

MICROBIAL OCEANOGRAPHY
Ecology and Diversity of marine Microorganisms (ECODIM VIII) of ASI
January 6 - 25, 2014
at the Marine Biological Station Dichato of the University of Concepción (UdeC), Chile
Course Report



14 students, 15 full time and part time instructors, technical assistants and symposium speakers from 7 countries (Chile, Argentina, Brazil, Colombia, Ecuador, Cuba, France, Spain, USA, The Netherlands and Switzerland) participated in the ECODIM course this year. The course is part of the International Graduate Course Series in Oceanography offered by the Austral Summer Institute of the University of Concepcion, Chile.

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It was the first time that the course could return to its original home in Dichato after the entire facility and scientific equipment were washed to sea by the 2010 Tsunami.

The former research and lecture building was completely destroyed and has not been rebuilt yet, but restored buildings and labs are well suited to offer courses in Dichato again.



Course Announcement & Organization

The advanced course on the ECOLOGY AND DIVERSITY OF MARINE MICROORGANISMS (ECODIM) was offered for the eighth time as part of the International Postgraduate Course Program in Oceanography, organized by the Universidad de Concepción. The course was announced early 2013 with a poster (Appendix 1) that was sent to academic institutions world-wide and on the Internet under the addresses <http://www.profc.udec.cl/ecodim/> and <http://www.isme-microbes.org/events/meetings/past>. Relevant course information from past courses was also provided at the address <http://www.microeco.uzh.ch/chile/chile.html>. The latter site will continue to be available for students with the intention of establishing lasting interactions among ECODIM alumni across geographical and institutional borders and to promote collaborations in the fields of microbial ecology in oceanography.

The course was organized by the 14th Austral Summer Institute (ASI XIV) of the Departamento de Oceanografía, Universidad de Concepción (UdeC). For the first time after the destruction of the Dichato station by the Tsunami of 2010, the course was held again in Dichato. Lectures took place in the recently completed PIMEX building and the research was carried out in the restored Laboratory Building. It will still take a few years to reinstall the full operation of the station, but by moving laboratory equipment temporarily from Concepción to Dichato and maintaining a rapid supply service from the main campus to the station it was possible to fulfill the requirements for course work almost as in the past.

The course was directed by the team of instructors and technical assistants listed in the appendix (A2 and A6). Organizational details are provided in A3. The course was held under the auspices of the Universidad de Concepción. Funding for the course and the minisymposium was provided by the Agouron Institute, the Gordon and Betty Moore Foundation, the Graduate School of UdeC and the Faculty of Biological Sciences of PUC and the University Mayor.

Making an excellent ZEISS microscope available free of charge and for the entire duration of the course by the Reichmann company, and the expert advice by Ger van den Engh from BD contributed significantly to the success of the course.



This year's studies focused on the water column and the sediment in the redox transition zones in two time series station on the shelf in Concepción Bay.

Course Participants and Staff

The 2014 course was attended by 14 students from 11 different academic departments in 7 Latin American and European countries (Argentina, Brazil, Chile, Colombia, Ecuador, Cuba and Spain). The course was open to graduate students and interested professionals. 15 students from a total of 48 applicants were selected on a competitive basis. One participant had to quit at the beginning of the course due to personal problems that interfered too much with his full commitment at the course. Finding a replacement in a short time was, unfortunately, not possible. In addition to the selected participants listed in A5 ASI also received applications from Croatia, El Salvador, Pakistan and the United States. The list with the full names of the selected participants, their home institution and their research interests are given in A5.

The course was taught by instructors from Chile, France, the USA and Switzerland (A6). Speakers for the course symposium and for special lectures came from Chile, Spain, the USA and France (A8). Most symposium speakers spent some extra days with the course and got involved in lectures and seminar-type activities with the students. The course instructors stayed with the course for 8 to 22 days.

Course Description and Structure

The main goal of ECODIM courses is building intellectual capacity, opening minds of young scientists for research and to transfer knowledge in areas of growing scientific interest in marine microbiology. This is achieved by teaching concepts and providing an overview of the fields of microbial ecology, marine genomics, the diversity of microorganisms and their roles in biogeochemical cycling. This year's course brought together various aspects of environmental, molecular and microbiological, as well as chemical, physical and bioinformatics methodologies needed to approach questions of geochemical, ecological, phylogenetic and genomic interests. For the first time, we were able to include marine virology as a course topic. In order to not overload the three week program, we had to reduce aspects of culturing microorganisms, unfortunately.



