R/V *Tioga* Cruise #746
Cruise Report

April 25th – 27th, 2014

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National Science Foundation Ocean Acidification Grant # OCE- 1316040 (PIs: Lawson, Maas and Tarrant) *"Ocean Acidification: Seasonal and ontogenetic effects of acidification on pteropods in the Gulf of Maine"*

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2. Acknowledgements

The success of this cruise would not have been possible without the dedication, skill and professionalism of Captain Ken Houtler and mate Ian Hanley of the *R/V Tioga*. We are grateful for the efforts of Ann Tarrant who assisted with transferring the water collected on the first day to Woods Hole Oceanographic Institution's Environmental Systems Laboratory and Mike Lowe who helped with demobilization. This cruise was supported by a grant from the National Science Foundation OCE-1316040 (Lawson, Maas, and Tarrant).

3. Background

As a result of increases in atmospheric carbon dioxide (CO_2), the ocean is taking up extra CO_2 and becoming more acidic, in a process referred to as ocean acidification (OA). Certain coastal regions, such as the upwelling system along the U.S. West Coast, are more susceptible to the effects of ocean acidification than others, because their waters are episodically or seasonally naturally higher in CO_2 concentration and lower in pH and saturation of aragonite (a calcium carbonate mineral). In such OA 'hot-spots,' continued anthropogenic perturbations to the carbonate chemistry will quickly push the system towards a more corrosive (aragonite under-saturated, $\Omega A < 1$) environment that many calcium carbonate shell-forming organisms may not tolerate. Coastal acidification in the Gulf of Maine (GoME) has generally not been considered to be a pressing concern, but new data (Wang et al. 2013) suggest that in the deep waters of the GoME low seawater pH may already cause aragonite saturation states (ΩA) to be close to a chemical and ecological threshold (i.e. $\Omega A = 1$).

In conjunction with this spatial variability in CO_2 is a documented seasonal variability in the surface carbonate chemistry of the GoME. The spring season in the GoM corresponds to the timing of the lowest levels of documented CO_2 (~250 ppm) in contrast to highs of ~550 ppm evident in early winter (Fig. 3; Irish et al. 2010; Vandemark et al. 2011). Unlike other regions, where natural CO_2 fluctuations are influenced primarily via upwelling events and eutrophication, this large variability in CO_2 is driven primarily by riverine outputs, seasonal cycles in primary production, and sea-surface temperature changes (Previdi et al. 2009; Salisbury et al. 2009). This pattern continues to be monitored at UNH by a NOAA CO_2 buoy moored at 43° N 70° W in the GoME: http://www.pmel.noaa.gov/co2/story/GOM

The implications of this spatial and temporal variability in carbonate chemistry are that organisms in this system may already be exposed to conditions of undersaturation. This cruise part of a series designed to determine the physiological response of the local population of thecosome pteropod, *Limacina retroversa* to seasonal variability in environmental CO_2. These aragonite-shelled individuals are found throughout the year in the GoME and by exploring what conditions they experience *in situ*, and then brining animals back to the lab for metabolic, gene-expression and calcification studies, we can determine whether there are seasonal sensitivities to CO_2 exposure on time-scales relevant to acclimation responses. Paired with this primary objective is the opportunity to continue an ongoing seasonal time series designed to more fully understand the vertical and spatial variability in the carbonate chemistry of the GoME.

4. Cruise Objectives

The central goal of this cruise was to document the abundance and vertical distribution of the pteropod species *Limacina retroversa*, to capture live individuals for experimentation, and to sample the carbonate chemistry profile of two sites in the GoME. The long-term goal of this research is to understand variability in the carbonate chemistry of the Gulf of Maine and to understand how these changes in the natural environment impact the local planktonic calcifiers, specifically the thecosome pteropods. The specific goals are to:

1. Quantify seasonal patterns in the abundance of the pteropod *Limacina retroversa* and its vertical distribution relative to concurrent measurements of water column chemical properties, testing the hypothesis that this species is absent in the acidic waters of the near-bottom nepheloid layer.
2. Test whether there are seasonal patterns of gene expression, shell quality and metabolic rate linked to seasonal exposure.
3. Determine how experimentally enhanced levels of CO_2 influence the gene expression, shell quality and metabolic rate of *Limacina retroversa* that are exposed for a period of 1-14 days in the laboratory and explore whether these responses are mediated by seasonal exposure.
4. Maintain the seasonal carbonate chemistry sampling time-series associated with Tioga cruises 668, 700, 715, and 725. This dataset will allow us to test the hypotheses that deep waters of the GoME are already seasonally under-saturated with respect to aragonite saturation state.

