CORSACS
(Controls on Ross Sea Algal Community Structure)

Science Plan & Project Descriptions

Science Plan

Major goals of the project can be divided into three areas of research including the initial transect sampling, manipulative deckboard experiments and our survey work. The initial transect sampling work will include: (1) test CTD and trace metal clean rosette deployments, (2) trace metal profiles in the ACC, (3) ice algae sampling in the pack ice, (4) IVARS transect stations, and (5) IVARS mooring deployment. The manipulative deckboard experiments will include batch incubation experiments, semi-continuous and continuous culture experiments focusing on the interactive effects of iron, light and CO_2 on algal community structure and various biogeochemical parameters. Our survey work will allow us to occupy stations in distinct biogeochemical regimes. We will strive to find both high and low pCO_2 regions as well as areas that are dominated by either *Phaeocystis* and/or diatoms.

Initial Transect Sampling

Underway sampling will begin for some groups across the Antarctic Circumpolar Current (ACC) en route to the Ross Sea. Tentatively, we plan to transit south close to 170^o E. This will be an ideal time to test all analytical equipment, especially the CTD. We will stop and perform a test CTD station somewhere in the ACC (weather permitting). All groups that intend to collect CTD water in the Ross Sea should do so at this test station. Thus, we will treat this deployment as a real station. We will need to elect a "water cop(s)" to orchestrate the sampling in the Ross Sea (no previous experience is necessary to fill this position but a cheerful disposition is a must). Mak Saito will also deploy his trace metal rosette sampler on some short casts several times across the ACC. Within the pack ice we will also stop to collect some sea ice algae samples for biogenic sulfur measurements. Anyone else interested in collecting ice samples should make their intentions known ASAP to the chief scientist. This ice sampling will only be performed a few times.

Upon entering the Ross Sea our first goal will be to complete the IVARS transect and deploy moorings in the southern Ross Sea. Hence, our second station is tentatively planned for 76^o 54.57'S, 171^o 47.04'E (we will not designate the test stations in the ACC or the ice sampling sites with station numbers but rather with the letter notations of 1a, 1b

1c etc…). This labeling will also correspond to the IVARS station numbering scheme as we will occupy and label IVARS station No. 2 as our CORSACS station No. 2. We will transit SE and perform 11 stations ending at 77^{o} 51.92'S, 177^{o} 52.66'W (Station 11). This transit and mooring deployment should be completed in approximately 24-36 hr. IVARS sampling will have precedence on the CTD water. However, there should be adequate seawater available for others to sample. We will calculate a CTD water budget on the transit south. Please estimate your CTD water needs. If any major analytical problems arise with instrumentation on the transit south we should be fairly close (about a 12-36 hr steam depending on ice conditions) to helo support when we arrive at Station 2.

Following the completion of the IVARS stations, our next objective will be to collect seawater for manipulative deckboard experiments. Our survey of the IVARS transect line might provide us with an ideal site to reoccupy and collect water for the incubations. Underway sampling for dissolved iron (Sedwick) and CO_2 (Dunbar) during the IVARS transect should reveal the relative concentrations of these compounds in this region. Ideally, for our manipulative experiments we are looking for a site that has low Fe (ca. 0.1 nM), low CO_2 (ca. 150 ppm) and a mix of both diatoms and *Phaeocystis* present in the near surface waters (upper 10 m). If we do not find these conditions along the IVARS transect then we will utilize satellite imagery to make our best guess of where these conditions are the closest.

Manipulative Deckboard experiments

Once we find our "ideal" station we will perform a CTD to characterize the water column. At this point, trace metal clean seawater will be pumped onboard using the towfish system (Sedwick). Tentatively, we plan to pump clean seawater into either the trace metal clean working area of the Hutchins van or a clean sampling "bubble" area will need to be constructed. We will remain on this station indefinitely until all bottles have been filled and all experiments have been successfully initiated. At this point it is not clear how long these deckboard experiments will last. The chemostat experiments may go the longest (probably with a minimum of 14 days). Depending on results from the first experiment and other factors, it is possible there will be sufficient time to start a second chemostat experiment. We will convene periodic PI meetings to discuss the status of the deckboard experiments and the timeframe for starting additional experiments.