The most efficient training: Learning from each other in the field, in the laboratory and in the lecture hall

The course comprised field experiences, lectures and exercises in the morning, guided research work in the laboratory and tutored computer exercises in the afternoon, colloquia and open study time in the evening and two symposium days, one by invited speakers on January 10 (The Microbial Tree of Life and Beyond: The Legacy of Carl Woese 1928-2012), the other organized by the students themselves on January 24 (Discoveries in Microbial Ecology). Preparatory discussions, exercises on particular course subjects, introduction to bioinformatics software packages, computer aided thermodynamics in geochemical processes and metabolism were offered as exercises in small groups. With the didactic mix between lectures and talks by experts, workshops with tutorials and the students' own efforts (formulating a research proposal, presentations, experimental design, practical laboratory work, summarizing research results, designing posters, writing abstracts), course participants were introduced into topics from the research front in microbial oceanography, but still were taught the physico-chemical and biological concepts that underly research results. Students were also asked to suggest special topics and thus to define their particular needs.

During the first week, the students introduced themselves with a short presentation about the research topics that They carry out at their "home institution" and their personal research interests. This activity provided to the participants and to the staff an overview of the background knowledge of each student.

On a one-day sampling cruise, we determined a number of environmental parameters at two sites on the continental shelf in Concepción Bay and collected inocula for experiments with pico-eukaryotes as viral hosts and nutrient-dependent growth. On a cruise to nearby coastal water sampling stations, some course participants gained first-hand experience of CTD measurement and water and sediment sampling and observation techniques of coastal and shelf environments. All students were exposed to these marine habitats and collected samples for the extraction and isolation of DNA. With the aid of molecular techniques, they investigated the presence of major genes and examined the morphological and functional diversity of microbial communities using DAPI staining and advanced epifluorescence microscopy and flow-cytometry. For the first time in the history of ECODIM, we made an attempt to enrich for viruses and planktonic microbes employing fractionated filtration techniques.

An interactive website for the exchange of course materials, lecture notes, papers, exercises etc. was created for this year's course on OLAT (A4). It will remain active for a few months after the course has ended.

The laboratory part was designed to educate students in current techniques and to encourage independent research. Students carried out investigations in groups and independently with the aid of faculty and teaching instructors. The course culminated with the student project presentations; results are summarized in posters (Appendix A13 – A16).

By writing a research proposal, students were trained to search and review the published literature via the internet, to screen and select the proper information, to quickly review the papers, to understand and extract key elements, integrate them in a didactically logical way into a research proposal, be able to present it to the course and respond to questions relating to the topic.

Course Contents

Introductory lectures and practical exercises emphasized the chemical, phylogenetic, metagenomic and energetic basics of marine microbial ecology, photosynthesis and ocean biogeochemical cycling, as well as the molecular and culturing methodologies, microscopy and flow cytometry for the study of the molecular ecology of microorganisms in ocean waters and sediments. The 2014 course addressed close to 30 topics in lectures, workshops, tutorials and minisymposia (A7, A8, A9).

ECODIM-VIII focused on topics with the following contents:

- Microbial Ecology:** (Osvaldo Ulloa, Ger von den Engh, Rodrigo De la Iglesia, Alejandro Murillo, Kurt Hanselmann) Field observations and collection of environmental determinants (metadata); nutrient and energetic constraints in ocean habitats; molecular evolution of biogeochemical processes (N-, S-cycling); microscopic and cytofluorometric analyses of population changes; techniques for growing and isolating microorganisms.
- Diversity:** (Daniel Vaultot, Norman Pace, Kurt Hanselmann) Molecular and microscopic methods for the identification of OTUs and microorganisms from all three domains of life and viruses, pigment analyses; collection of DNA and its sequence analyses; detection and description of genotypic diversity; studying phylogenetic relationships and constructing trees based on statistical clustering techniques.
- Virology:** (Matt Sullivan) Role of viruses in nutrient cycling, regulation of structure, size and dynamics of microbial populations; gene flow.
- Genomics:** (Nicole Trefault, Daniel Vaultot, Matt Sullivan, Adriana Lopes Dos Santos, Alvaro Plominsky) High throughput sequencing approaches; bioinformatics, data analyses and annotating genomic information; database searches for full genome and metagenome comparison.
- Biogeochemistry:** (Osvaldo Ulloa, Alejandro Murillo, Kurt Hanselmann) Metabolic potential of microorganisms, relating in situ measurements to mass fluxes, designing geochemical cycles, bioenergetics,.
- Bio-Informatics:** (Daniel Vaultot, Nicole Trefault, Adriana Lopes Dos Santos, Rodrigo De la Iglesia) Introduction to statistical sequence analysis and presentation packages, multivariate analyses, presenting data with the r- software package)
- Academic Skills:** (Alvaro Plominsky, Carles PedrósAlió, Kurt Hanselmann) Basics of writing research proposals, poster design, presentation rules.
- Mini-Symposium:** (Norman Pace, Linda Amaral Zettler, Carles PedrósAlió, Matthew Sullivan, Daniel Vaultot) To honor the work of Carl Woese, special program (A8)