The specific goals of this particular cruise were to:

1. Catch *L. retroversa* with a vertically stratified net system to quantify their size class, abundance and vertical distribution in the context of the carbonate chemistry and season.
2. Collect *L. retroversa* to preserve in ethanol (70%) for shell studies and in RNAlater for gene expression studies to explore the seasonal response to CO_2.
3. Collect surface water and *L. retroversa* for live animal laboratory experiments.
4. Measure the carbonate chemistry of the water column at multiple sites in the Gulf of Maine, targeting the sites which were sampled during Tioga cruises 668 (May 2013), 700 (August 2013), and 715 (October 2013) to provide a seasonal contrast in the measurements.
5. Measure the carbonate chemistry in the nepheloid layer to provide a seasonal contrast in the measurements.

5. Survey Design

On Thursday April 24th the R/V Tioga was packed at WHOI and left port around 17:00 to transit to Provincetown. The scientific crew followed by car and stayed at the Cape View Motel. On Friday April 25th the boat left port at ~7:00 am and traveled to Murray Basin, an offshoot of western Wilkinson Basin (standard station 2/consecutive station 1 in Fig. 1; ca. 260 m) where we deployed the CTD twice, the MOCNESS twice (both due to mechanical difficulties) and collected water with the little pump for experiments. We then transited to standard station 3/consecutive station 2 (Fig 1; ca 135) where we deployed one Reeve net. We were met at the dock by Ann Tarrant and transferred water to a 55 gallon barrel and to multiple trash cans aboard a truck that she returned to WHOI. The next day we left at 7:30 am and traveled to standard station 3/consecutive station 2 where we deployed the CTD and the acoustics package. At this time we pumped new water for animal handling and then conducted 6 Reeve nets. Animal capture was non-ideal (many animals but most of a very small size), so we began a semi-systematic search for higher numbers of medium to large adults. At consecutive station 3 we deployed 2 Reeve nets, at station 4 we deployed 1 net, and at station 5 we deployed 1 (very successful) Reeve net then returned to port for the evening. Animals from the Reeve nets were transferred from the refrigerator into two coolers and to Peter's car. These were driven to WHOI where they were placed in the cold room. The morning of the 27th a reduced crew of five scientists left the dock at 7:15 am and traveled to consecutive station 6. We put in our first Reeve net at 7:30 and continued traversing the area for the rest of the day, completing 11 Reeve tows by 16:00. Occasionally the boat repositioned as there was a front in the area across which the catches of medium to large animals diminished. Upon return to port at ~16:30 some gear was offloaded and the scientists returned to WHOI. The animals remained in the refrigerator aboard ship as the R/V Tioga began the transit back to WHOI. The boat was met at the dock at ~21:30 WHOI and the jars of animals and the final trash cans with water and animals was craned off and transported to ESL via truck. The rest of the gear was unloaded and moved into the staging room and Lawson lab for cleanup and stowage. Full information about casts and stations can be found in the Event Log (Appendix 1).

6. Cruise Narrative

Day 1: Friday April 25th

Friday morning the scientific crew met the R/V *Tioga* at the Provincetown dock and left port at 7:00. Waves were quite high this day (maxing at ~10ft) creating an atmosphere that diminished scientific crew productivity. As a consequence event logging was patchy for the following activities. We reached the first station (consecutive 2, station 1 in Figure 1) at ~9:40. This site was chosen as it has been sampled previously for carbonate chemistry multiple times in 2013 and for zooplankton distribution in 2011 and 2013. Our CTD sampling had a reduced number of bottles than previously (12 vs. 16 on the rosette), the oxygen and transmissometer were absent from the unit and there were two bottles that did not fire. During CTD water sampling the MOCNESS was prepped and deployed. There were communications errors with the unit which caused it to be returned to deck and serviced. While the MOC was being worked on, two small pumps were deployed to obtain water from ~30 m depth for animal culture. Six 40 gallon trash cans were filled in ~45 minutes. After pumping the MOCNESS was redeployed. Despite continued communications difficulties, a full cast of the water column was completed. The top two nets were particularly full of phytoplankton, somewhat slowing processing. Samples of *Meganyciphanes norvegica* (??) from net 4 (n=2) and net 0 (n=1) were put in crovials and preserved in liquid nitrogen during the sampling. While the MOC samples were being processed the CTD was redeployed to provide higher resolution of the bottom water and to fill in the dataset for the two bottles which misfired. Once the CTD was onboard the ship began to transit to standard station 3/consecutive station 2 (Fig 1). Here a Reeve net was deployed to collect pteropods. There were many small individuals (~200) and 15 medium size adults. We then left station and steamed towards Provincetown which we reached by ~19:00. At the dock we were met by Ann Tarrant and transferred water off the boat via the small pump. Ann, Ali and Maja drove the water and the 3 jars of animals back to WHOI where she and Ali Thabet transferred the water into the tank for filtration.

