESERVATION.

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Reverse Weathering (RW) reactions in marine sediments have been shown to have significant impact on global geochemical cycles of major (K, Mg) and trace (Li, F) elements. Silica is an essential element in the formation of new diagenetic aluminosilicate phases, the main products of RW reactions in marine sediments. Silica is derived from the dissolution of biogenic silica particles and/or detrital aluminosilicates and re-precipitates in newly formed aluminosilicates as dispersed particles, as coatings on detrital particles or other buried bioparticles (e.g. diatom frustules, pollen particles) and as altered/reconstituted diatom frustules. The net result is silica storage in marine sediments not only in the form of biosiliceous particles but also as siliceous phases formed during early diagenesis. Despite the close association of Si with organic carbon in biologically produced particles in the marine environment (e.g. diatoms), cycling of Si and organic carbon during early diagenesis in marine sediments is largely considered uncoupled. Contrary to this perception, studies in Mississippi deltaic sediments indicate close correlations between reactive Si and organic carbon in diagenetically altered sediments while they show a lack of correlation in riverine and recently deposited sediments, pointing to an early diagenetic Si control on the burial and preservation of organic carbon. These strong indications for linkages between reactive silica and organic matter cycling raise the possibility for potential biological controls on early diagenesis of Si. Initial results from incubation experiments with Congo deep sea fan sediments suggest a rapid and significant influence of microbial activity on early diagenetic recycling of Si in marine sediments.

## SI CYCLE AND THE COASTAL ZONE: WHAT ABOUT MAN IN THIS STORY?

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Diatoms are playing a key role in the functioning of coastal ecosystems; hence, the Si biogeochemical cycle is being explored to better understand the dynamics of these ecosystems located at the land-ocean interface. Pelagic diatoms are key players, but they are not the only ones as the role of benthic diatoms, sponges, cyanobacteria is being revealed. This cycle, as others, is being profoundly perturbed by human activities such as eutrophication, damming, or the proliferation of exotic and invasive species linked to climate change or the global transportation system. These modifications, in turn, affect the functioning of these ecosystems and they may have deep implications in terms of integrated coastal management. So, what about Man as a key player in this context? As a source of perturbation, but also as a key actor for the sustainable development of coastal areas. But if the N or P cycles can be manipulated by man for example to reduce eutrophication, what about the Si cycle? Is the Si cycle taken into account by decision makers, or have they even heard about it? Using the social-ecological framework developed by Collins and his collaborators (2011), we will explore the different questions that may arise when trying to link our beloved Si cycle to human perceptions, behavior, practices, and decision making towards a more sustainable coastal zone. We will use the example of the evolving Bay of Brest ecosystem, subject to several aspects of global change (eutrophication, invasive species, climate change...) which imply decision-making and public action defined at the intersection of different spheres: scientists, professionals (agriculture, fisheries...), NGO's, managers and decision makers. The idea is to explore how nutrient cycling, and in particular the silicon cycle, is or is not taken into account in this process of public action.

## SESSION 6B: OPEN OCEAN ECOSYSTEMS

## PHYTOPLANKTON DETECTION FROM SPACE BASED ON THE SYNERGY BETWEEN OCEAN COLOUR AND IN SITU OBSERVATIONS: PRINCIPLES AND PERSPECTIVES

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Over the last past years, a large number of new approaches in the domain of ocean colour have been developed, leading to a variety of innovative descriptors, from estimation of particles size in the surface to the detection of phytoplankton groups.

One of these methods, the PHYSAT one, currently allows the detection of five main phytoplankton groups from ocean color measurements when they are dominant (Alvain *et al.*, 2008, 2005), at the global scale. Even if the PHYSAT outputs, in its last published version, are largely used in various applications and projects, one limitation still prevents developments: the detection of dominance cases only. Indeed, dominance is not always the most adapted products in terms of users needs. However, according to the theoretical explanation provided by Alvain *et al.* (2012), it is expected that a development of PHYSAT is still possible toward the detection phytoplankton communities (regional diatoms species composition for example) or cells developments stages. Based on these theoretical results, recent works consisted in evaluating various approaches in order to improve PHYSAT products. After a quick presentation of PHYSAT principle and main results at this stage, current strategies and needs for future improvement of the products will be presented.