Southern Ross Sea Survey

We will use satellite imagery to guide our survey track of the southern Ross Sea. Ideally, we want to sample in waters that are high and low in CO_2. Iron concentrations and pCO_2 at this time of year should be relatively low in many regions that we sample in the southern Ross Sea. We will perform a grid with uniform spacing of stations to allow adequate mapping of our study area. We are tentatively scheduled for refueling at the ice edge on January 15. As a result we will need to remain somewhat flexible on our proposed transect work.

Project Descriptions

DiTullio Group, B-272, (ditulliojj@cofc.edu)

Participants:
Jack DiTullio (CORSACS, Chief Scientist)
Peter Lee, Post-doc
David Jones, Research Associate
Aimee Neeley, Research Assistant
Jay Francella, Grad student
Brian Taylor, Research Assistant

Our group will focus on the interactive effects of iron and light on phytoplankton community dynamics. We will perform batch growout experiments utilizing an Fe-UV light matrix. Samples from these experiments will be collected for HPLC pigment analyses, nutrients, biogenic sulfur compounds, FRRF measurements, flow cytometry measurements (algal and viral) and mycosporine amino acids (MAA's). We will work closely and collaborate with both the semi-continuous (Tortell) and continuous culture experiments (Hutchins) investigating the interactive effects of light, Fe and CO_2 on algal pigments and the biogeochemical cycling of various sulfur compounds (including DMS, DMSP and DMSO). We will also measure the DMSP lyase activity in phytoplankton as an indicator of oxidative stressful conditions. Results from the manipulative experiments will be compared with transect data.

Sea ice algal samples will be collected for biogenic sulfur measurements. These samples will be primarily collected in the pack ice on our transit south from New Zealand. Underway sampling on transects will include: HPLC pigment samples, FRRF, flow cytometry samples and biogenic sulfur compounds. We will also use both HPLC analyses and molecular methods to ascertain the physiological state of the algal population with respect to iron based on the ferredoxin/flavodoxin ratios.

Dunbar Group, B-258, (dunbar@stanford.edu**)**

Participants:
Rob Dunbar, P.I., Stanford University
Dave Mucciarone, Lab Manager, Stanford University
Eduard Costa, Post-doctoral fellow, Stanford University
Matt Long, Ph.D. student, Stanford University
Christina Riesselman, Ph.D. student, Stanford University

Interests:
C system dynamics, in batch incubators as well as in the water column How does primary production vary in space and time in the Ross Sea relative to community structure and environmental forcing Community responses to interannual variability (and to our perturbation experiments) in terms of diatom species and diatom/Phaeo ratios
Controls on isotopic composition of diatoms and Phaeo.

Workplan:
Our group will measure inorganic C parameters in samples obtained from 1) the underway uncontaminated seawater (TSG) line, 2) the CTD rosette sampler for depth profiles from the water column, and 3) inflow/outflow waters from the batch processors. The goal is to measure rates of net community production and respiration and rates of drawdown of pCO2. We are also prepared to measure very small levels of net community calcification/dissolution if we encounter a carbonate community. Associated with the basic C rate measurements as described above, we will take selected samples for stable isotopic analysis (N15 and C13) of POM, DIC, DOM. In this regard we are particular interested in the time evolution of isotopic composition in evolving communities or in end-member communities (e.g., dominated by a single taxa). One goal here is to better understand how to use stable isotopic analyses of seafloor sediments to examine past changes in community structure and rates of primary production. Our group has built quite a bit of specialized semi-automated sampling gear for the cruise – along the lines of what we said we would provide in Table 3 of the proposal. We have 3 independent systems for measuring TDIC (2 coulometers and 1 IR system). The automated coulometers have been regularly achieving a reproducibility of 0.2 to 0.3 µmol kg-1 with a cycle time of about 8 minutes/sample. The Licor IR system is faster but a little less precise (1 µmol kg-1). Our optical pH system has a precision of about 0.002 pH units. We also have a precise spectrophotometric automated discrete sample alkalinity system that yields a reproducibility of ± 0.5 µmol kg-1. If you know this business, you will know that these numbers are about as good as anyone can get and in fact is a little better than the WOCE standard.