Short introductions to the particular conditions in oxygen minimum zones (OMZ) along the continental shelf of Central Chile during the first days were followed by a one day cruise to collect samples from the water column and sediments at two stations off Dichato.

Computer labs were designed to familiarize students with the most common data bases available for phylogenetic and metagenomic analyses. The tutorials focused on the design and validation of nucleic acid probes and the application of bio-thermodynamic models to examine metabolic processes in geochemical cycles.

Lectures and tutorials were delivered by the course instructors (A6). The goal of all lectures was to illustrate how basic concepts in microbial ecology and geochemistry can be applied to a number of questions relating to microbial oceanography. Additional speakers were invited to deliver special lectures during the mini-symposium in Concepción on January 10 (A8). All lectures and the details of the course activities are outlined in the weekly schedules (A7). Most speakers made their slides available for personal use by the course participants.

Places visited for Field Work

This year, the main course topics dealt with the richness of microbes and their metabolic activities at two time series station in the near shore waters of the South Pacific Ocean of Central Chile. This allowed us to study the regulation effects of viruses on community selection.

The water column and the sediments at the sampling sites represent redox transition, in particular oxygen depletion zones. Important ecological changes occur through this gradient: changing biodiversity with decreasing oxygen availability, high density of prokaryotic cells under oxic conditions and short food chains. Each redox zone contains characteristic microorganisms that are well adapted to live under the particular conditions.

Course Research

The research part in the lab focused on growing pico-eukaryotes, heterotrophic Bacteria, Archaea and Viruses from the above mentioned ecosystems, isolating DNA for molecular analyses and ribosomal RNA gene detection employing PCR methodology with corresponding primer sequences and RFLP with a number of endonucleases, microscopy, flow-cytometry, and elucidating the regulation of growth in microcosm experiments. It was the goal to study the trophic and organismic interactions, and changes and the role microbes play in the geochemical cycling of matter in the particular marine environments. The techniques were instructed to small groups in rotations. Molecular, flow-cytometric was applied in particular for the study of the diversity and abundances of marine pico-eukaryotic microbes. The techniques were applied to small research projects, and their range of applicability and the limitations were studied by four student research groups. Course participants gained experience in the collection and preservation of samples in the field, they were trained in designing experiments, including the use of controls and replication, in maintaining microbes alive and in the analysis and interpretation of results using phylogenetic methods as well as biosystem modelling with the aid of databases and in bio-thermodynamics applied to the understanding of microbially mediated processes in biogeochemical cycles.

The results of the student projects are summarized in posters, which are available as appendices A13 – A16.

Group A:

Response of the microbial Community of the Oxygen Minimum Zone (OMZ) in the SE Pacific, Chile, to upwelling Conditions: empirical and molecular Approaches.

Group B:

Exploring pico-eukaryotic Viruses in marine Upwelling Systems.

Group C:

Are Nitrate and Phosphate limiting Growth of Pico-Phytoeukaryote Assemblages in the seasonal coastal Upwelling Zone off Central Chile?

Group D:

Viruses regulate microbial Communities in the Surface Ocean in the Upwelling System off Central Chile.