## DIATOM STRATEGIES FOR PERSISTENCE, GROWTH AND REPRODUCTION IN THE EASTERN EQUATORIAL PACIFIC: AVOIDANCE OF NUTRIENT LIMITATION VS. BLOOM-AND-BUST

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Surface waters in the eastern equatorial Pacific upwelling zone are characterized by low, potentially limiting concentrations of both silicic acid (typically 1–4  $\mu\text{M}$ ) and dissolved iron (50–250  $\text{pM}$ ). Diatoms in those waters exhibit two distinctly different strategies for growth and reproduction: Small pennate forms, by far the most abundant diatoms in the system, grow at moderate rates (1–2  $\text{d}^{-1}$ ), without strong limitation by either Si or Fe. Their populations are held in check by both microzooplankton and mesozooplankton grazing. In contrast, large centric diatoms are rare, have very high maximum growth rates ( $\mu_{\text{max}}$ :  $>3 \text{ d}^{-1}$  and perhaps  $>4 \text{ d}^{-1}$ ) but their growth rates are strongly limited in situ by low [Fe]. They are grazed primarily by mesozooplankton, and persist at low numbers despite strong Fe limitation. Their ubiquitous presence, high  $\mu_{\text{max}}$  and strong in situ limitation by Fe are all reflected in the fact that their populations explode over 2–5 days in mesocosms when Fe is added at 2 nM. These two very different strategies –moderate  $\mu_{\text{max}}$  which is approached in situ despite low ambient [Fe] and [Si] (small pennate species) and very high  $\mu_{\text{max}}$  which is not approached in situ because of severe Fe limitation (large centric species) – clearly represent different evolutionary responses to the same habitat, under genetic control. From an ecological and biogeochemical perspective, it would be interesting to know whether information on differences in the genomes of these two diatom types can shed light on how this works at the cellular level.



## PARMALES AND BOLIDOPHYCEAE, A PICOPLANKTON GROUP CLOSELY RELATED TO DIATOMS

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The order Parmales (Heterokonta) is a phytoplankton group containing small eukaryotic marine, 2 to 5µm in diameter, with a silicified cell wall composed of several various shaped plates. Recently, we isolated the first ever culture of Parmales, from the coastal Oyashio region of Japan. Phylogenetics and photosynthetic pigments analyses of this culture indicated that Parmales lies within the class Bolidophyceae which are autotrophic naked flagellates and a sister group of diatoms. Recently several new strains of Parmales and Bolidophyceae have been isolated allowing to refine the relationships between these two groups. The analysis of metabarcoding data from the *Tara Oceans* project permitted to map the distribution of the different taxa throughout the oceanic realm. We will also present recent physiological and genomic data obtained for this important group.

## INTERSPECIFIC VARIABILITY OF SILICIFYING ACTIVITY AMONG DOMINANT DIATOMS OF THE PHYTOPLANKTON COMMUNITY IN THE SOUTHERN OCEAN (KEOPS 2 PROJECT)

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Since the pioneer works of Hutchins and Bruland (1998) and Takeda (1998), it has been commonly admitted that iron-limitation induces the growth of heavily silicified diatoms with higher silicon (Si) to carbon (C) elemental ratios than in iron-replete conditions. In the iron-depleted Southern Ocean waters, this would have large implications on the global biogeochemical cycles by limiting Si supply to low latitude waters over the other nutrients. The Kerguelen Plateau (KP) region represents a unique natural laboratory to study the impact of iron on the functioning of a plankton community and the associated biogeochemical cycles because it is characterized by recurrent extensive blooms contrasting with surrounding HNLC waters. The northeastern part of the KP was investigated during the KEOPS2 cruise in early spring (October-November 2011). As part of this cruise, we investigated the link between the diatom community composition and the C and Si biogeochemical cycles by assessing diatom specific Si and C biomasses as well as silicification degree at the cellular and specific levels. We evidenced unexpected high diatom Si:C biomass ratios in the iron-fertilized area and high variability in the silicification degree between species and regions differently impacted by iron-availability. Our observations suggested that diatom specific Si and C biomasses were highly dependent on specific composition of diatom assemblages and on their specific responses to environmental conditions rather than on the iron availability alone. This emphasizes the importance of studying the specific composition of diatom assemblages to better understand their impact on the biogeochemical cycles.