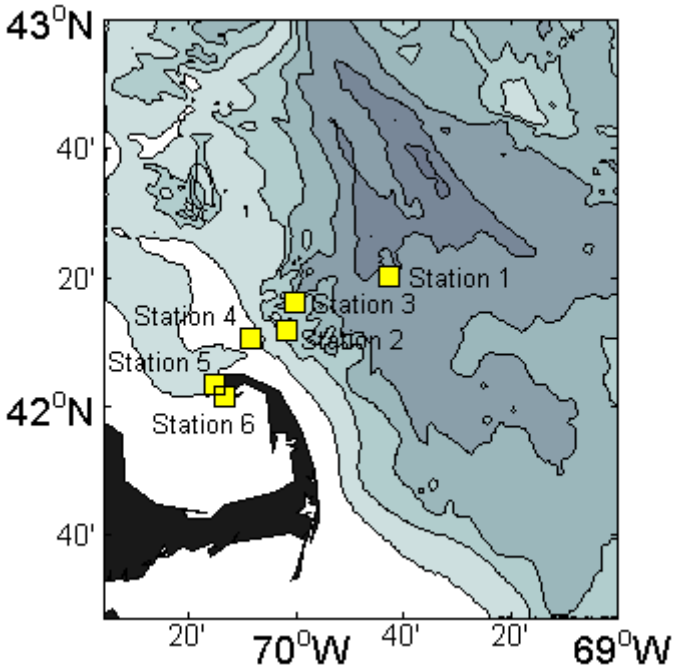


Figure1 – Gulf of Maine Map. We sampled with the CTD pup and MOCNESS at Station 1 (standard station 2) on Day 1 (April 25th). At Station 2 (standard station 3) on the 25th we did a Reeve net and on the 26th we did a CTD, pumping, acoustics and a Reeve tow. We began trying other stations on the 26th, Reeve sampling at stations 3-5 in search of larger animals. On Day 3 (April 27th) we did all of our Reeve sampling near Station 6.

Day 2: Saturday April 26th
We left Provincetown at ~7:30, and set out for standard station 3 (consecutive 2). The sea state was still a bit rough, but improved throughout the day. On the way out ethanol was replaced in the MOC samples which were most in need. We reached the station and the CTD was deployed at 8:50. During CTD sampling the acoustics package and the pump were put in the water and we filled the 1 L sampling jars which were kept in the refrigerator. Simultaneously two trash cans were filled. When the pump was done we retrieved the acoustics packages and started Reeve netting. Reeve nets were conducted at a number of depths to try to maximize catch and minimize wire time. Although there were a high number of animals, most were too small for the experiment. After six tows we had only ~56 large animals in jars, and 25 in RNAlater. At this point we began to change stations to attempt to find higher densities of larger animals. These stations were at the Edge of Wilkinson' Basin (~10 useable animals), the SE corner of Stellwagen Bank (~15 useable animals) and within Cape Cod Bay. The final tow was the most successful resulting in ~200 small but useable animals. We left station at 17:12 and started to transit back to Provincetown. We returned to port at ~17:40. All the pteropods were packed into two coolers and taken by Peter Wiebe back to Woods Hole. The remaining scientific crew went to Stop and Shop to buy more jars (wide-mouth quart ball jars).

Day 3: Sunday April 27th
We left Provincetown at 7:15 am and traveled to consecutive station 6. At this site was a large aggregation of cetaceans, seabirds, and large fish. The Reeve netting at this station (11 in total) yielded relatively large animals in high numbers during most tows. The number of animals was great enough that any tiny individuals, which are usually kept for culturing experiments, were transferred to one of the trashcans for transport so that all jars could be dedicated to large experimental animals. At one point we moved across a front (obvious via temperature, flocculent phytoplankton in the net and a change in zooplankton community) during one tow which reduced the catch number. After that we made a point of staying on the other side of the front. We left the station at 16:05 and got to port at 16:33 where we unloaded much of the carry-on gear. The science crew went for dinner and then back to WHOI, while the Tioga made the transit carrying all of the full pteropod jars. Back at WHOI the crew was met by Ali and Mike Lowe who joined up to unload the truck full of gear. Everyone then meet the boat at 21:30 and the heavy gear was offloaded via crane and transported to the staging room (nets) or ESL (two trashcans and all of the pteropods in jars.