Christina Riesselman has been to the Ross Sea before and is working on diatoms from the seafloor sediments for one part of her dissertation. She is an ARCS scholar and has been awarded an ANDRILL graduate fellowship. She has been well-trained in Amy Leventer's lab and is interested in the distribution of diatoms in the Ross Sea water column during our cruise and is available to do microscope work in support of the overall project as needed. Matt Long is a 2nd year Ph.D. student who will make the core C system measurements an important part of his thesis. He is interested in following up on some ideas developed in part within Colm Sweeney's and Stephanie Rubin's papers. Eduard Costa is a post-doc from Barcelona, Spain. Eduard holds a Spanish Fullbright award and is an expert in radiochemistry. On this trip he is helping with the C cycle measurements. His main work at Stanford as a post-doc involves work on jumbo piston cores through Holocene sections that we collected off East Antarctica from the NBP in 2001.

Hutchins group (dahutch@udel.edu)

Participants:
Dave Hutchins (PI),
Clint Hare (grad student),
Yuanyuan Feng (grad student),
Sara Handy (grad student),

Julie Rose (postdoc)

My group will take responsibility for CHN, biogenic silica (BSi) and particulate organic phosphorus (POP) core measurements for the transect and experimental sampling. POP will be measured as both total POP (seawater washed) and "interior" or "intracellular" POP (oxalate-washed). Student Sara Handy will take primary responsibility for coordinating our core analysis sampling.

Our main experiments will use our two shipboard continuous culture systems to examine the effects of varying pCO_2, iron concentration, and light availability on algal community structure and biogeochemical parameters. In keeping with the general aim of the project, we hope to examine experimentally how these three variables may affect shifts in diatom vs. *Phaeocystis* abundance. Our experimental matrix will consist of: 1) present-day CO_2 (~380 ppm) and 750 ppm CO_2, 2) Low light (~10% of incident?) and high light (~50% of incident?) and 3) Ambient Fe (~0.1 nM?) and Fe-enriched (1 nM). We will accomplish this by running the two chemostat incubators at two light levels; within each incubator, 12 incubation bottles will be run in a triplicated complete CO_2/Fe matrix. We will try to run two complete experiments of up to 2 weeks each; we may want to substitute temperature for light as a variable in the second set of experiments. Input from collaborating groups on the detailed design of these experiments is welcomed. These experiments will be closely coordinated with the semi-continuous experiments being carried out by the Tortell group, and we're counting on the other cruise participants for experimental sampling and analyses of Fe, CO_2, HPLC, sulfur, P vs. E, Fv/Fm, and Chl. All of Hutchins participants will be involved in these experiments, but particular responsibility for them will fall on Yuanyuan Feng and Clint Hare.

Postdoc Julie Rose is supported on a newly funded OPP fellowship to collaborate with our experiments. Her project will examine the effects of changes in temperature, CO2, light and Fe on top-down control of phytoplankton by microzooplankton growth and grazing. Hypotheses being tested in her experiments include 1) Microzooplankton growth and grazing is strongly affected by low temperature relative to the growth rates of phytoplankton, which reduces top-down control of phytoplankton in cold waters and contributes to the formation of algal blooms. 2) Temperature changes of a few degrees Celsius in a perennially cold ecosystem such as the Ross Sea would be enough to significantly increase rates of herbivory. 3) Shifts in present-day phytoplankton community composition in the Ross Sea due to light or iron availability may cause significant changes in rates of herbivory. 4) Future potential shifts in phytoplankton community composition in the Ross Sea due to climate change-induced increases in temperature and pCO2 levels would also result in significant changes in rates of herbivory. 5) Increases in rates of microzooplankton herbivory would also significantly increase microbial foodweb carbon flow and nutrient remineralization in this environment. She will estimate growth rates of numerically dominant taxa within the microzooplankton assemblage during the chemostat experiments by microscopy. She will estimate specific ingestion rates of numerically dominant individual microzooplankton taxa within the chemostat experiments by measuring uptake of fluorescently labeled algae. Microzooplankton grazing rates will also be estimated using

disappearance rates of fluorescently labeled algae. These complementary measurements of microzooplankton growth and grazing should give reasonable estimates of top-down control by microzooplankton on mixed assemblages of phytoplankton in our experiments.

Saito Group (msaito@whoi.edu**)**

Participants: Mak Saito (PI),
Abigail Noble (grad student),
Erin Bertrand (grad student).