## QUANTITATIVE DETERMINATION OF GROUP-SPECIFIC SI UPTAKE IN FIELD DIATOM ASSEMBLAGES

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A new method allows taxon-specific silica production rates to be determined within natural diatom assemblages. This technique has the potential to improve our understanding of the diatom groups dominating total silica production across ocean regions and to reveal how competition for silicic acid influences diatom group succession, thereby informing both food-web and biogeochemical models. We tested the power of the approach in the waters off the central California coast (USA) by combining traditional Si-tracer methods (radioisotope  $^{32}\text{Si}$ ) with an improved quantitative method for measuring silica incorporation by single diatom cells using the fluorochrome PDMPO. While 10 genera were present in the assemblage, *Chaetoceros spp.* was the most abundant, accounting for 93% of cells; this group was responsible for 87% of total silica production at ambient silicic acid concentration ( $0.47 \mu\text{M}$ ). *Pseudo-nitzschia spp.* and *Odontella spp.* were present in much lower relative abundances, 1.3% and 0.2%, respectively, but their contributions to total silica production were approximately eight-fold higher than their abundances (9.7% for *Pseudo-nitzschia spp.*, 1.7% for *Odontella spp.*). Using  $^{32}\text{Si}$ , the half-saturation concentration for the assemblage was  $2.6 \mu\text{M}$ . Within this assemblage, the degree of silicification in response to increasing silicic acid availability was determined for four diatom genera (*Chaetoceros*, *Pseudo-nitzschia*, *Skeletonema*, *Odontella*), and half-saturation concentrations ranged between  $<0.4 \mu\text{M}$  and  $3.0 \mu\text{M}$ . The results provide unprecedented detail regarding interspecific variation in silica production in the field. Widespread application of this methodology will provide new information regarding the functional variability of diatoms within natural assemblages.

## SPATIO-TEMPORAL VARIABILITY OF THE RELATIONSHIP BETWEEN PHYTOPLANKTON COMMUNITIES AND PARTICULATE MATTER STOICHIOMETRY IN THE GOLFE DU LION (NORTHWESTERN MEDITERRANEAN)

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Data on nutrients and the composition of particulate matter (particulate organic carbon, nitrogen, and phosphorus, biogenic silica) were acquired from May 2010 to December 2014, on a monthly basis, at four contrasting (inshore and offshore) stations of the Golfe du Lion. The stations were located at the eastern entrance, mostly oligotrophic, and the western exit influenced by Rhône River and small coastal tributaries inputs. In parallel, the composition of the phytoplankton community was followed at each station by microscopical examination, and enumeration, as well as cell size measurements enabling calculation of biovolumes then converted to carbon biomass. The latter procedure allowed to partition the carbon biomass between the different components of the phytoplankton assemblages. We present the spatial and temporal evolution of phytoplankton assemblages with special attention on the ratio between siliceous (diatoms and silicoflagellates) and non-siliceous components (dinoflagellates, nanoflagellates), and the relative importance of living vs. detrital particulate organic carbon in the coastal and shelf ecosystems of the Golfe du Lion. These results are compared to the stoichiometry of nutrients (nitrate, ammonium, phosphate, silicic acid) and particulate matter (C/N/P/Si ratios), and the silicification degree of diatoms and silicoflagellates, which is evaluated on the basis of biogenic silica measurements by chemical analysis and microscopical carbon biomass evaluation of each species or group of species at different seasons.

