TARA
OCEANS

Scientific Programs

Summary

It is now unequivocal that global warming is occurring at a rate 100 times greater than documented in geological studies. The major part (>50%) of global primary biomass production occurs in the upper 200 m surface water layer of the oceans (the photic zone) and this also drives most of the global elemental cycling involved in climate regulation. Key actors in these elemental cycling processes are thought to be plankton protists, which produce oxygen and recycle carbon dioxide. However, protists are embedded in a network of plankton organisms that range from viruses to fish larvae and the complex, dynamic food webs that these organisms form remain largely un-investigated.

TARA OCEANS will analyze plankton ecosystems in relation to physicochemical conditions throughout the world's oceans, assessing their adaptation to and feedback on a rapidly changing earth system. Large scale genomics and metagenomics projects coupled to newly developed high-resolution and high-throughput imaging methods will allow quantitative studies of plankton ecosystems and identification of the plankton composition within these ecosystems. Integrative bio-informatics analyses of the imaging, genomes, physicochemical and climate data generated in the project, will be used to assess plankton biodiversity and activity in the diverse ocean ecosystems sampled. All of the data generated through the project will go to form an open-source multidimensional bio-oceanographic database that will allow generating predictive models of the spatio-temporal evolution of plankton ecosystems.

The results of this project will have outstanding implications for our understanding of early life evolution, global biogeochemical cycles and spatio-temporal evolution of the earth climate.

Coordination
Scientific Consortium

Steering Committee

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Olivier Jaillon, Genoscope, FR	Genomics
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Gilles Reverdin, UPMC, FR	Physico-Chemistry
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University of Milan Bicocca, Italy
MNHN, Paris France
James Cook University, Townsville, Australia
Museum of Tropical Queensland, Townsville, Australia
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University of Warwick, Coventry, UK
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Cross-disciplinary oceanography, sampling and navigation infrastructure Tara
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University of Maine, Orono, USA
ACRI-ST, Sofia-Antipolis, France
LEGOS/CNRS, Toulouse, France
GIP Mercator Océan/CNRS, Ramonville St Agne, France
METEO France, Toulouse, France
Satlantic Inc., Halifax, Canada
Hydroptic Ltd., Lisle en Dodon, France
LOCEAN / UPMC, Jussieu, Paris, France.

Specific cross-disciplinary collaborations
CNRS/UPMC, Paris, France
IFREMER, Brest, France
University of Hawaii, USA
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Cross-disciplinary cellular/organism imaging and cytometry infrastructures

EMBL, Heidelberg, Germany
CNRS / UPMC, Villefranche-sur-Mer, Roscoff, France
University of Washington, Seattle, USA
School of Biology and Environmental Science, UCD, Dublin , Ireland
Monterey Bay Aquarium Research Institute, USA.

Sequencing and Bioinformatics infrastructures

Genoscope, Evry, France
EMBL, Heidelberg, Germany
EBI, Cambridge, UK - Stazione Zoologica, Naples, Italy
IOBIS,/Cmarz / Census of Marine Life, Washington, USA.

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Prof. Mohamed Said	National Institute of Oceanography and Fisheries	Egypt
Dr George Zodiatis	Oceanography Centre	Cyprus

I. TARA OCEANS FOR PLANKTON

I.1. Oceanic Protists, Symbiosis, and extracellular DNA

Contact:
Colomban de Vargas, Station Biologique, CNRS / UPMC, Roscoff, France.

The Roscoff team will focus on protists - unicellular micro organisms possessing a nucleus (eukaryotes). Everyone has heard of viruses, bacteria, plants, or animals but most people still ignore protists, which make up a fifth compartment of Life on Earth and are present in all Oceans. Marine protists appeared more than a billion years ago. They have since evolved into a bewildering diversity of organisms whose complex structures, genomes, and metabolisms significantly contribute to the energetic and biogeochemical resources of our planet.

Among planktonic organisms, plant-like protists are particularly important as they produce massive amounts of oxygen. They also absorb CO2 thus generating fluxes of carbon from the atmosphere to the oceanic depths and sediments. Despite their essential role in global ecology and climate, we probably know less than 1% of the existing protist species. Our project **POSEIDON** – **P**r**O**ti**S**tan **E**colog**I**cal bio**D**iversity in Tara-**O**cea**N***s,* funded by ANR, proposes novel technological and methodological approaches for exploring the genetic and morphological diversity of oceanic protists. Using massive sequencing of DNA markers, we will characterize the most important groups of protists in the oceans, and analyse their morphology and function in marine ecosystems. For the first time we will explore the entire size-range (from 1 to 5000 microns) of protists and their biodiversity. This should lead to an understanding of ecological relationships, and in particular symbioses, between protists of different sizes.

We will also coordinate research on planktonic biodiversity. We have established tight collaborations with the GENOSCOPE for sequencing and bioinformatics, the EMBL Imaging team in Heidelberg for high-throughput microscopy, the Polytechnic University of Marche in Ancona for exploration of extracellular DNA. We will coordinate research carried out by a network of international experts (Tara-Oceans protist consortium) to analyze and interpret the data generated by the expedition.

Four types of experimental data will be obtained from the same water sample: *genetic* (DNA sequences), *morphological* (light and electron microscopy), *archival* (DNA, RNA, and cellular samples), and *contextual* (ecological parameters). The nuclear, mitochondrial, and chloroplast biodiversity markers will be massively sequenced using the *454*-DNA technology in collaboration with GENOSCOPE (France) and the Consortium for the Barcoding of Life initiative (Canada). Cellular structures and morphologies will be analyzed by high-throughput fluorescence, light, and electron microscopy protocols, as well as by flow cytometry.

I.2. A "Genetic" Measure of Biodiversity

Contact: Olivier Jaillon, GENOSCOPE, CEA, Evry, France

GENOSCOPE - the French National DNA Sequencing Centre - is participating in the inventory of marine micro organisms using two complementary genetic approaches:
- *A measure of species diversity*: We will sequence massively the genomes of micro organisms sampled from three different ocean depths (subsurface, Deep-Chlorophyll Maximum-DMC and aphotic zones). Species will be collected on filters and distributed according to their volume using different micrometer pore sizes. - *A measure of gene diversity*. In parallel with the first approach we will carry out extractions and sequencing of the copies (RNA) of genes expressed in the micro organisms present in the samples. This approach will lead to a better knowledge of the "active" portion of the micro organisms as well as knowledge of the set of genes utilized by micro organisms to adapt to their environment.

Among the various micro organisms collected during the Tara Oceans expedition, GENOSCOPE has chosen to study a specific category of marine plankton - the protists - as well as a group of giant viruses - giruses (Mimivirus) - which have been discovered recently. Unicellular eukaryotic protists represent a resource which is still poorly known and unexploited and could be valuable for industrial applications. Discovery of new biocatalysts could lead to novel methods of chemical transformations less polluting for the environment than synthetic chemical processes used today.

I.3. GENEXO: Quantitative relevance, sources and GEnetic imprint of EXtracellular DNA in the world's Oceans

Contact: Antonio Dell'Anno and Cinzia Corinaldesi, Polytechnic University of Marche, Ancona, Italy.

Extra cellular DNA (i.e. DNA not associated to biomass or to viral particles) is a ubiquitous component of the organic matter pool in the world's ocean. Extracellular DNA plays a key role in biogeochemical and trophodynamic processes and is potentially involved in recombination processes through horizontal gene transfer. At the same time the genetic information contained within the extra cellular DNA pool, which integrates biological sources and processes, represents the "background noise" of biodiversity and dynamics of pelagic food webs.