7. Equipment Configuration

7.1. *Deck configuration*

The collapsible plastic crate with 4 garbage cans was strapped down to the starboard side of the back deck. The MOCNESS was tied down on the starboard aft, the CTD was positioned mid-ship. The VPR was forward of the wheelhouse, although it ended up not being used. The Reeve net was stowed on the port aft rail. We used the same cable for all deployments. When the back deck space was needed for other deployments the CTD was moved to the forward port side of the deck. A standard refrigerator was strapped down forward of the winch and housed a number of glass jars for specimen collection. There was a table with a built in sink bolted down at the forward port portion of the back deck that was used for chemistry sampling and net processing. The ethanol for preservation was strapped down furthest aft of this table to allow for easy jar filling. Under the sink were the coolers full of jars for live animals and a tote containing the MOCNESS sample processing gear. During CTD processing this space also contained sample bottle totes. On the second day a tarp was placed above the sampling station to prevent rain from into the work area.

7.2. *Lab configuration*

The main lab aft counter housed the laptop which was used for event logging and VPR processing. The starboard counter had MOCNESS sampling supplies, chemistry sampling equipment, and the BioSonics

acoustic setup. On the floor was more MOCNESS backup/sampling equipment which continued into midship. The rest of the backup supplies, foul weather gear and personal belongings were stored in the port bunk space. The MOCNESS computer unit was housed in the wheelhouse. A small dry shipper with liquid nitrogen was strapped down to the starboard side of the stairs heading down into the lab.

8. Hydrography: CTD

8.1.1. Introduction

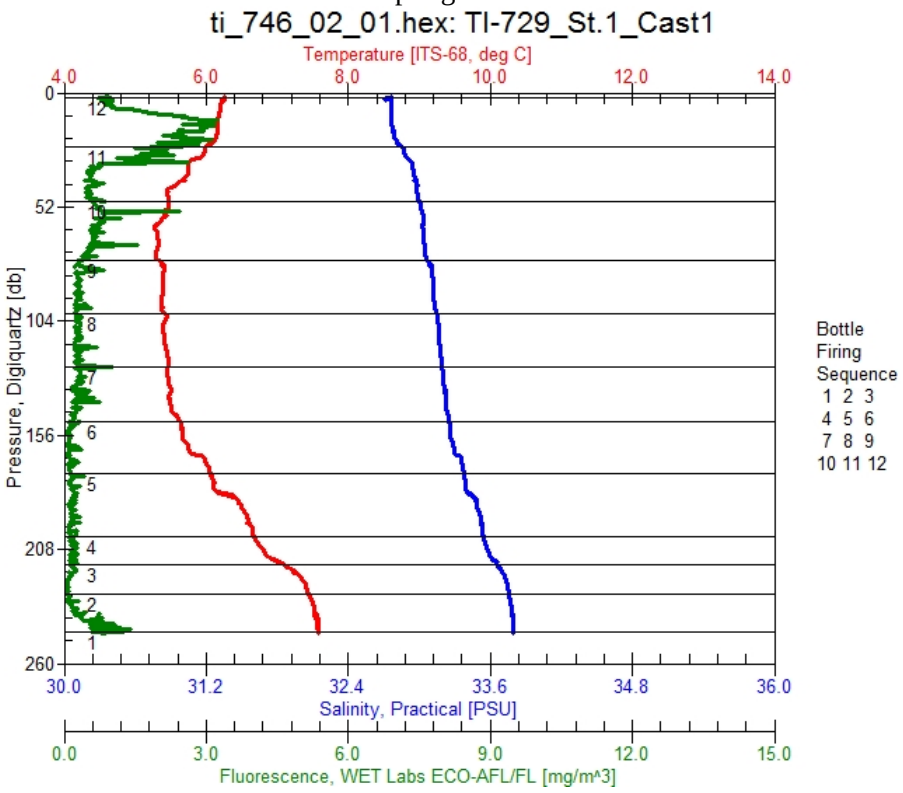
CTD rosette casts were used to get both a profile of the water column and to allow Niskin bottle sampling for the carbonate chemistry analysis. These CTD measurements of environmental conditions will provide key correlates of the distribution and abundance of pteropods with hydrographic features. Depths were chosen to characterize the bottom nepheloid layer (BNL) and then to continue at pre-designated intervals throughout the rest of the water column.