## SILICATE PUMPING OF EXCESS CARBON IN COASTAL UPWELLING ECOSYSTEMS

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Diatoms, designated the « workhorses of the sea » by Victor Smetacek, require silicate to construct their frustules. They dominate new production in upwelling ecosystems and control the rate at which nitrate and associated carbon are assimilated into the euphotic zone. The Redfield ratio defines the average ratio of major nutrients C:N:P in phytoplankton as 106:16:1 and the phytoplankton uptake of nitrate (new production in the ocean) is often used as a proxy for carbon production and export of carbon to the deep ocean. Elevated nitrate and  $\text{CO}_2$  in upwelled water is rapidly drawn down by diatoms, usually within 3-5 days. From the Redfield ratio the ensuing drawdown of  $\text{CO}_2$  would be expected to be a function of the initial concentration of nitrate. If the ratio of Si:N in upwelled water is 1:1 or greater, and the canonical ratio of Si:N in coastal diatoms is taken as 1:1, silicate in excess of nitrate would be expected to have no effect on carbon drawdown and export. However, in large volume enclosure experiments using silicate-rich San Francisco Bay water, silicate drawdown continued well after nitrate was exhausted by phytoplankton growth. Similar grow-outs made with upwelled water from Point Reyes, California showed the same pattern of silicate drawdown continuing past nitrate exhaustion. Dissolved inorganic carbon (DIC) concentrations were also measured with the unexpected result that DIC drawdown followed silicate drawdown stoichiometrically and continued after nitrate exhaustion. The implication of these preliminary results is that coastal upwelling basins rich in silicate, e.g. North Pacific, may account for substantially more drawdown/absorption of  $\text{CO}_2$  than would be calculated from upwelled nitrate concentrations. The excess carbon exported to the underlying waters by silicate pumping also may be a factor in the development of hypoxia along the U.S. West Coast upwelling areas. In these circumstances where there is a direct role of silicate in  $\text{CO}_2$  drawdown, new production may be better defined in terms of silicate rather than nitrogen and be a better indicator of biological carbon processes.

# SILICAMICS

## POSTER ABSTRACTS

## SILICON UPTAKE BY SPONGES IN THE BAY OF BREST

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Sponges make a group of ubiquitous organisms that often elaborate siliceous skeletons by taking up and polymerizing silicic acid (DSi). Little is known about how the DSi uptake works, with kinetic information available for only two genera, which makes complicated to derive reliable estimates of field DSi demands. Herein, we present the kinetic uptake of two abundant sponge species at the Bay of Brest, *Hymeniacidon perlevis* and *Tethya aurantium*. Uptake rate increases with increasing ambient DSi availability following a Michaelis-Menten kinetics, which is in agreement with the enzymatic processing of DSi known for siliceous sponges. Despite these two sponges living at the same habitat, they show notably different half saturation and saturation parameters in their respective kinetic equations. Interestingly, both sponges attain saturation at experimental DSi concentrations that are more than one order of magnitude than field DSi concentrations over the year cycle. Within the natural DSi concentration range in the Bay of Brest (0.05 – 18  $\mu\text{M}$ ), the silicon (Si) consumptions estimated for *H. perlevis* and *T. aurantium* range from -0.003 to 0.025 and from 0.001 to 0.065  $\mu\text{mol Si per h and ml of sponge tissue}$ , respectively. Hence, both studied sponge species live in Si limited conditions, with low DSi uptake rates at natural DSi concentration, being clearly adapted to higher DSi concentrations.

## SILICON ISOTOPES AND BIOGEOCHEMICAL PROCESSES IN INDIAN ESTUARIES

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Si, a specific nutrient needed by diatoms and its availability relative to other nutrients determines the phytoplankton composition while land use and climate change alters the supply of nutrients in estuarine and coastal ecosystems functioning. In order to understand the biogeochemical cycling of Si and its supply to coastal ocean, here we present the first study on silicon isotopes ( $\delta^{30}\text{Si}$ ) in more than 18 tropical Indian estuaries during dry period. Average  $\delta^{30}\text{Si}$  is  $1.9 \pm 0.4 \text{ ‰}$  ( $n=58$ ) which is almost 1 ‰ heavier than the world river supply to the ocean estimated so far. Indeed, contrary to a typical weathering control on riverine  $\delta^{30}\text{Si}$  as generally observed, there is no positive correlation between Dissolved Silicon (DSi) content and  $\delta^{30}\text{Si}$ . More specifically in western estuaries,  $\delta^{30}\text{Si}$  are lighter ( $1.2 \pm 0.2 \text{ ‰}$ ) with negative and positive correlation with DSi and Biogenic Si (BSi), respectively. This cannot be ascribed to diatoms since these variations are not related to diatoms pigments. In contrast, the eastern estuaries have heavier  $\delta^{30}\text{Si}$  ( $2.1 \pm 0.3 \text{ ‰}$ ) with no clear relation with either DSi or BSi contents across the salinity gradient. Interestingly, there is good correlation between BSi and diatom pigment ( $r=0.65$ ) that might be indicative of the significant role of diatoms.

