**Research protocol, study sites and science party list for the upcoming cruise on New Horizon, October 14th-October 27th, 2009**
**Sailing from Mazatlan, returning to San Diego**

*"Collaborative research: Non-local bacterial transport of nitrate within sediments underlying Oxygen Deficient Zones: A new twist in the N cycle"*

## I. Introduction

The focus of the proposed work is sedimentary denitrification, the largest loss term for the biologically available nitrogen in the marine nitrogen cycle. Previous studies in oxygen deficient zones of the oceans (along Peru-Chile margin and in the California Borderland basins) have showed that, in contrast to a widely accepted paradigm, a large proportion of nitrate in the water column may be lost via biological, active (or non-local) transport into the sediments and subsequent denitrification, rather than a passive diffusive flux driven by the sedimentary consumption of nitrate.

We will investigate the link between the non-local nitrate transport and regional nitrogen losses in oxygen deficient regions in the Eastern Subtropical North Pacific. The bacteria *Thioploca* and *Beggiatoa,* found in this region, are known to accumulate up to 500 mM of nitrate intracellularly. They transport nitrate into the sediments, up to depths of tens of centimeters where nitrate is used for $H_2S$ oxidation. This non-local nitrate transport (within laminated sediments) clearly 'breaks' the rules of diagenetic sequence. We hypothesize that non-local transport of nitrate may be a significant but previously overlooked sink in the regional and global N budgets of oxygen deficient zones.

## II. General objective

The overall goals of the proposed work are to:

1) examine the geochemical framework favorable for non-local transport of nitrate;
2) determine the relative contribution of this process to net regional losses of fixed N;
3) identify and quantify the effects of this phenomenon on the isotopic composition ($\delta^{15}N$) of nitrate in the overlying water column; and,
4) investigate the possible coupling of nitrate transport to other reactions that reduce nitrate, such as anammox and iron reduction.

## III Research protocol

### *1) Sample collection*

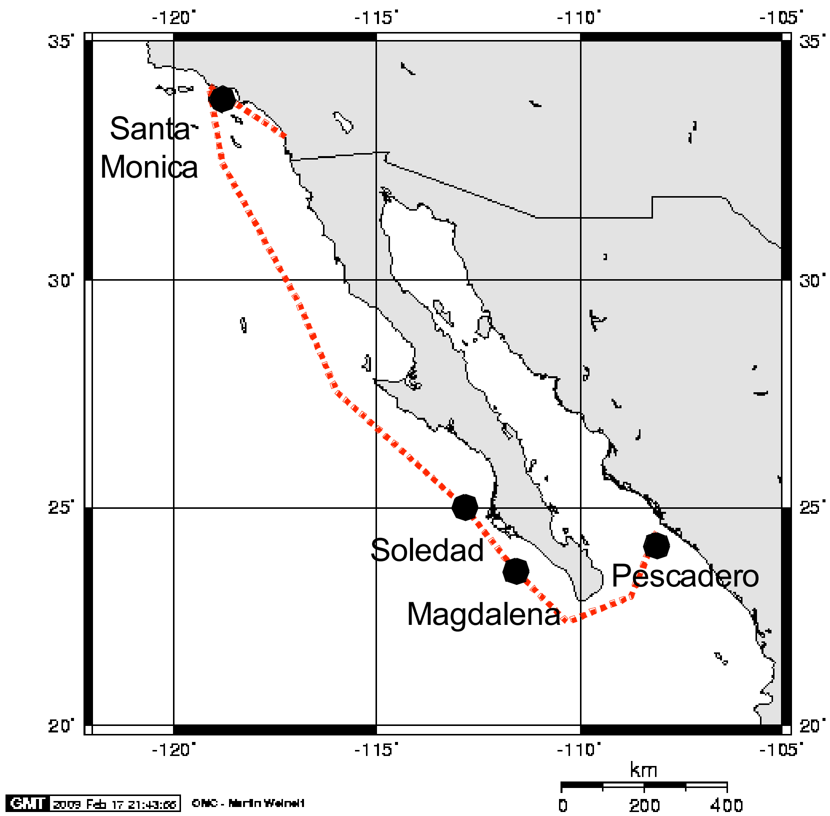
Four stations will be occupied in the Mexican waters (see the map in the SRE application): Pescadero, Soledad Basin and Magdalena margin. If time permits, a forth station, to the north of Magdalena margin may be occupied as well. At all of the stations, we will collect water column samples using CTD rosette and sediments, using multicore and gravity core. Pore water will be extracted from the sediments, using whole core squeezer and rhizomes. Water column and pore water samples will be analyzed as described below.