1. Research goals:

The biogeochemistry of cobalt, cadmium and zinc will be investigated in the Ross Sea using voltammetric speciation techniques applied to water column samples and bottle incubations. This proposed study would add a Co, Cd, and Zn trace metal chemistry component to two NSF-OPP funded R/V Palmer cruises to the Ross Sea in 2006 (PIs: DiTullio, Hutchins, Smith, Sedwick, Dunbar, Tortell, Bowie). One of the primary objectives of the DiTullio cruises is to study the interactions between iron limitation and carbon dioxide utilization/limitation using novel trace metal clean chemostats incubation methods. Because carbon acquisition by marine phytoplankton is dependent on the Zn, Cd, or Co metalloenzyme, carbonic anhydrase, adding Co, Cd, and Zn trace metal analyses to the research program should be highly complementary to the cruise objectives. Total dissolved concentrations and chemical speciation analyses will be measured on field samples to ascertain the ambient geochemical conditions as well as on bottle incubation samples to measure chemical transformations and drawdown of these micronutrients. Cobalt analyses will be carried out using cathodic stripping voltammetry with dioxime electroactive ligands. Cadmium and zinc will be analyzed using anodic stripping voltammetry. DNA samples will also be collected from the Ross Sea and amplified for genes homologous to ferric reductases. This enzyme has been shown to be involved in the acquisition of ferric iron, including organically chelated forms, but molecular analyses of marine phytoplankton ferric reductases have barely begun. Sequences of these genes will be amplified using polymerase chain reaction and degenerate primers, and clone libraries will screened for sequences of interest. Diversity of this functional gene will be assessed by DNA sequencing and alignment of sequences. This study of ferric reductases will also be highly complementary to the cruise research activities focused on iron limitation, and is an important first step towards future analysis of ferric reductase gene expression in environmental samples. Samples will also be taken for vitamin B_{12} uptake and inorganic cobalt uptake in seawater and measurements of B_{12} in seawater. Finally, frozen samples will be taken for measurements of methyl mercury and total mercury in seawater.

2. Wire time needs: We will be bringing our trace metal rosette sampler and 10L Go-Flo bottles. The sampler has 11 bottle 2.5L bottles and will be attached to the Kevlar non-conducting line. It is triggered independently by timer. The rosette is significantly faster than sampling by 10L Go-Flo bottle (we will be bringing two bottles), but miscasts do occur due to the problems with this new technology. We

can typically accomplish a 1500m cast in ~3 hours and a shallow 400m cast in 1hr. Ideally, we would really like to obtain 6 profiles on the transit down to the Ross Sea across the polar front. If CTD casts are occurring we would like to do a long cast before and short 300m cast after the CTD to try to obtain a high resolution (22pt) profile. Once in the Ross Sea we would like to obtain 5-7 profiles per transect (total 10-14) utilizing 2-4 hr of wire time. I should discuss with Pete about coordinating sampling too.

3. CTD water requirements: We may need 1L from 4-6 depths when conducting uptake studies, and we may need 4L from 4 surface depths for RNA and DNA. However, some of this sampling may be shifted to trace metal clean surface pumps and our Go-Flo samplers depending on time of day of sampling (uptake studies are 24h).
4. Trace Metal Clean water for incubations: 30-50L per incubation set, at 6 times during the cruise. Do we have a mixing carboy?
5. Liquid nitrogen needs: We are experimenting with RNA-later solutions so our LN requirements may be reduced from initial expectations. However, to be conservative I will need 15-25L every 10 days to recharge a shipper/flash freezing dewar.
6. I'm hoping to do a number of +Co, +Fe, +Cd +Zn style experiments, if you want them to overlap with chemostat experiments let me know. I've got 1L and 2L bottles, maybe I should try to do 2L at these sites to allow pigs?

Sedwick Group B-267-N (psedwick@bbsr.edu)

Participants are:
Peter Sedwick (PI, BBSR)
Chris Marsay (research associate, BBSR)
Maeve Lohan (post-doc, U Plymouth, UK)
Ana Aguilar-Islas (grad student, UCSC)
Juliette Tria (grad student, U Tasmania)

The principal aim of our work is to define the spatiotemporal distribution, sources and sinks of biologically-available Fe in surface waters of our study region.

Our primary work will involve measurements of the concentrations and speciation of the lithogenic trace metals Fe, Mn and Al in samples from the water-column (collected in kevlar casts), surface water (collected from towfish pumped supply), and, if possible, snow/sea-ice/glacial-ice samples.

At selected stations, we will also use the towfish pumped supply to collect large volumes of uncontaminated whole seawater for the shipboard incubation experiments.