The  $\delta^{30}\text{Si}$  of monsoonal headwater samples are not significantly different from those of the dry period ( $1.64 \pm 0.55 \text{ ‰}$  and  $2.01 \pm 0.61$ , respectively) despite there is almost no discharge during dry period. This indicates that the Si source to estuary is insensitive to the season even though it appears to be controlled by weathering as seen on the inverse relationship  $\delta^{30}\text{Si}$  vs.  $1/\text{DSi}$ . Then, within estuary during monsoon,  $\delta^{30}\text{Si}$  is little changed and seems to be mainly controlled by mixing.

## SILICON ISOTOPE FRACTIONATION BY DIATOM SILICIC ACID TRANSPORTERS

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Diatoms fractionate silicon isotopes by discriminating against the heavier isotope ( $^{30}\text{Si}$ ) during uptake. Understanding the physiological factors that may influence the Si quota and the isotopic signal is vital for interpreting biogenic silica as a paleoproxy. Si fractionation during uptake is potentially species dependent and it has been suggested that Si isotopic discrimination in diatoms is set by the Si transport step rather than by the polymerization step. Diatom cell membranes contain specific silicon transporters (SITs), such as ToSIT protein for *T. oceanica*, which are essential for Si uptake. SIT proteins, native to individual diatoms species, were microinjected into *Xenopus laevis* oocytes and the media from the subsequent transport studies was analysed for silicic acid concentration and isotope fractionation. The results showed isotope fractionation occurring during Si transport through the SITs. We varied the SIT proteins and initial silicic acid concentrations to simulate uptake by different species under different environmental conditions. This study will enhance our understanding of Si uptake by diatoms by tracking the silicon isotopes fractionation and consequently the interpretation of silicon isotope from diatom fossils.

## ACTIN ASSOCIATED GENES: IMPLICATED IN HIGHER ORDER SILICA STRUCTURE FORMATION IN THE DIATOM *THALASSIOSIRA PSEUDONANA*

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The intricate and diverse patterns of diatom silica cell walls have inspired extensive work aimed at understanding the processes behind their formation. Through a variety of biochemical and bioinformatic techniques, proteins and other organic molecules associated with silicification have been identified. In addition cytoskeletal elements including actin and microtubules have been implicated in determining silica structure, particularly at intermediate and large scales. There is still relatively little known about the proteins which may facilitate the cytoskeleton's influence on silicification. The *Thalassiosira pseudonana* genome contains a host of genes with actin associated domains; transcriptomic data obtained over a time course from a synchronized culture of *T. pseudonana* demonstrates that a subset of these genes are also up regulated during valve formation. The proteins encoded by these genes present optimal candidates to evaluate actin's influence on silica structure. Gene silencing was used in order to characterize the morphological impacts of a subset of these genes. Our data suggests that characterized genes are involved in higher order silica structure formation. This work demonstrates the potential of combining transcriptomic data and genetic manipulation to elucidate the impact of specific genes on silica cell wall formation in diatoms.

## FUNCTIONAL STUDIES OF SILICON TRANSPORT PROTEINS IN THE XENOPUS OOCYTE EXPRESSION SYSTEM

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The incredible nanostructured silica cases (frustules) of diatom species are extremely diverse and abundant, yet the mechanisms of uptake and polymerisation of environmental silicic acid under ambient conditions are still largely unknown. Being able to recreate and control this process artificially is of great interest in many fields including engineering, nanotechnology and pharmacology. Silicon transport proteins (SITs) are trans-membrane proteins with a role in the uptake of silicon. Many SITs have been identified in different diatom species and their sequences known. Our research focuses on understanding the mechanism of action of these proteins using the *Xenopus laevis* oocyte expression system.

*In vitro* transcription and microinjection into *Xenopus* oocytes allows expression of SITs in a single cell system, enabling analysis of protein function without interferences such as presence of other proteins involved in silicon metabolism. Thus far, we have successfully expressed eight SITs from six diatom species. Analysis of their silicon uptake ability, by measurement of changes in media silicon concentration over short time periods, indicates both inter- and intra-species variation.

Future work aims to fully characterise the transport mechanism of individual SITs, and identify whether the differences observed between the proteins could play a role in the diversity of frustule patterning.

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