Mini-Symposia

A one-day symposium entitled “THE MICROBIAL TREE OF LIFE AND BEYOND: THE LEGACY OF CARL WOESE (1928-2012)” took place as a „public“ event offered by the course on Friday January 10 at the main campus of the Universidad de Concepción in Concepción. It was organized by Nicole Trefault from the Universidad Mayor and Rodrigo De la Iglesia from the P. Universidad Católica de Chile and took place in the auditorium “Claudio Gay” of the Facultad de Ciencias Naturales y Oceanográficas. The detailed program is enclosed (A8). Established investigators representing different research institutions from France, the USA and Spain presented their research, related it to the importance of Carl Woese’s phylogenetic approach, stimulated discussion on newest developments, initiated new research ideas among the symposium participants and strengthened interactions between different research groups and centers. The speakers were Norman Pace, Linda Amaral Zettler, Carles PedrósAlió, Matthew Sullivan and Daniel Vaultot. Steven Hallam from the University of British Columbia, Canada, had to cancel his participation for personal reasons in the last minute. The course students were in charge of introducing the guest speakers and hosting them for after-symposium discussion groups. The symposium was sponsored by the Agouron Institute, the Gordon and Betty Moore Foundation, the Austral Summer Institute, the Universidad Mayor and the P. Universidad Católica de Chile.

At the end of week two the four student working groups presented the proposal that connected their actual course research to what is known from the scientific literature. We consider writing a proposal an essential skill in scientific research and tried this for the first time in an ECODIM course. We introduced the process and the procedures in a seminar and are pleased with the proposals that were submitted. Each group followed the guidelines to address four parts:

- What is the question to be investigated? (Definition of the problem, concepts)
- What can you learn from the scientific literature that relates to your question? (Background information)
- How do you intend to solve the question experimentally? (Approaches, methods employed)
- What results do you expect and how can they be integrated into the existing knowledge base? (Discussion of results and conclusions)

A second mini-symposium (DISCOVERIES IN MICROBIAL ECOLOGY) was organized in Dichato as a course activity on January 24, with 4 group presentations offered by the course participants themselves. Each student group reported on the preliminary research results obtained during the course and linked it with the background information assembled in the research proposal. The one-day symposium was also open to interested researchers and students from other UdeC research groups. The titles and the symposium program are available in A9.

Student Evaluation

All participants were highly-motivated, actively-participating, hard-working students who expressed great interest in every aspect of the subjects which were taught. We expect ECODIM students, who successfully completed the course to become leaders in their fields in their countries and to contribute significantly to marine microbiology in the coming years. Since the course was taken by several students for credit, an evaluation was made for all students. Full credit required:

- a) giving a 2 minute „Elevator Talk“ followed by a 15 minute presentation on the work the student is presently involved in at his/her home institution (at the beginning of the course).
- b) presenting the essence of a research proposal, selected and formulated by each student group, in 30-40 minutes (incl. discussion) in English and being able to respond to questions related to it. The proposal had to address the research questions that were investigated during the course (microbiology, virology, ecology, microbial diversity in ocean habitats) and the particular scientific interest of the students (counted as group effort). The four proposals were reviewed by instructors and are available in written form to all course participants. The contents became part of the final research presentations as listed in A9.
- c) the presentation of the course research results and the contribution to the final poster design (counted as a group effort)

All students passed these exam requirements and were given credits for a grade A or B (A10), according to the scale recommended by UdeC

Grade scale:

Points	Grade	Scale	Points	Grade	Scale
100 - 88	A	excellent	< 50	D	insufficient
87 - 68	B	good	0	F	failed
67 - 50	C	sufficient		I	incomplete
				X	withdrawn

Students, assistants and instructors received a certificate for successful passing and participating to the course, respectively (A11)

Thanks

It is one of the most important components of an intensive course like ECODIM that the group spirit remains high in spite of the late hours and sometimes the shortage of sleep. A special thank goes to all those who supported the course in one way or another. Uncomplicated interactions, friendship and a great hospitality made the course work enjoyable, easy, productive and memorable for the students as well as for the instructors. Investigators from UdeC contributed to the course by sacrificing research time, equipment, and funds for course experiments. Everybody was very helpful in getting together the necessary infrastructure for the lab, the computer exercises, and the field work in a timely manner.