Despite the advancement of knowledge on the dynamics of the extra cellular DNA pool in marine environments, many questions remain. What is the quantitative relevance, origin and ecological significance of extracellular DNA in pelagic marine ecosystems over large spatial scales and in different ecological settings?. Do extra cellular DNA concentrations change in relation with latitudinal and longitudinal oceanic gradients? What are the main ecological factors influencing concentration and distribution of extra cellular DNA on a global scale? Which are the major biological sources of extra cellular DNA pools? Is the genetic imprint of extracellular DNA coupled with biodiversity and community structure of the pelagic food webs? To address these major issues, wide sampling efforts and joint scientific collaborations are required. The Tara-Ocean expedition offers a unique opportunity to investigate the quantitative relevance, main sources and genetic imprint of the extracellular DNA pool in the world's oceans.

Using chemical, biochemical and molecular biological approaches we will analyse the extra cellular DNA pool. This is expected to improve the understanding of the functioning and biodiversity of the oceanic ecosystems and the role of natural and/or anthropogenically mediated drivers. This research will be carried out by the Department of Marine Science, Polytechnic University of Marche, Ancona.

I.4. **TANIT - TA**ra Ocea**N**s Prokaryiot**I**c Func**T**ioning and Diversity**,**

Contact: Silvia Gonzalez-Acinas, Barcelona, Spain.

TARA-Oceans is a global circumnavigation ocean sampling project that brings together an international team of scientists to **explore ocean life and its sensitivity to climate change.**

TANIT (**TA**ra Ocea**N**s Prokaryiot**I**c Func**T**ioning and Diversity) represents a consortium of scientists within TARA-Oceans that will inspect the functioning ecology and biodiversity of bacteria in the oceans. TANIT will perform a 3 years worldwide study of the bacterial biogeography of the surface layers and DCM samples. Bacterial assemblages will be correlated with physico-chemical environmental data and the relationship with other fractions of the marine plankton such fish larvae, zooplankton, algae and viruses will be explored.

TANIT is motivated by the very rapid climate changes and increasing ocean temperatures and acidification, which may affect the health status of major oceanic ecosystems. Bacteria are a relevant component of the ocean's food networks; they are responsible for 30% of the primary biomass production and 95% of the respiration of the ocean accounting with 10^{29} cells in the global ocean. Bacterial biodiversity and ecological functioning are still poorly known but this knowledge is essential to understand carbon flow and shifts in marine bacterial communities patterns in respond to environmental changes. A global quantitative and qualitative survey of prokaryotic life will be performed through high-throughput genomics/transcriptomics and other molecular ecological approaches as a baseline for future studies and to supply data to dynamic models of ecosystem responses to the changing climate.

The Labs involved in the TANIT consortium is integrated by 15 groups of multidisciplinary scientists from Europe (France, Germany, Netherlands, Italy and Spain) and the US (MIT, UC) specialized in oceanography, microbial ecology, environmental microbiology, genomics and bioinformatics providing an unique opportunity to shed light into the current status of bacterial "life" of the oceans.

TANIT will pave the way to explore global ocean bacterial genomes and will develop innovative methods generating an extensive and coherent inventory of genetic and functional diversity in this important component of the ocean's food networks. New prokaryotic species will be discovered and biogeography of bacterial species will be for the first time comprehensively explored in a global oceanographic context. TANIT will contribute to monitor the quantitative evolution of planktonic ecosystems and base future conservation decisions on firm scientific knowledge

I.5. Functional genomics of diatoms

Contact : Chris Bowler, CNRS/Ecole Normale Supérieure, Paris, France

Diatoms are one of the most important components of marine phytoplankton and are the main players in the biological carbon pump (sequestration of CO2 from the atmosphere to the ocean depths). During the Tara Oceans expedition, the SeaFlow cytometer will continuously monitor diatom concentrations in surface waters. In addition, water samples will be collected that are enriched in diatoms, both from surface waters and from the Deep Chlorophyll Maximum (DCM) zone. We will use these for microscopy observations, both onboard using live samples, and onland using fixed samples, in order to characterize diatom populations at the species level.

To facilitate species identification we will use the fluorescent dye FITC-silane, which specifically labels the silicified cell walls of diatoms. Cells from water samples will be collected on filters and DNA and RNA will be extracted. DNA and rRNA will be sequenced in order to quantify species abundance at the molecular level, and mRNA will be sequenced to reveal gene expression profiles in different oceanic contexts.

Key outcomes expected are a global evaluation of diatom communities and diatom gene expression profiles in a range of different oceanic contexts. The results will provide a basis for understanding how diatoms will be affected by climate change-induced phenomena in the future.

Laboratories involved are the ENS in Paris, Stazione Zoologica in Naples, University of Washington Seattle, EMBL in Heidelberg, Genoscope in Evry, EBI in Cambridge , and the Station Biologique in Roscoff.

I.6. Biogeography of viral diversities and metabolisms

Contact: Matt Sullivan, University of Arizona, Phoenix, USA

Microbes drive the biogeochemical engines that run the planet, and we've recently learned that - at least for cyanobacteria - their viruses interact at the level of the core metabolism - photosynthesis - and in a big way: 60% of microbial photosynthesis core machinery in the surface oceans is viral. However, our understanding of the types of viruses that occur in the wild, and their probable large impact on modulating the biogeochemistry in the global oceans is minimal.

The goal of the Project OViD (Ocean Virus Diversity) is to map the biogeography of viral diversities and metabolisms along the Tara Oceans circum-global transect. Our laboratory will initially target one of the most abundant bacterial double stranded DNA (dsDNA) virus groups observed in the oceans, the T4-like viruses.

There are two other virus labs involved in the project, that will complement these efforts: Markus Weinbauer's laboratory (Observatoire de Villefranche sur Mer) will examine the molecular diversity of dsDNA viruses using a fingerprinting technique, while Mya Breitbart's laboratory (University of South Florida) will document the ssDNA and RNA viral diversity. We will use these datasets, along with the extensive biotic (basic viral measures + measurements of other organismal class) and abiotic (chemistry and physics) data, to select a sub-set of the 375 sampling sites for viral metagenomic sequencing. Mya Breitbart and I were recently funded to improve viral (meta)genomics by the USA National Science Foundation as Project PHANTOME: Phage Annotation Tools and Methods.

The goal of the metagenomic work is to examine the diversity of the whole viral community, as well as to further our understanding of the metabolic capacity and evolution of the whole viral community. In addition we are prepared to examine the viral signal in Tara Oceans microbial metagenomic and metatranscriptomic datasets as directed by partner researchers. Hiroyuki Ogata's group (University of Marseille)will be examining giant viruses (Giruses) and has coordinated the sampling and analyses with the project TANIT (the prokaryotes group) because of these large viruses being collected with the "prokaryote" fraction.

I.7. Hunting “Giruses” in the oceans

Contact : Hiroyuki Ogata, Nigel Grimsley, Université de la Méditerranée, Marseille, France

Giant viruses (called “Giruses”) are large double stranded DNA viruses. Giruses infect a wide range of eukaryotes, including fishes, shrimps as well as climatically important photosynthetic plankton, and are thus considered to affect the food web worldwide in the sea. Today, the largest virus known is the amoeba-infecting Mimivirus with its 0.7 micron particle and 1.2 mega-base pairs genome.

Girus genomes are expected to be an important source of new genes used in developing new genetic tools, for example in medicine. However, our knowledge of marine giruses is still very limited. To discover new giruses, to assess their genetic diversity and to understand their ecological and evolutionary roles, we will collect organisms and water samples containing giruses at stations visited by the Tara schooner.