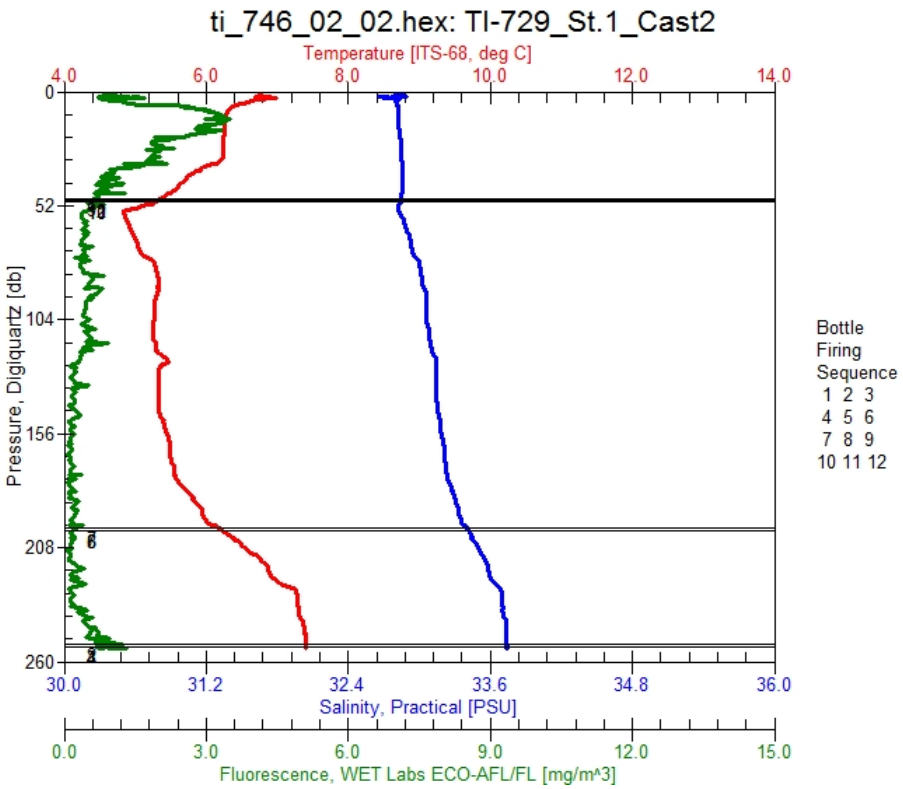
8.1.2. Methods

The R/V *Tioga* CTD rosette had a12 bottle rosette with 3-L Niskins, and a SBE3/SBR4 sensor set. Niskin bottle sampling provided water for the carbonate chemistry analysis. Depths for bottle sampling generally are chosen to characterize the bottom nepheloid layer (BNL) and then to continue at pre-designated intervals throughout the rest of the water column. The typical protocol for the CTD is to sample upper 100m at 10 m intervals, 100-200m at 20 m intervals, and less frequently below. At the near-shore station (i.e. Standard Station #3) the water depth necessitated only the firing of 10 bottles, following the pattern established on previous cruises (Appendix 2).

8.1.3. Preliminary Results

Due to miscommunication with ops, both the O_2 sensor and the transmissometer were absent from the CTD. The reduced bottle number and some miss-firing of the CT during the first cast necessitated a second cast at consecutive station 1 to achieve full sampling of the water column.

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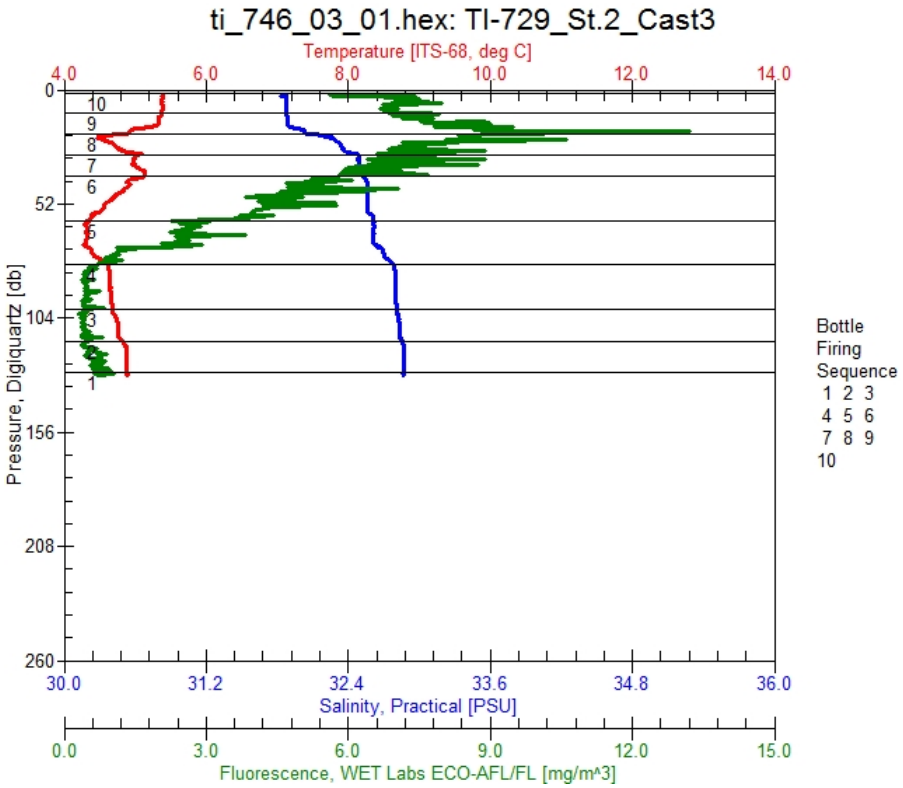
7 8 9
10 11 12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33.6
34.8
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Salinity, Practical [PSU]
0.0
3.0
6.0
9.0
12.0
15.0
Fluorescence, WET Labs ECO-AFL/FL [mg/m^3]
Bottle
Firing
Sequence
1 2 3
4 5 6
7 8 9
10