Wire time needs. I'd say we would do a maximum of 1 kevlar cast per day, requiring ~3 hours. As much as possible, we would like to fly the towfish between stations. If all goes smoothly, it should take 30-60 minutes to deploy and recover the fish.

I would definitely like to know who needs TM clean water and how much. At present, I am only aware of Hutchins (~500L? for chemostats) and Tortell/DiTullio (volume? for semi-continuous batch experiments and Fe/UV experiments) who will need water at 2 stations.

We plan to provide clean seawater for 2 big chemostat experiments but that will depend on several factors that we can discuss later. If anyone needs clean water at the chemostat stations please let me know.

Smith Group (wos@vims.edu**)**

Participants:
Walker Smith, PI
Jill Peloquin, Post-doc
Sasha Tozzi (grad student)
Carol Pollard (grad student)
Jennifer Dreyer (grad student)

Our group will contribute to the overall group efforts of investigating the interactive effects of iron, CO2 and light on phytoplankton growth by conducting routine productivity and photosynthesis/irradiance measurements. As a subset of this, we will conduct size fractionations of each (GFF and 20 um), focusing on Phaeocystis dominated stations, to assess the 14C uptake of colonies vs. solitary forms. Similar fractionations for various biomass estimates will also be conducted.

We also will be conducting more detailed experiments during the cruise. For example, we will conduct a growth rate-temperature response experiment by measuring the 14-chlorophyll pigment labeling rate on samples incubated at a variety of temperatures under constant light. Long-term temperature responses will also be assessed by growing assemblages at a variety of temperatures (over weeks) and measuring their growth rates (and perturbations from that temperature). We will also measure the single-cell responses using a PAM-fluorometer linked with a microscope (Peloquin). Bulk PAM measurements will also be made routinely, as will rapid light curves using the PAM.

Finally, we will be conducting the last year of IVARS during this cruise. That means simply that we will want to occupy a series of stations in the southern Ross Sea (our "survey") in which we collect biomass determinations in the upper 150 m (Chl, HPLC pigments, BSi, POC/PON, microscopy, nutrients, POP, productivity, P/E measurements) and CTD to the bottom. We also will deploy moorings with in situ fluorometers, microcat, current meters, sediment traps, nitrate sensor) at two locations for the duration of the cruise. They will be recovered during NBP06-01A.

Tortell Group (tortell@ubc.edu)

Participants:
Phillipe Tortel (PI)
Celine Gueguen (postdoc)
Chris Payne (grad student)

Our main objective is to examine the physiological mechanisms of inorganic carbon acquisition in diatom and Phaeocystis-dominated communities, and to study the effects of natural and experimental CO_2 gradients on phytoplankton species composition. This work is motivated by our previous field studies of C acquisition in natural phytoplankton communities, including preliminary evidence that the diatom / Phaeocystis ratio may be sensitive to ambient CO_2 concentrations. Our research goals will be accomplished by combining sampling of in-situ assemblages and CO_2 – controlled incubations. At many of the major CTD stations, we will collect phytoplankton assemblages to determine: a) the relative proportions of HCO_3^- and CO_2 taken up by cells; the kinetic properties (K_m and V_{max}) of the Ci uptake system; 3) the in vivo activity of extracellular carbonic anhydrase; and the in vitro activities of RubisCO, total Carbonic anhydrase, and PEP-carboxylase (these will be determined in the lab on frozen material). We also hope to work with Dunbar's group on the interpretation of stable carbon isotope signals. Ultimately, we are looking for a relationship between ambient CO_2 concentrations and the activity of cellular carbon concentrating mechanisms. Preliminary results from the NE Pacific suggests that such a relationship can indeed be observed in field samples. CO_2 effects will also be examined directly using semi-continuous incubations, and we are particularly interested in potential CO_2-dependent species shifts. These incubations will run in parallel to the chemostat experiments of Hutchins' group.

In addition to the studies outlined above, we shall also undertake underway measurements of various dissolved gases using our membrane inlet mass spectrometer (MIMS). This system is capable of measuring (with high accuracy and precision) the concentrations of CO_2, O_2, N_2, Ar, and DMS, with a frequency of more than once per minute. Each of these gases provides a unique tracer for some biological or physical process, and we are particularly interested to look at the fine-scale structure of gas distributions in the polynya. The DMS work will be in support of DiTullio's program.