Hunting giruses requires their isolation in the laboratory. To this end, we will grow a number of previously untested protists (“unicellular eukaryotes”) in laboratory cultures in our laboratories as hosts to permit discovery of novel giruses. GENOSCOPE will decipher the genetic information in these giruses using high-throughput sequencing technologies.

This first global inventory of marine giruses will provide an essential baseline for assessing the future impact of climatic changes on marine girus-host cell microbial communities. As “Girus Hunters”, we hope to encounter record-breaking giants of the viral world through our expedition.

The laboratories involved are the Institut de Microbiologie de la Méditerranée (Marseille), the Observatoire Océanologique (Banyuls), the Station Biologique (Roscoff), GENOSCOPE (Evry), and the Instituto de Ciències del Mar (Barcelona).

I.8. Marine Biology Imaging

Contact: Dr Emmanuel G. Reynaud, University College Dublin, Ireland

Tara Oceans Marine biology Imaging platform (TAOMI)

The aim of the plateform is to allow every team member and laboratories involved in Tara- Oceans to image its particular plankton, coral or sea weed species of interest on board during collections for live observations. Samples will also be observed more thoroughly on land. The compact onboard imaging platform combines novel instruments (including prototypes) as well as up to date microscopy equipment and underwater imaging instruments. The on land platform relies on a network of laboratories in Heidelberg, Villefranche, Roscoff, Dublin. The TAOMI platform will allow high throughput analysis of plankton, confocal microscopy as well as X-ray micro-tomography. These facilities will be freely available to all Tara-Oceans network members.

Poseidon High Throughput Imaging Analysis (PHITIA)

This program developed in parallel with Tara-Oceans is part of the ANR POSEIDON project. In order to understand the complexity of the plankton community it is essential to identify and quantify all organisms present in the water column.

Each sample collected and fixed on board Tara during the cruise will be used to identify and quantify the different species it contains. This requires the development of a unique high throughput imaging platform allowing the imaging of various organisms of different sizes, shapes, optical properties and mineral constituents (silica skeleton, calcium carbonate skeleton…). Moreover, it will be necessary to develop specific image analysis software and applications to allow access and analysis via a web interface for researchers who request it. All the images will be stored in the Tara-Oceans BioBanks.

The Tara Oceans cruise is a unique chance to thoroughly test newly developed equipment, prototypes and also to adapt “terrestrial” microscopes to boat conditions. We will keep on developing the equipments on board as well as introduce and develop new technologies.

“An image is worth a thousand words”

The TAOMI platform includes aquariums and a macro-photography and HD video studio as well as the best microscopy and underwater imaging equipment. We will certainly obtain astonishing images of marine organisms suitable for education and communication. We will ensure that images taken by participating laboratories are properly disseminated.

I.9. Operational Oceanography

Contact: Gabriel Gorsky, LOV, CNRS / UPMC, Villefranche sur Mer, France.

Several projects will be carried out by the members of LOV (Laboratoire d'Oceanographie de Villefranche) laboratory and a network of partner laboratories.

Rapid determination of faunistical composition of zooplankton

-We will undertake a comparative study of the zooplankton faunistical composition in the different oceanic systems sampled during the expedition.
-We will estimate the carbon cycling of the different ecosystem structures.
-We will set up a digital image bank for zooplankton related in different oceanic regions.
-We will provide a world wide distribution of photosynthetic pigments in superficial waters (300 m) using HPLC methods.

The analysis of Plankton samples will be done using the ZooScan. It is an integrated system built in Villefranche/mer for acquisition and processing of digital images from preserved samples. The liquid samples are digitized and processed to detect, quantify, measure, and identify planktonic organisms. Zooscan also delivers size and mass estimations.

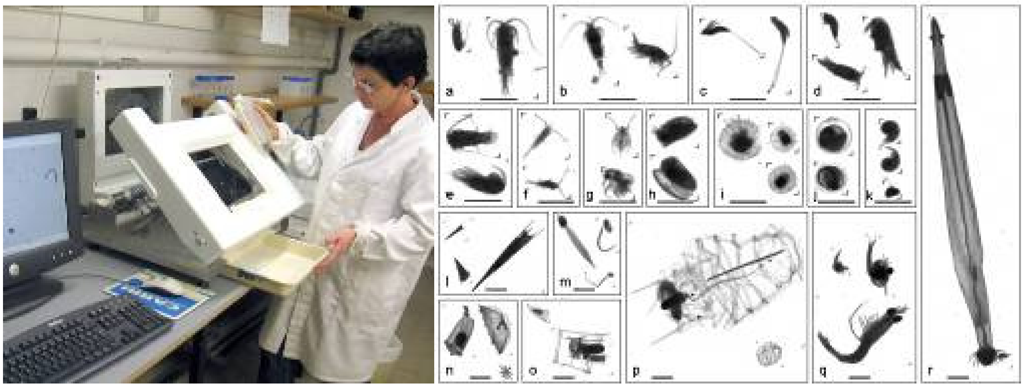


Figure 1: Right: sample recovery from the ZooScan following the digitisation, left: Mediterranean zooplankton ZooScan image.

Vertical distribution of zooplankton and of particulate matter in relation to the hydroclimatic conditions. Estimation of the vertical export of carbon.

We will estimate carbon export from the first kilometre along the Tara Oceans route using the Underwater Vision Profiler (UVP). Images and data will be treated onland.The laboratories involved are the LOV,France, Stazione Zoologica in Naples, Italy, IMS METU in Turkey, and MCM in Cape Town, S. Africa..

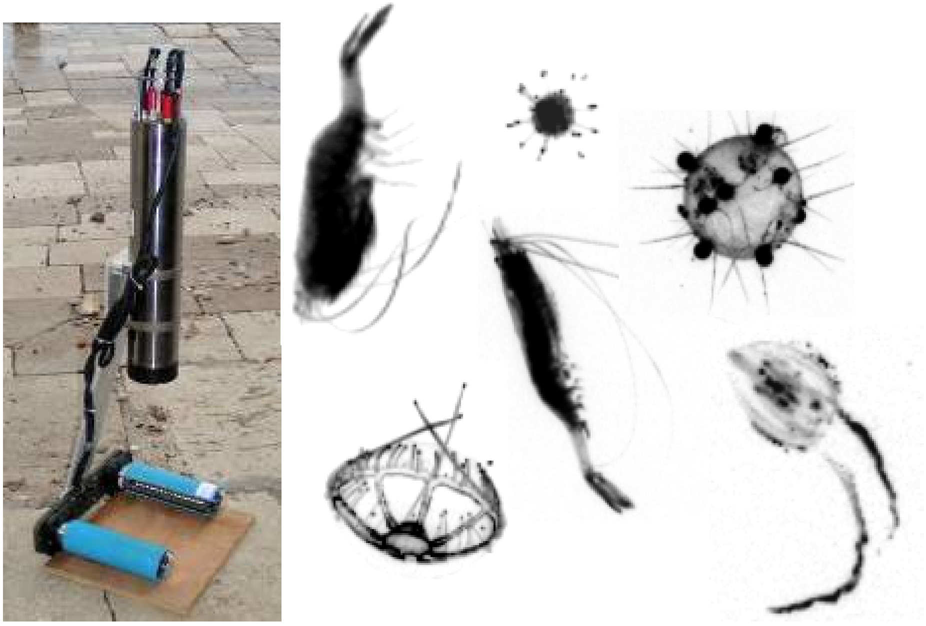


Figure 2. The UVP stand-alone version. The working range of the UVP is 0- 3000 meters and the volume of each image acquired at the frequency of 5 im/s is 1,2 litres. The abundance and size of zooplankton is sorted *in situ* and visualized onboard.