9. Chemistry

9.1. *Introduction*

Carbonate chemistry samples are to be analyzed by Dr. Zhaohui Aleck Wang's group from the Department of Marine Chemistry and Geochemistry at WHOI. Aboard ship we collected discrete bottle samples of seawater for later measurement of carbonate chemistry parameters. Measuring these parameters allows us to calculate pH, the carbonate compensation depth and the calcium carbonate saturation state, three important variables that may influence the formation of aragonite shells by pteropods.

9.2. *Discrete Measurements of Dissolved Inorganic Carbon and Total Alkalinity*

9.2.1. Methods

Discrete dissolved inorganic carbon (DIC) and total alkalinity (TA) samples were collected from the surface to near-bottom. Depths were chosen to follow previous sampling patterns (See CTD Methods). DIC and TA samples were collected in 250mL Pyrex borosilicate glass bottles after being filtered with a 0.45um in-line capsule filter. Each bottle was rinsed three times, filled completely, and then the sample was overflowed by another one and one half bottle volume. Air head space of about one percent of the bottle volume (~3 ml) was left in each sample bottle to allow room for expansion. Each sample was then poisoned with 100uL of saturated mercuric chloride, capped with an Apiezon-L greased stopper, thoroughly mixed, and then tied with a rubber band over the glass stopper. Duplicate samples were collected at random depths of selected stations to evaluate the precision of the measurements. These samples will be measured for DIC and TA back in the Wang Lab at WHOI.

9.3. *Discrete Nutrient Measurements*

9.3.1. Methods

Nutrient samples were collected in acid cleaned Kimble 20mL plastic bottles. Before the cruise, the bottles were soaked in 10% hydrochloric acid for four hours, rinsed three times with de-ionized water, and then dried in the oven at 50°C for 48 hours. During collection, the sample was filtered with a 0.22um Pall capsule filter. The bottle was rinsed three times with the sample and then filled. Collected samples were put into the plug-in freezer aboard ship immediately upon collection. When the R/V *Tioga* reached WHOI these samples were taken to the WHOI Nutrient Analytical Facility for analyses. Concentrations of ammonium, nitrate plus nitrite, nitrite, orthophosphate, and silicate will be determined by a Lachat Instruments QuickChem 8000 four-channel continuous flow injection system, using standard colorimetric methods approved by U.S. Environmental Protection Agency.

10. Zooplankton Sampling

10.1. *MOCNESS*

10.1.1. Introduction

A standard 1/4-m^2 Multiple Opening/Closing Net and Environmental Sensing System (MOCNESS; Wiebe et al., 1985) was used to collect zooplankton to determine the vertical distribution and abundance of the thecosomatous pteropods.

10.1.2. Methods

The MOCNESS was equipped with nine 150-um mesh nets (nets 0-8). The underwater unit used was #169; temperature probe was #535 and conductivity probe was #120. Unlike previous deployments the strobe system was not employed during this cruise. The MOCNESS was deployed from the aft winch and A-frame. Upon recovery the nets were all hosed down with seawater and the cod-ends were

sequentially removed, placed in buckets, and transferred to the forward sink and table. Net contents were preserved in 70% ethanol. All samples had the ethanol replaced with fresh 70% ethanol ~24 hours after sampling.