The course profited immensely from the hard work and dedication to ECODIM of:

- MONICA SORONDO, the course coordinator,
- ESTEBAN ALARCÓN who guaranteed safe transportation,
- BRUNO JEREZ from W. Reichmann Y Cía. Ltda. for the use of the Zeiss microscope,
- NATHALIE DELHERBE MARTÍNEZ, a 2010-student, who dedicated her love to ECODIM 2014.
- ALEJANDRO MURILLO, a 2006 ECODIM student who introduced his postdoctoral project on the OMZ to the course and helped on the sampling missions.
- The staff of the CABAÑAS who cared for our meals and accommodations.
- ALL instructors, technical specialists in the lab and on the research vessel, and speakers of the course; they are listed in A2 and A6.

Outlook, Outreach and Recommendations

January is a good time for an intensive microbial oceanography course like ECODIM, because the students have finished their academic commitments for the year and could fully dedicate their time to the course. The facilities in Dichato still suffers from the damages of the 2010 Tsunami, but great efforts are on the way to restore buildings and improve the infrastructure. In a few years the Dichato site might be fully operational again to host activities like the Microbial Ecology and Diversity Course. For the 2014 course the laboratories were temporarily equipped with the necessary instruments and internet connections that served the course needs. Students and instructors were housed in Cabañas near the station. Keeping the entire group together favors active exchanges and leads to full dedication to the course objectives.

Most of the demands, which emerged from previous courses could be fulfilled again. For future ECODIM courses it is recommended:

1. that the basic structure of the course and the timing be maintained and that its duration be extended by a few days at the end,
2. that the mini-symposia be maintained as a means of introducing the students to frontier research, with the participation of additional speakers from abroad,
3. that enough time be reserved for the course research project and other laboratory work, since these are activities during which the students get practical experience and when questions emerge,
4. that enough independent study time be integrated into the program for reading, discussing key papers and formulating a research proposal,
5. that a facility allowing for larger scale growth experiments, the culturing of anaerobic organisms, and the sorting of cells into growth media, be made available,
6. that relations with equipment and supply companies be used further for course purposes. Lending modern equipment, microscopes etc. to an international course and having students work with good reagents and molecular products is attractive for the companies involved as well as to provide state-of-the-art materials for the course.
7. The need for fast Internet connection for journal and library access, as well as for bioinformatics and modelling exercises which need software that is only available online remains a necessity for future courses.
8. Thanks to generous grants from the Agouron Institute and the Gordon and Betty Moore Foundation it was possible to allow 14 students to work together with experts and efficiently on a research project in an interdisciplinary group for 3 weeks.

Given the favourable circumstances, the strong financial support by a number of institutions (A1), and the dedication of people from various university campuses, the course instructors were able to achieve the course objectives with almost everything they had intended to do. The organizers were glad that ASI decided to offer the course, in spite of the still reduced activities at the Dichato station. Since it will probably still take a few years to rebuild Dichato completely, we are thankful for the flexibility of the Oceanography Department to help out with equipment, chemicals and personell.

Since its start in 2000, ECODIM has established itself as a widely recognized training opportunity for marine microbiology in South America. A number of people, who were tained by ECODIM are now active in the field in Chile and in numerous other South American countries, in the US and in Europe. This not only strengthens the field, it also assures the successful continuation of ECODIM. The course has also gained attention outside of the field of marine sciences and microbial ecology through coverage in public media.

Students' responses to the course content, the teaching approaches and the facilities were positive and encouraging. The contacts which have been established among the students and between the students and the instructors will last into the future and may lead to study periods abroad or to joint research activities with people who met during ECODIM-VIII.

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Dichato, January 27, 2014

Appendices (pdf files)

A1_ECODIM-VIII_2014_announcement_poster.pdf A2 Organizers

A2_ECODIM-VIII_Organizers.pdf

A3_ECODIM-VIII_2014_Organization.pdf

A4_ECODIM-VIII_2014_website_welcome_page.pdf

A5_ECODIM-VIII_2014_Student_interests.pdf

A6_ECODIM-VIII_2014_Teaching_Team.pdf

A7_ECODIM-VIII_Course_Schedule.pdf

A8_ECODIM-VIII_Symposium_Program.pdf

A9_ECODIM-VIII_2014_Student_symposium_140126.pdf

A10_ECODIM-VII_2012_Grading.pdf

A11_ECODIM-VIII_2014_Certificates.pdf

A12_ECODIM-2014_Report.doc

A13 ECODIM-2014_Poster_Group_A

A14 ECODIM-2014_Poster_Group_B

A15 ECODIM-2014_Poster_Group_C

A16 ECODIM-2014_Poster_Group_D