I.10. An integrated Biobank

Contacts: Maria Krestyaninova, EBI, United Kingdom, Stephane Pesant, PANGAEA®, Germany, Jeroen Raes, EMBL, Germany

Integrating this wealth of information ranging from oceanographic and satellite data to images and sequences is a challenge. A tool needs to be developed that will allow to establish correlations between these various sets of data.

Classically, oceanographers store their environmental results in specific « databases » and their Imaging data about plankton in other repositories. New initiatives tend to bring all this together but efforts are not yet fully integrated. Genome data from marine organisms are also stored in other places and the integration of all this information, so crucial to the understanding of marine ecosystems, is still largely missing.

TARA OCEANS will develop a new integrated data processing and storage system to integrate all the data acquired as described in the previous sections. The goal is to provide a tool to extract functional correlations between genes, the diversity of organisms present in a given region and the physical environment. This will also provide information that modelers working on the dynamics of oceanic ecosystems will be able to use to predict how oceanic life will react to rapid climate changes. This work will be carried out at EMBL Heidelberg and the EBI, in collaboration with PANGEA in Bremen.

II. TARA OCEANS FOR CORALS

II.1. Bacterial community composition and diversity

Contact: Dr Christine Ferrier-Pagès and Dr Didier Zoccola, Centre Scientifique de Monaco.

Keywords: Bacteria, biodiversity, coral-bacterial associations, coral diseases

Since the last century, coral reefs have been rapidly declining. Human activities, as well as climate change coupled to extreme El-Nino oscillations, are considered as some of major causes of the occurrence of new diseases in the reefs. The amplitude of this stress can be severe in some regions, constituting a major ecological problem. Increase in seawater temperature favours pathogens development, and decreases host resistance. Procaryotes (viruses, bacteria) are among the most diversified organisms in the world and many are living in close association with animals or plants. In corals, procaryotes are found in the mucus layer, in the tissue as well as in the skeleton, but coral-bacterial associations are still largely unknown. Bacterial diversity in corals is linked to the health of such corals. However, few studies have yet been performed to understand the composition and functioning of the bacterial populations in healthy corals, and even less in stressed ones. The project aims to compare the bacterial diversity associated to three coral species in different reef throughout the Indo-Pacific Ocean. This comparison will help us to highlight bacterial species closely associated to each coral species independently of their location suggesting a real symbiosis. Symbiotic interactions will be studied by DNA characterisation of bacteria associated to corals both using the classical DGGE electrophoresis and sequencing of discrete bands or using pyrosequencing. For this purpose, both seawater and coral samples will be taken in the studied reefs, on several colonies and locations. The expected results are a better understanding of coral reef species and their relation to symbionts and associated bacteria, and potential applications for environmental management and health of coral reef ecosystems.

II.2. Understanding and forecasting climate change impacts on coral reefs: A baseline study of the algal symbiont communities (*Symbiodinium* spp.) in reef-building corals from understudied and remote reef regions in the Indo-Pacific

Contact: Dr Andrew Baker, University of Miami, Florida, USA

Keywords: Symbiodinium, zooxanthellae, symbiosis, biodiversity, Scleractinia

The aim of this project is to characterize the algal symbiont communities in reef corals from understudied reef regions in the Indo-Pacific, and use this information to better understand the functional biodiversity of reef corals globally, and their response to climate change. We propose to relate *Symbiodinium* communities to: (1) environmental parameters, particularly temperature and recent bleaching history at the site; (2) the identity of the coral host (link to the coral systematics team); (3) other microbial partners of corals (link to the coral microbial team). We will use this information to better understand whether the distribution of putatively heat tolerant symbionts (in *Symbiodinium* clade D) are found more commonly in warmer areas and specific coral taxa, and whether there are impacts on associated microbial communities as a result of these partnerships. This research will help build our global understanding of functional biodiversity in reef corals, and help us understand whether and how coral can adapt to warm environments by hosting heat-tolerant algal symbionts. These results will contribute to addressing a pressing question in coral biology, ecology and conservation: do reef corals possess a novel symbiotic mechanism for mitigating temperature change? Can they switch between physiologically and genetically distinct algae when exposed to different thermal conditions? This information will provide important data for an assessment of how coral reefs will respond to future global climate change.

II.3. Algal diversity in the Indian and Pacific oceans

Contact: Dr Line Le Gall Museum National d'Histoire Naturelle (MNHN – UMR 7138), France.

Keywords: Algae, Rhodophyta, Pheophyceae, Chlorophyta, biodiversity, biogeography

In the following project proposal we aim to study green, red and brown algal diversity to infer the patterns and processes of speciation events for macroalgae. The Indian and Pacific Oceans provides a unique opportunity to study processes and patterns of speciation in a marine environment:
i) it is a "hot-spot" of marine biodiversity
ii) it represents both continental and insular coasts which provides continuous and isolated habitats to evaluate the influence of factors such as reproductive isolation and long distance dispersal on genetic divergence
iii) the tormented geological history of these oceans provides a time line to infer the rate at which speciation events occur over geological time.

Toward this goal, an extensive sampling will be performed across the Indian and Pacific Oceans. Reliable species assignment will be conducted on every sample using a DNA-barcode in addition to morphological and anatomical observations. On the one hand, phylogenetic analyses will be performed to ascertain taxonomic affinities of every characterized species of green, red and brown algae in a worldwide context. On the other hand phylogeographic inferences will be conducted to study the biogeographic affinities of the Indian and Pacific Ocean flora. This study will considerably improve our knowledge on algal diversity and biogeography; and will constitute a reliable starting point to monitor floristic changes influenced by human footprints including the global warming. It will also have important implications for natural resource conservation and management.

II.4. Benthic Foraminifera Biodiversity: latitudinal and longitudinal patterns

Contacts: Prof. Daniela Basso University of Milano - Bicocca (UNIMIB), Italy

Keywords: Benthic Foraminifera, sediment, biodiversity

Foraminifera are the most abundant marine Protozoa in the benthic, epypelagic and upper mesopelagic realm. Because of the complexity and diversity of habitats, especially in the shallow benthic realm, foraminifera show high biodiversity and abundance as an effect of their different ecological requirements. Short life cycles and the possibility of genetic reorganization by sexual reproduction enable rapid reaction to environmental changes. This makes foraminifera ideal bio-indicators for short- to long-term changes of marine environments, from global to extremely local scales. The main regulation factor for abundance and diversity is trophication: oligotrophic and mesotrophic conditions induce higher diversities, whereas eutrophication leads to the development of a few opportunistic species.

We will work on the biodiversity patterns of benthic foraminiferal bio and thanatocoenoses present in soft sediment surface samples collected at all sites. Research on benthic foraminifera distribution and biodiversity will allow us to obtain useful information on the main ecological factors affecting their assemblage composition, such as, food supply, sediment texture and energy state of the substrate as function of geographical and depth gradients. Furthermore, the possible presence of planktonic and epiphytic individuals could be used respectively as proxy for export production and ecological association with other benthic groups. This project will allow: 1) to obtain quantitative data on benthic foraminifera assemblages from reefal and peri-reefal environments, along all the select geographical gradients; to use benthic foraminifera assemblage biodiversity data as proxy for changes in geographic and/or environmental patterns.
We plan to obtain bottom sediment samples from all the selected TARA team stations (1-7).