Oblique casts with the MOCNESS were made to ca. 10 m off bottom (based on the Knudsen echosounder on the ship's depth estimate) with a ship speed nominally of 1.5-2.5 kts. Maintaining this speed on the Tioga can be difficult. Consistent with previous cruises, sampling for the top 5 nets was intended to be taken at 150-100, 100-75, 75-50, 50-25, and 25-0 m. For the bottom two nets the goal was to choose depths to cover the lower water column and ensure that the lowest net occurred exclusively in the nepheloid layer (see Appendix 3- for sampling sheets). Cast depth has been to 10m off bottom. Typically there has been a region of fairly constant or gradually changing transmission at the bottom of the BNL, with a region of steeply changing transmission at shallower depths, ending at the top of the BNL where transmission levels off. The protocol has been to sample with net 1 from the max depth to the top of the region of constant/gradually changing transmission and then with net 2 from there to the top of the region of steeply changing transmission (i.e. to the top of the BNL). On this cruise there was no transmissometer and so we had to guess at the BNL depth (Appendix 3).

During the MOCNESS tow we repeatedly lost communication with the underwater unit. The system was recovered but on deck it worked fine with both the test cable and through the winch and wire. The system was therefore redeployed but again repeatedly lost communication. Each time we were eventually able to re-initiate communication but the gaps in connection sometimes occurred just as a net was being tripped, leading to some uncertainty in exactly what depths were being sampled by which net. Peter Wiebe was not on board the day of the MOCNESS tow but the following day joined the cruise and did some sleuthing to figure out the most likely sequence of events and depths sampled (Appendix 3). The final conclusion was that the depth of the nets were approximately 0-50, 50-100, a failed net, 100-150, 150-200, 200-225, 225-235. Ultimately the most likely explanation for the communication issues is some weak link somewhere in the termination/wire/sliprings chain.

10.1.3. Animal sampling

Animals were sampled from the MOCNESS nets for gene expression analysis. *Meganyciphanes norvegica* individuals were handpicked from net 4 (n=2) and net 0 (n=1), put in cryovials and then preserved in liquid nitrogen.

10.2. *Reeve Net*

10.2.1. Introduction

The objective of Reeve net sampling was to gently collect live specimens to be sampled for physiological and genetic analyses. These trawls were short in duration and aimed to maximize pteropod catch.

10.2.2. Methods and Approach

A 1-m diameter Reeve net with a 150-um mesh net was deployed via the A-frame. The book-clamp to attach the net was borrowed from Carin Asjian's lab. Ship speed during tows was ~1-1.5 knots. The depth and duration of deployment varied widely. We occasionally focused on deep (60-80) or shallow (0-30) when searching for animals in the deep stations, but had our greatest success with slow up-down tow between 15 m and the bottom (~40 m) at the shallow stations (See Table 1 and Appendix 4 for details).

On the bench installed on the back deck, the cod end was promptly divided among a number of buckets. Since pteropods tend to sink, the bottom buckets were examined first. The contents were swirled and the pteropods sucked up of the center of the bottom using a plastic pipette. These were then concentrated, again taking advantage of the pteropod tendency to sink, and then sorted to create two size classes – the

"large" (big enough for respiration experiments) and the "small". Animals were put into seawater filled pre-cleaned glass jars that had been kept in the fridge before and after filling to maintain ambient temperature (see Pump section for details on filling the jars).

10.2.3. Preliminary Findings

The Reeve nets at the deeper stations had only a few *Limacina retroversa*. Larger, whole ones (from R6) were put into RNAlater. Most individuals collected on Day 1 and Day 2 were dedicated to culturing/egg laying experiments. Most of the large animals (~1960 individuals) were caught on Day 3 at a very shallow station close to port (station 6). At the end of the day we had filled > 50 jars with 40 individuals each. We also collected a trash barrel of very small animals for culturing experiments (> 10,000). 2 vials of 5 individuals each were also put into liquid nitrogen on Day 3 (from R14). Details of the tow depths, duration and success is in Table 1.

Table 1: Success of Reeve net capture.