### *2) Incubations*

In order to link sedimentary and water column processes and quantify the exchange between the two environments, fluxes across sediment-water interface will be determined by incubations, both *in situ* using benthic landers and on deck, using sediment cores. During the course of incubations, water samples will be collected and analyzed as described below. Sediments will be also incubated to determine the anammox activity.

### *3) Analysis*

Dissolved gases ($N_2$/Ar ratios) will be measured on board during the cruise using a Membrane Inlet Mass Spectrometer (MIMS). Dissolved $N_2O$ will be analyzed onboard as well. Other analysis will be conducted back in our university laboratories. Those will include measurements of $TCO_2$, pH, dissolved $Fe^{2+}$ and $Mn^{2+}$, $[H_2S]$ and sulfate to establish a geochemical context the nitrate transport, as well as $[NH_4]$, $[NO_3]$ and $[NO_2]$, $N_2/Ar$ ratios and $[N_2]$ and isotopic analyses - $\delta^{15}N$ and $\delta^{18}O$ of pore water nitrate, $\delta^{15}N$ of ammonium. In order to investigate the coupling between nitrogen and sulfur cycle, sulfur isotopes will measured on sulfate, sulfide and other, intermediate sulfur species. We will also apply common molecular methods to determine the composition of relict bacterial and protist community structure at all study sites.




| | Lat, N | Long, W | Nombre |
|---|---|---|---|
| Sitio 4 | 33.835 | 119.028 | Cuenca Santa Monica |
| Sitio 3 | 25.080 | 112.950 | Cuenca Soledad |
| Sitio 2 | 23.420 | 111.230 | Margen Magdalena |
| Sitio 2a | 23.160 | 110.950 | Margen Magdalena |
| Sitio 1 | 24.170 | 108.120 | Cuenca Pescadero |

**Application of results**

The shuttling/advecting of nitrate into the sediments may play a unique role in the material exchange between the water column and the sediments: by bypassing the diffusion limitations, this process may account for a significant proportion of total benthic denitrification, depending on the final fate of the transported nitrate. It is ultimately the $C_{org}$ oxidation that drives this process by generating reducing potential in form of $H_2S$ (or reduced metals), but non-diffusive transport linking nitrate consumption directly to $H_2S$ oxidation apparently increases total denitrification, and increasing the sensitivity of the sedimentary denitrification feedback to the $C_{org}$ rain from the overlying water. Non-local transport of nitrate into the sediments may have important consequences for S, metal and C cycles.

The study of mechanisms and rates of N consumption in suboxic sediments also addresses important practical issues. The naturally occurring oxygen deficient zones (ODZ) in the oceans (Black Sea, Baltic Sea and Gulf of Mexico excluded) presently occupy ~ 764,000 $km^2$ (defining suboxia by $[O_2] < 10$ μM. This accounts for 5% of the continental slope area (depth 0.2-1 km) and < 0.3 % of the global sea floor. However, natural climate change is capable of greatly changing the volume of suboxia. Anthropogenic eutrophication of coastal waters and global warming may also result in the growth of areas of suboxia, such as those presently occurring in the Gulf of Mexico , Baltic Sea and elsewhere. While the suboxic condition itself is clearly an environmental cost of human activities, the associated N consumption by denitrifiers and other organisms could be a beneficial side-effect (removing N so as to reduce algal growth), providing a shunt to the eutrophication cycle.

People--
Maria Prokopenko--USC--co I--nitrogen cycling studies
Will Berelson--USC--co I--nitrogen and carbon cycling studies
Oscar Gonzalez-Yajimovich--UABC Ensenada--sedimentology
Tim Riedel--USC--graduate student
John Fleming--USC--grad student
Laurie Chong--USC--grad student
Amy Townsend-Small--UCI--nitrogen cycling studies
Lynda Cutter-USC--trace metal studies
Jim McManus--OSU--trace metal studies
Sean Loyd--USC--sulfide studies
Person #1-Ensenada--student with Oscar
Person#2-Ensenada--student with Oscar
Marissa Hirst--UCD--anoxic eukaryotes
Loreto De Brabandere--USDenmark--nitrogen cycling
Heidi Jensen --USDenmark--nitrogen cycling
Ann Dekas--Caltech--nitrogen cycling
Lt Palacois--Mexican Navy, observer