II.5. Coral biogeography and Biodiversity: latitudinal and longitudinal patterns

Contact: Dr Carden Wallace Museum of tropical Queensland (MTQ)

Keywords: Scleractinia, Acropora, Isopora, Astreopora, biodiversity, biogeography

We will work on the biogeography and biodiversity patterns of the coral family Acroporidae, with emphasis on the genera *Acropora*, *Isopora* and *Astreopora*. We have a project well underway, using 26.400 *Acropora* and *Isopora* specimen-based records, in which we are analysing a range of patterns connected with latitudinal and longitudinal patterns, for example depth range changes with latitude, gradients north and south of the equator, longitudinal spread of species. This program is generating a series of hypotheses about the origins and future prospects of Indo-Pacific coral distribution, taking in past and present climate conditions. In this project, we are monitoring species range extensions north and south of the equator in the Pacific Ocean. The Tara Ocean research program would dovetail with the research of the other members of the coral biodiversity team, and complement their selection of coral genera and families.

Our project plan would have the following components: 1) Biogeographic affinities and origins of eastern and western Indo-Pacific Acroporidae; 2) Population genetics of established and establishing species (W. Indian Ocean); Refinement of hypothesis of Central Indian Ocean biodiversity origins, previously developed for Central Indonesia using the WWAC; Development of an analogue model for Acroporidae evolution and biogeography, using the small genera *Astreopora* and *Isopora* (combining data from this project and fossil data); Development of a unified revision of the worldwide biogeography of Acroporidae, as a baseline for monitoring of species boundary change in the three nominated genera; Development of a unified hypothesis of the origins and future prospects of world coral distribution, based on Acroporidae, the dominant Indo-Pacific coral family and previously in Europe and the Caribbean. We will use fossil research data from our studies in Caribbean and European palaeontology to complement the biological data from this research, to develop the overall hypothesis of the future response of coral distribution patterns to global climate change

II.6. Insular and fragmented populations and the phylogeography of Scleractinian corals

Contacts: Prof. Michel Pichon[1] and Fabrizio Stefani[2] Museum of tropical Queensland (MTQ)[1], University of Milan-Bicocca[2]

Keywords: Scleractinia, Psammocora, phylogeography, insular/fragmented populations

Tara Oceans which will be calling to isolated and seldom studies coral reef locations in the Indo-Pacific Ocean. Sampling in such locations will allow to obtain first hand data on the phylogeography and biogeography of selected and geographically widespread coral species. This should provide key information on the faunal relationships between the various, and at times very seemingly very isolated populations of such species. Recent taxonomic studies on morphospecies belonging to the scleractinian coral families Psammocoridae and Siderastreidae (as they are presently understood) have provided new and reliable data on species boundaries between close-related species, and have at the same time deeply altered our previous knowledge on their distribution and biogeography. There remains, however, many gaps in our knowledge of the detailed distribution of such species, and as is often the case for geographically largely distributed species, on their inter-regional and inter-population genetic variability. In this respect, isolated areas to be visited during the Tara Ocean Expedition, and which are not easily accessible during the course of normal research projects, provide a unique opportunity to study the morphologic and genetic make up, and to better understand the relationships between population of the same or of sibling species in distinct and more or less isolated area of their overall distribution range. It is expected that the project will generate results on the phylogeny and biogeography of species and species groups in the families Psammocoridae and Siderastreidae.

II.7. Species boundaries in Scleractinia: implications for systematics and biogeography

Contacts: Dr Francesca Benzoni and Dr Fabrizio Stefani University of Milano - Bicocca (UNIMiB)

Keywords: Scleractinia, species boundaries, integrated systematics, biodiversity, biogeography

The study of reef dwelling Scleractinia (Cnidaria) biodiversity has received considerable input in the last decades thanks to extensive sampling and field observations. However, on the one hand many remote locations still await for exploration. On the other hand coral taxonomy and species distribution remain unstudied for most taxa. As a consequence, most coral species and their distribution need to be validated.

Besides its intrinsic importance as such, the species boundaries problem in scleractinian corals has important downfalls on several aspects of coral reefs biology, ecology and conservation. The re-evaluation of traditional morphologic characters of the skeleton, the study of the polyp morphology and the discovery of previously disregarded macro, micro and nano structures have proved phylogenetically informative when combined with molecular results. The aims of this project are: 1) the investigation of species boundaries of a selected number of problematic taxa with a large distribution range through a multidisciplinary approach; 2) the biodiversity assessment of reef-dwelling Scleractinia in remote and/or poorly studied locations; 3) the build up of a reference collection for each sampled coral of skeleton, preserved tissues, *in vivo* images and information on the environmental conditions at the time of sampling. The search for species boundaries and phylogenetic relationships in and between selected taxa will be pursued through a multidisciplinary approach aiming to match information coming from different tools including SEM imaging, CT-scanning, morphometric analyses, type material and original descriptions re-examination, molecular phylogenies. As a result of the species delineation, more accurate species distribution will be obtained for the studied taxa. Assessing a species presence in different environments and its range of plasticity will provide, in turn, important information for the study of the ecology of coral reefs and of their biodiversity in the different locations.

II.8. Diversity and resilience of reef corals in the Indian Ocean

Contacts: Dr David Obura Coral Reef Degradation in the Indian Ocean (CORDIO - East Africa)

Keywords: Scleractinia, resilience, biodiversity, biogeography

The biogeography of shallow marine organisms in the western Indian Ocean (WIO) is poorly known, though there are indications of a peak of biodiversity (species and genus distributions) in the region at the northern end of the Mozambique Channel encompassed by northern Madagascar, northern Mozambique and southern Tanzania. This region is fed by the South Equatorial Current passing to the northern tip of Madagascar, with some indications of the formation of a gyre around the Comoro Islands and the formation of large eddies that subsequently move southwards in the Mozambique Channel. The aim of the proposed project is to determine the biogeography and relative abundance of coral species to: fit into a regional dataset on western Indian Ocean corals (Obura 2008); 2) identify patterns of diversity between the Iles Eparses and adjacent coastlines on the larger islands and mainland; 3) assess the resilience and current ecological state of coral reefs on the Iles Eparses; 4) identify any latitudinal gradients in the above and their relevance to biogeography and climate change vulnerability. The research proposed for Tara Oceans is to sample using ecological and biogeographic techniques used in a) ongoing characterization of the biogeography of reef corals in the WIO (Obura 2008), and b) a study to determine if there is a centre of diversity and reef resilience for the WIO. The data collected will have relevance to regional conservation planning as has been carried out in the East Africa and Western Indian Ocean marine ecoregions (supported by World Wildlife Fund, WWF, and the Indian Ocean Commission, COI), and to long term vulnerability of the region and any peripheral regions dependent on it as a larval source, to climate change.

II.9. From transects to population dynamics: a mixed assessment-prediction approach

Contacts: Prof. Bernhard Riegl and Dr. Samuel Purkis Nova Southeastern University

Keywords: Scleractinia, population trajectories, phototransects

Expeditions are efficient in reaching many different areas and are therefore able to rapidly assess many different reefs and species. We aim to use this spatial efficiency during Tara Oceans and combine it with sampling that provides detailed information required to investigate life-history parameters of rare coral species. The aim of the proposed project is to use snap-shot information obtained on an expedition to model population trajectories of rare and uncommon coral species. Reef-scale satellite remote sensing will be used to provide spatial context into which the field-survey will be nested. Satellite imagery is a well proven time- and cost-efficient technology for mapping the distribution and health of reef habitats across entire depositional systems. Expedition data-sets traditionally provide rapid snap-shot assessment of community aspect (living cover), community composition (taxonomic rankings), and health. We will augment these data by extracting from photo-transects: transition dynamics of corals between different stages, predictability of future population/community trajectory, capability of hindcasting past disturbances. From satellite data coupled with rapid ground-assessment, we will deliver habitat maps with meter-scale resolution that describe the spatial mosaic of reef assemblages, morphometric statistics to quantify the spatial complexity of reef units, and their relationships to one another (transition probabilities), bathymetric maps extracted directly from the satellite data, charts of rugosity to identify ‘hot-spots’ of three-dimensional complexity. Bathymetric data, in unison with the satellite imagery, will be used to derive a three-dimensional model of the reef surface. The satellite maps will provide quantitative information on the size, shape, and complexity of the different facets that make up the reef mosaic. This information has value in providing spatial context into which the small-scale work with phototransects can be placed, hence assuring representative sampling of the habitats present. Additionally, the spatial complexity of reef systems in comparable environments have been shown to follow mathematical scaling laws that facilitate the prediction of the heterogeneity from very large to very small scales. The existence of such predictability can be used to reveal the forces driving the geomorphology of the reef system, and in particular the likely existence of repetitive disturbances in the region, and the influence of antecedent (ancient) topography on the modern reef structure.