			# Animals caught		Sampling depth	Duration
St.	Reeve	Notes	Xpt.	Culture	(m)	(min)
1	R1		15	lots	30-60	
2	R2		16	lots	15-30	
2	R3	25 RNAlater	5		30-60	30
2	R4		5	~200	30-45 x2	28
2	R5		5	~200	60-85	
2	R6		25	few	60-80	34
2	R7		0		0-30	17
3	R8	Edge of Wilkinson's	~10		25-60	28
3	R9	Edge of Wilkinson's	~15		50-100	
4	R10	SE Corner of Stellwagen Bank	120	lots	0-50	26
5	R11	Cape Cod Bay	210	Lots	0-45	
6	R12		200	Trash can	0-35	17
6	R13		200	Trash can	0-35	17
6	R14	2 cryovials x5 Limacina	200	Trash can	12-35	20
6	R15		200	Trash can	12-35	34
6	R16		200	Trash can	16-52	30
6	R17		280	Trash can	14-40	24
6	R18		160	Trash can	16-39	19
6	R19	1 cryovial Clione	160	Trash can	16-45	23
6	R20		120	Trash can	0-35	18
6	R21		120	Trash can	16-45	20
6	R22		120	Trash can	14-40	18

10.2.4. Troubleshooting

The spring phytoplankton bloom was in full force during the cruise and the Reeve net was coming up clogged with algae. Cleaning the net before redeployment took a significant amount of time and effort. In an attempt to expedite the process the net was put overboard without the cod end and dragged in the propeller wash. Over time this resulted in tears in the net and a kink in the wire, so we returned to the procedure of washing the net thoroughly with the hose. This was improved significantly with the addition of an adjustable rate nozzle on the end of the hose.

11. Pump

11.1. *"Little Giant" Pump*

11.1.1. Introduction

The objective of the "Little Giant" pump was to retrieve water from depth for animal culture. This goal is to have large amounts of water of the appropriate salinity, DIC/TA and temperature to replicate the conditions the animals experience in situ. To achieve this, six "pteropod ptransporter" garbage cans were brought onboard and held in a plastic cage or strapped to the port rail.

11.1.2. Methods and Approach

First two separate 100 feet of heavy duty hose was lowered into the water attached to the winch line (~30 m depth). At intervals a carabineer had been attached to the hose with electrical tape and then affixed to the hose to keep the hose close to the line. Once the hoses were at depth a pump was attached to each hose. The pumps were "Little Giant" sub-pumps with a watertight spliced extension cord. Another hose was attached to the outflow of these units. The pumps were lowered until just below the surface using a safety line, lashed down and then turned on. The outflow hoses were strapped to the trash barrels using clamps and filtered through a 63 micron sieve.

When we got to port we transferred water a truck that had been driven to Provincetown by Ann Tarrant. The truck pre-positioned on the dock and then we dropped the pump directly into the trash bins and extended the two 100 ft hoses via the outflow to the truck. We filled up a 55 gallon drums and four trash bins on the truck.

The following day the pump was used again, filling two trash cans. While this was occurring we filled the 1-L glass jars with filtered water. These jars were placed in the refrigerator to maintain as close to ambient seawater temperature as possible for pteropod transportation.

12. Acoustics

12.1. *Introduction*

In an effort to improve our ability to localize the pteropods in the water column in terms of vertical position, as well as to help identify regions of high pteropods abundance, on this cruise we brought along the WHOI Biology Department Biosonics DT-X echosounder. The system has two single-beam transducers, operating at 120 and 200 kHz, from a single deck transceiver. The system is very user-friendly and required minimal extra effort. The transducers could be deployed using the port-side boom that Jay Sisson developed for rocky Geyer, which Andone Lavery has used previously with the Edgetech system, but for this cruise given the limited success from the previous cruise and the inclement weather we decided to keep things simple and just hung the transducers bolted to a plate over the starboard side, tied off to the rail. The transceiver and a control laptop were housed in the main lab.

Relatively little scattering was evident over much of the water column, and nothing indicative of pteropod patches or layers. No effort was made therefore to target net sampling at particular acoustic features. The transducers were deployed at a time that didn't interfere with the progress of other, more project-relevant, operations were underway. Given the sometimes rough conditions, the lack of dedicated personnel to monitor the acoustic data, and the limited indication of any pteropod-like scattering (or scattering of any kind), the transducers were only used on this cruise for a limited time.

13. Cruise Participants

Science Party

1	Amy Maas	Chief Scientist	WHOI	Biology
2	Gareth Lawson	Scientist	WHOI	Biology
3	Alex Bergan	Graduate Student	WHOI	Biology
4	Taylor Crawford	research Assistant	WHOI	Biology
5	Peter Wiebe	Scientist	WHOI	Biology
6	Sophie Chu	Graduate Student	WHOI	Chemistry
7	Leocadio Blanco Bercial	research Faculty	UConn/WHOI	Biology
8	Ali Thabet	Guest Student	Assiut University/WHOI	Biology
9	Maja Edenius	Graduate Student	WHOI	Biology

Officers and Crew

1	Ken Houtler	Captain
2	Ian Hanley	Mate

14. References

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