II.10. Mollusk Biodiversity: latitudinal and longitudinal patterns

Contacts: Prof. Daniela Basso and Prof. Elio Robba University of Milano - Bicocca (UNIMIB)

Keywords: Foraminifera, Mollusca, Scleractinia, death assemblage, biodiversity

Modern marine sediments contain the remains (shells or other skeletal parts) of several generations of benthic dwellers, which cumulate *in situ* or are introduced from adjacent sites by post-mortem transport, together with other sedimentary particles. Shelled gastropods and bivalves are usually the best preserved and dominant groups, and due to the time-averaging of multiple generations their associations are very diverse and frequently contain also rare or poorly known species. Therefore, sublittoral mollusks death assemblages are a surrogate of local biodiversity and their study is a fast way to depict patterns of geographic or environmental gradients. We will work on the biodiversity patterns of death assemblages of selected taxa of benthic dwellers occurring in the soft bottoms of reef environment. Our main objectives are: to obtain quantitative data on molluscan death assemblages from selected reefal and peri-reefal environments, along geographical gradients; to use molluscan death assemblage data as surrogate of local molluscan biodiversity, in order to identify geographic or environmental patterns; to compare patterns of molluscan biodiversity with patterns of coralgal biodiversity.

II.11. In situ impact of global ocean acidification: Study of massive coral growth

Contacts : Dr Stephanie Reynaud and Dr Eric Tambutté Centre Scientifique de Monaco

Keywords: calcification, banding, stable isotopes, trace elements

In addition to bleaching, a process leading to the disruption of the symbiotic association between coral and its microalgae, coral reefs suffer from seawater acidification. This process consecutive to the dissolution of the excess of carbon dioxide into oceans (linked to human activities) leads to a decrease of seawater pH. The pH decreased from 8.3 at the glacial time to about 8.1 today, and a pH of 7.7 is predicted for the next century. Some coral biologists even suggest an almost total extinction of coral reefs around 2100. It has been shown in experimental conditions that the rate of calcification of corals exposed to the predicted more acidic condition of pH 7.7 decreased by about 54 %. Unfortunately, only very few studies deal with *in situ* conditions. A large uncertainty exist on the field importance of ocean acidification on coral growth. The present project will aim to fill this gap by a general survey along the Tara route. Massive corals, like *Porites* genus, are important archives for geochemical proxies that have increased our understanding of past ocean chemistry and climate change and could be helpful to predict the future of coral reefs. For many years cores have been used in these species to study linear extension. Growth bands can be used as a chronological clock and the sum of a dark and a clear band equal one year. In this project, we propose to measure the linear extension (growth) by analyzing skeletal density banding of several colonies of massive *Porites* and revealing the bands by X-Ray radiography. Data should then be compared to other environmental data which are not yet available. For this reason we suggest to perform in parallel isotopic (□^{18}O, □^{13}C, □^{11}B) and trace elemental analysis (Sr/Ca, Mg/Ca) since these values are used as proxies of environmental parameters. Boron analysis will be performed on the same cores since its uptake in carbonates is controlled by *p*H. Indeed this parameter will be used to better understand the modification of the seawater chemistry at the site of calcification (within the coral). As a result, we expect to determine if calcification rate is presently decreasing or not since the last century (approximately 30 years). This study will give information on the effect of global change on corals (ocean acidification, temperature, etc.).

II.12. A proteomic approach to the study of bio mineralization in corals

Contacts : Dr Eric Tambutté, Dr Sylvie Tambutté, and Dr Didier Zoccola Centre Scientifique de Monaco

Keywords: Calcification, organic matrix, 2D electrophoresis, Zooxanthellae clades

Hermatypic scleractinian corals, together with calcareous algae, are responsible for the edification of coral reefs. Coral reefs show high calcification rates of about 2 to 6 kg of calcium carbonate per m^2 and per year. These special high rates of calcification are due to a biomineralization process related to symbiosis and more specifically to symbiotic intracellular photosynthetic zooxanthellae living inside the animal cells. In corals, the biomineralized structure is a calcareous exoskeleton which shows unique properties of resistance and formation. The formation of this exoskeleton results from a complex series of cellular and biochemical events which gives the biomineral a specific micro- and macrostructure. As for any other biomineral, the coral exoskeleton is composed of an organic matrix embedded in a mineral structure. The organic matrix is composed of sugars, lipids and proteins and is involved in crystal nucleation, growth and inhibition of growth. In corals, even if the data are scarce, it is known that the synthesis of organic matrix depends on a biological/genetic control but also that its composition is influenced by zooxanthellae. The aim of the study during Tara is to answer the following questions: Does biomineralization in a species varies between different geographical sites? Does biomineralization in corals show inter-genera characteristics ? Can organic matrix proteins be considered as a taxonomic criterion for corals? Do clades/sub-clades influence organic matrix composition and thus biomineralization? We will study the micro- and macroarchitecture of the skeleton of different coral species in different sites and we will compare the proteomic pattern in gel electrophoresis. In addition to the inter-sites comparison, we will also make an inter-genera comparison. In parallel, we will determine the genetic characteristics of the symbiotic zooxanthellae in the different sites and in the different genera. As results we expect to determine if 1) different corals show common specific matrix protein characteristics 2) organic matrix proteins can be used as taxonomic criteria 3) the clades/sub-clades influence the protein pattern of organic matrix. This study will provide information both on the mechanism of biomineralization, the taxonomy of corals and the role of zooxanthellae in biomineralization.

II.13. Abundance and biogeography of internal bioeroders in coral reefs of the Indo-Pacific region

Contacts : Cornelia Maier [1] and Aline Tribollet[2] Laboratoire d'Oceanographie de Villefranche sur mer (LOV)[1], Institut de Recherche pour le Développement (IRD)[2], France.

Keywords: Bioerosion, macroborers, microborers, euendoliths

Internal bioeroders, which comprise macroborers (sponges, polychaetes, sipunculids, molluscks, crustaceans) and microborers (cyanobacteria, algae, fungi), are inconspicuous but very abundant in coral reef ecosystems. They colonize all kinds of carbonates, live and dead, by burrowing actively the substrate (chemically and/or mechanically). In live corals, internal bioeroders show relatively strong taxon specificity for their host which suggests co-evolution. It is well recognized that bioerosion is one of the most important and active processes involved in carbonate dissolution and coral reef decline. In some coral reefs, bioerosion by internal bioeroders can be detrimental to coral growth, and can threaten reef framework stability. It has been shown recently that internal bioerosion is enhanced by anthropogenic and climate change factors such as eutrophication, heat stress and ocean acidification, these factors being detrimental to coral growth. Internal bioerosion has been identified as a responsible agent in some coral reef drowning. To date, bioerosion has been poorly quantified by *in situ* monitoring techniques, thus data on internal bioeroder diversity and abundance in coral reefs including remote and disturbed areas, are extremely important to acquire. Studying at a large scale, i.e. across the Indo-Pacific Ocean the abundance of internal bioeroders will improve the understanding of the main factors (e.g. substrate, temperature, nutrient concentration, ocean pH) affecting and controlling internal bioerosion, and ultimately reef decline. The Tara Oceans expedition could provide an ideal platform for such a study because complementary sets of data will be gathered in parallel on water chemistry, coral, and algal diversity. Spatial variation of the diversity and abundance of the different studied organisms including internal bioeroders will be compared among sites all together.

III. ORGANISATION OF FLOW DATA

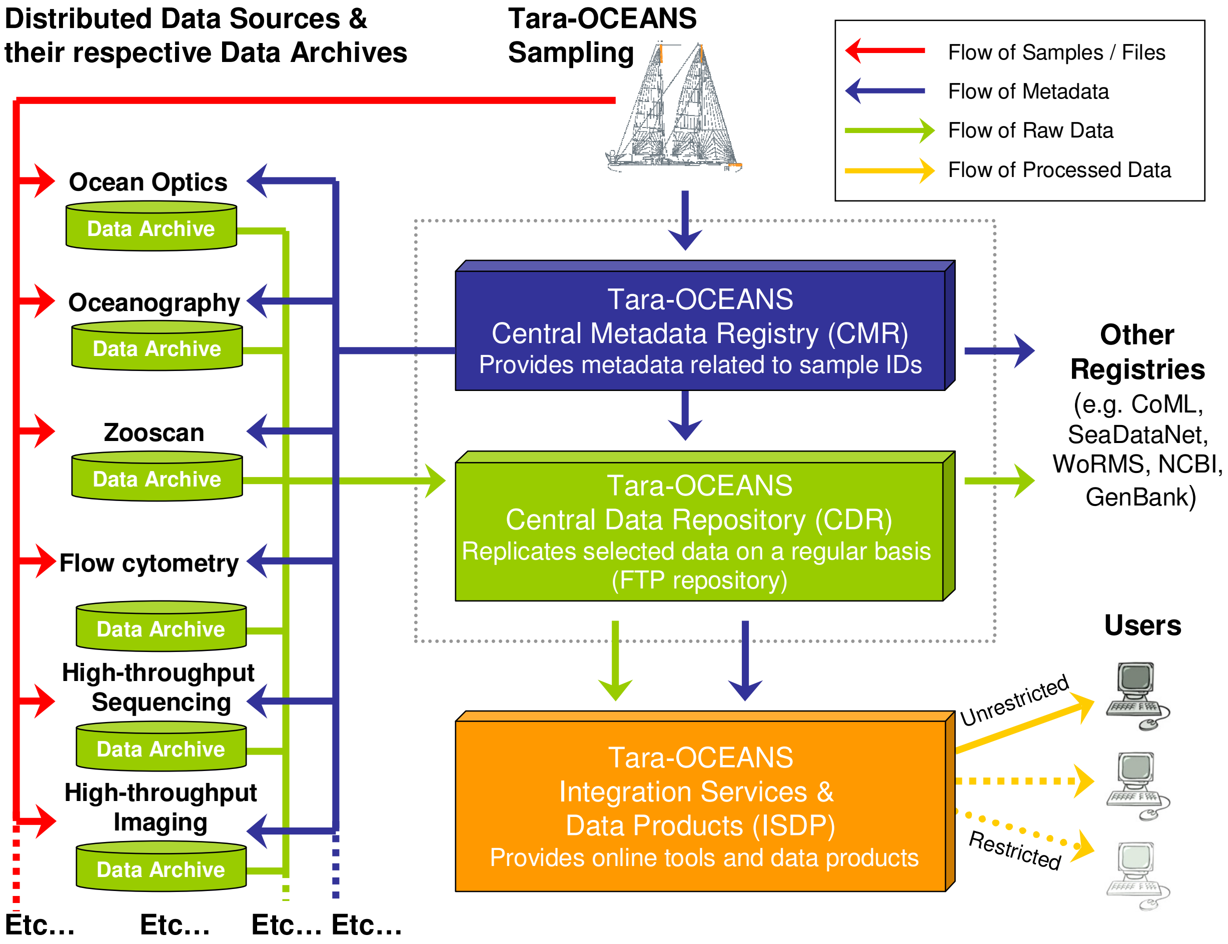


Figure Legend. The numerous samples and electronic records that will be generated onboard Tara will be distributed regularly (***red arrows***) to the scientific partners on land who will further analyse samples and electronic records, and archive data locally or in a data centre of their choice. Metadata about ***who*** measured ***what, where***, ***when*** and ***how*** will be recorded onboard Tara and will be archived in Tara-OCEANS' Central Metadata Registry (***CMR***) hosted at EMBL. These metadata are linked to samples and electronic records by a barcoding system. The CMR is therefore the key to track all samples, ensuring that scientific partners obtain (***blue arrows***) complete and up-to-date metadata for all samples and electronic records that they analyse. The CMR is also key to later assemble data generated by all scientific partners and to disseminate data and metadata to other registries regarding sampling events (SeaDataNet), biodiversity (CoML), taxonomy (WoRMS, ITIS and NCBI) and sequencing (DDBJ, EBI and GenBank). Raw data are quality checked by scientific partners before they are replicated (*green arrows*) into Tara-OCEANS' Central Data Repository (*CDR*), serving as a backup, share-point and data source for the development of Tara-OCEANS Integration Services & Data Products (*ISDP*) by specialised service provider (e.g. EBI, PANGAEA®) and expert communities (e.g. bioinformaticians, ecosystem modellers and taxonomists). The ISDP will include online tools (e.g. portals, warehouses) that allow users to access data (*yellow arrows*) and generate products (including some level of meta-analysis) that can be customised for both oceanographic and molecular biology communities, and most importantly, for their integration.

Contact: Stephane Pesant, PANGAEA®, Germany

IV. Expected Integrated Outcome

The approach and methods described above will first lead to an unprecedented discovery of new plankton species. This will also generate quantitative data about the relative abundance of organisms like viruses, bacteria, protozoa and small metazoans in various oceanic environments. Because we will use genomic methods, we will have access to the quantification of species in these ecosystems. Combined with imaging, the molecular data will bring new information about the correlation between shape and genome structures and most importantly about complex symbiotic events that occur in the sea as well as about horizontal gene transfer processes. Also, correlations between the presence of specific viruses and potential target organisms within various physico-chemical oceanic environments will provide important informations about the way oceanic ecosystems adapt to physico-chemical constraints.

The voyage will cross oceanic regions with very different pH (figure 3). We plan to study first the samples from ecosystems of areas having important pH gradients under various latitudes. Presently we do not know how pH variations may affect the susceptibility of protists or bacteria to virus attacks or how these pH variations may affect protists health. Because we will have access to recording of many other physico-chemical parameters, it is almost certain that some very interesting ecosystem patterns (e.g. in low oxygen systems or according to pH) will emerge. This should also tell a lot about the principles governing the evolution of microscopic life in the oceans. On the relatively long term (over 3 to 5 years from the beginning of the expedition) enough data should have been accumulated to build dynamic models of these ecosystems and begin to explore the relationship between climate and oceanic ecosystems evolution.

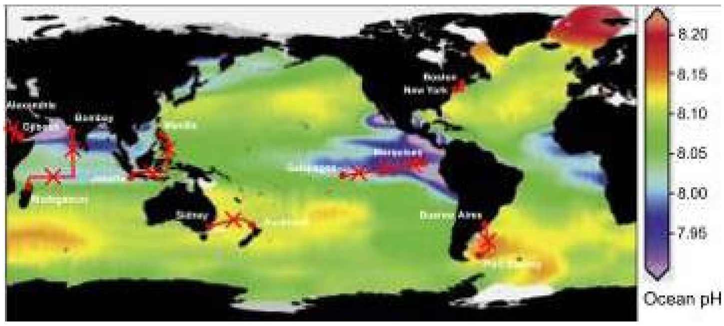
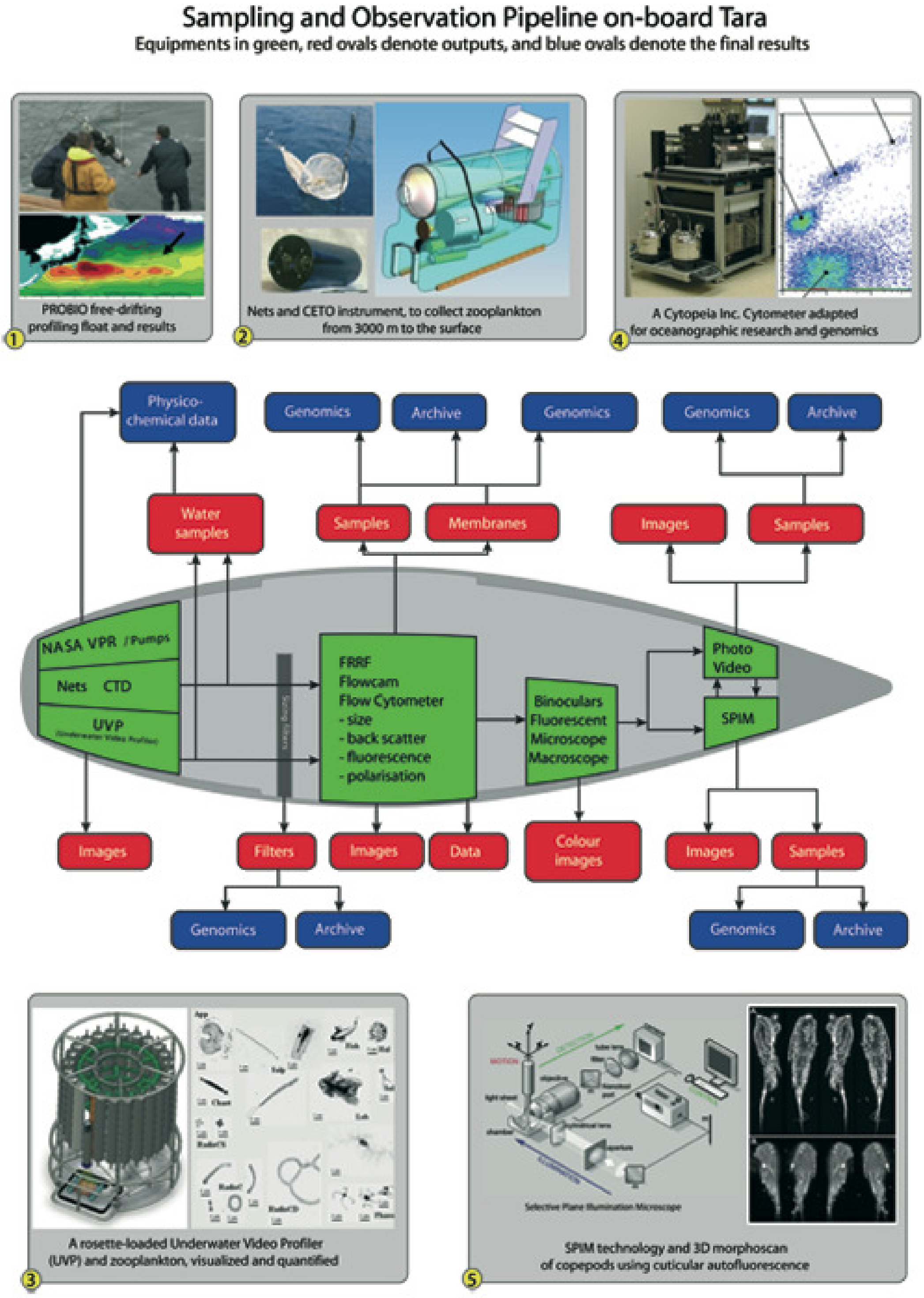


Figure 5: ***Location of 6 Tara-Oceans legs and 10 stations proposed, on a map of modern surface ocean pH.*** The mosaic of various pH domains is related principally to ocean circulation patterns.

Previous expeditions during the 19th century have had outstanding and long-lasting impacts on science and society (the expedition of the Beagle, and that of the Challenger for example). We expect the output of TARA OCEANS to become one of the most important oceanic metadata resources of the 21st century both for the scientists and through the importance of its legacy in terms of public awareness of the functioning of our planet.

Annexes
On board equipments



Sampling and Observation Pipeline on-board Tara

Equipments in green, red ovals denote outputs, and blue ovals denote the final results

① PROBIO free-drifting profiling float and results

② Nets and CETO instrument, to collect zooplankton from 3000 m to the surface

④ A Cytopeia Inc. Cytometer adapted for oceanographic research and genomics

③ A rosette-loaded Underwater Video Profiler (UVP) and zooplankton, visualized and quantified

⑤ SPIM technology and 3D morphoscan of copepods using cuticular autofluorescence

Physics/Chemistry	Zooplankton	Protists	Bacteria/Virus
TSRB Radiometry/hand CTD	Bongo 200um (500m-surface)	Pumping subsurface	CTD-Rosette HPLC (300 m)
CTD-Bio-optics (300 m)	Bongo 500um (500-surface) (2X)	Bongo 200um (subsurface, 2X)	CTD-Rosette DCM (2X)
CTD-Rosette HPLC (300 m)	Double Net (50 & 330 um) (100 - 0)	Bongo 20um (subsurface, 2X)	
long CTD Rosette (2000m)	Bongo 100/500um (500-surface) (2X)	CTD-Rosette (DCM) (2X)	
Imaging	Double Net (50 & 330 um) (100 - 0)	Closing Net 200um, 20um DCM (2X)	
	MultiNet (1000-0)		

The Imaging platform on Tara

The platform on board the sailing vessel is organized in « pipeline » allowing a analysis from cellular level to multicellular and planktonic populations.

Cytometry

- **FlowCam**, imaging in flux for fast and continuous sampling of micro-plankton
- **FRRF**, Fast Repetition Rate Fluorometer: biomass and photosynthetic activity measures
- **Cytofluorimeter TARA Influx,** in flux continuous measure of pico-nanoplankton organisms
- **Bio-optic system,** for nano and macro-plankton photosynthetic activities detection and dissolve and particular material.

Macro-microscopy plateform

- **Fluorescence** m**icroscope and macroscope:** imaging micro-organisms, protists and metazoairs after selection, dissection.

Microscopy with 3D confocal plateform

- **SPIM**: Single Plane Illumination Microscope for three dimension imaging

Macrophotography and HD video platform

Documentation on morphology and behaviour of plankton organisms.

In situ imaging (open ocean)

- **UVP,** Underwater Vision Profiler, for detection *in situ* and quantification of organisms and particules
- **VPR,** Vidéoplankton Recorder, and means of acquisition and analysis classification
- **Zooscan,** plankton analyseur for fixed samples to be completed by lab analysis.

Tara Oceans’ Map