Presented at the DLSU Research Congress 2016 De La Salle University, Manila, Philippines March 7-9, 2016



# $\begin{array}{c} Comparison \ of \ abundance \ and \ diversity \ of \ invertebrates \ in \ sediments \\ from \ CO_2 \ and \ non-exposed \ reefs \end{array}$

Karen Camille Perez and Ma. Carmen Ablan-Lagman <sup>1</sup>Biology Department, COS, De La Salle University \*Corresponding Author: ksren\_perez@dlsu.edu.ph

**ABSTRACT:** The continuous acidification of seawater contributes to the decline of coral reefs and to a decrease in abundance and diversity of associated invertebrates. Acidification can occur with increase in dissolved  $CO_2$  in the water. On reefs,  $CO_2$  venting sites are areas that display the possible effects of ocean acidification because of the increased dissolved CO<sub>2</sub> at these sites. In this study, the possible effect of ocean acidification on the abundance and diversity of marine invertebrates in sediments were compared between samples from water fractions collected from from Autonomous Reef Monitoring Structures (ARMS). The ARMS units were retrieved from shallow depths ranging from 13.8 m to 16.7 m. A total of five sites were represented in the area of Anilao, Batnagas but only four were used for the analysis of data due to the lack of collected sediments. The Acacia site represents the  $CO_2$  venting site reef while the Tingloy area serves as the marine protected site. Arthur's Reef and Twin Rocks sites serve as the reefs where there is constant anthropogenic involvement. The invertebrates were identified to phyla based on morphology and the number individuals per sample were counted for each site. Diversity and abundance were compared based o frequency and the Shannon-Weiner Species Diversity Index. No significant differences in the abundance and diversity of marine invertebrates were observed among the CO<sub>2</sub> venting reef site and the adjacent reefs.

#### Keyword: acidification, abundance and diversity, CO2 venting site, invertebrates

### 1. INTRODUCTION

One of the pressing matters that the ocean is now facing is the continuous increase in water acidity. This is mainly caused by the increase of carbon dioxide ( $CO_2$ ) released into the atmosphere. 93%  $CO_2$  comes from human activities (Sabine et al., 2004). The other 7% would come from naturally occurring  $CO_2$  venting sites in the ocean.

The continuous increase of  $CO_2$  in the ocean is a threat to the marine biodiversity, particularly to the crustaceans and corals. An increase in the acidity of water lowers the pH, which may affect the process of shell formation of most crustaceans. Corals may also suffer, as an increase in  $CO_2$  accelerates coral bleaching (Milman, 2014). Ocean acidification affects the survival, calcification, growth, development and abundance of marine organisms (Kroeker et al., 2013).

There are many possible causes of ocean acidification. One direct cause of the rise in acidity of seawater is through anthropogenic activities. CO<sub>2</sub> emissions from anthropogenic origin is one of the many causes of long-term changes in the pH level of the ocean, which may be concerning to the population of calcifying organisms that are located in coastal habitats (Duarte et al., 2013). Another cause is from the naturally released carbon dioxide from the environment. Continuous increase of atmospheric CO<sub>2</sub> may cause a reduction in pH, which will greatly affect the marine shelled molluscs or calcifying organisms (Gazeau et al., 2013). Effects would not only be seen in calcified invertberates but it will also affect the natural cycle, which include habitat structure for benthic organisms, water purification, and food source of other marine and terrestrial organisms (Gazeau et al., 2013).



Ocean acidification greatly affects the abundance of marine biodiversity. Smaller organisms may decrease in number dramatically and habitats may gradually decline. This can lead to a chain reaction that would affect not only the marine environment but the whole ecosystem dynamics.

Samples filtered through the 500 uM sieve from the four sites were analyzed and compared. Only the ARMS deployed in shallow depths were studied.

## 2. METHODOLOGY

### 2.1 Collection of Data



Figure 1. The sites were the ARMS units were retrieved.

The Autonomous Reef Monitoring Structure (ARMS) were recently recovered in Anilao, Batangas after being subjected underwater for three years. The retrieval of the ARMS was done during the month of May 2015.

Four out of the five sites (BAO, ART, TWI, BAA) were used to collect samples (Figure 1). The Kuala Reserve (KOA) site was disregarded due to the lack of samples collected. The CO<sub>2</sub>



Figure 2. The retrieval container (left) and disassembly tub (right).

venting site was discovered around the reef in Acacia (BAO). A marine protected area, Tingloy (BAA) was also considered.

The retrieval of ARMS unit was done using a recovery container. Once retrieved, the ARMS unit was transferred into the disassembly tub filled with sea water and has an air pump for oxygenation (Figure 2). After disassembling the ARMS unit, the water from the tub was filtered through 100 uM and 500 uM sieves. Only the samples sieved through the 500 uM were analyzed due to the nature and size of the invertebrates. Filtered samples were preserved using methanol contained in small bottles.

### 2.2 Analysis of Data

Three bottles of samples were analyzed for each site. A portion of each were examined under a dissecting microscope. This was done in order to check the abundance of invertebrates. Each individual was quantified and grouped together according to its phylum. A paired sample t-test was used to assess the biodiversity between reef sites. A statistical software called StatPlus was used to perform this test. To check for the diversity of each site, the Shannon-Weiner Species Diversity Index was used, which is defined as:

$$\mathbf{H'} = -\sum p_i ln p_i \tag{Eq. 1}$$

where:

 $p_i$  = proportion of each species in the sample

The results were tabulated and were compared to see the difference between each sites.

### 3. RESULTS AND DISCUSSION

The organisms found under the microscope from each of the sites were organized according to phyla. Table 1 shows the total number of individuals per phyla on each site.

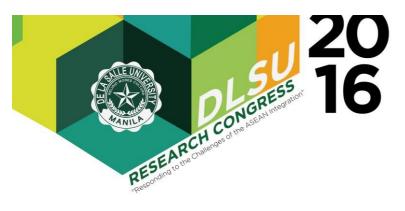


Table 1. Number of individuals per phyla observed in each site

	Sites			
Phyla	BAO	BAA	TWI	ART
Arthropoda	3	<b>5</b>	4	8
Chordata	1	-	1	3
Mollusca	13	8	20	18
Echinodermata	1	1	-	10
Porifera	8	9	2	2
Cnidaria	7	7	10	7
Algae	2	2	-	1
Foraminefera	2	-	6	2
Worm	2	11	4	7
Total	39	43	47	58

Table 2. Comparison of abundance of organisms		
among sites using Paired-sample T-test.		

Compare	Means				
	Descriptive Statistics				
VAR	Sample	Mean	St.	Var	
	Size		Dev.		
BAO	9	4.33	4.12	17.0	
BAA	9	4.78	4.18	17.44	
Test	0.35				
Statistic					
P-value	0.59				
= 0.05					
BAO	9	4.33	4.12	17.0	
ART	9	6.44	5.36	28.7	
Test	1.43				
Statistic					
P-value	0.59				
= 0.05					
BAO	9	4.33	4.12	17.0	
TWI	9	5.22	6.39	40.9	
Test	0.70				
Statistic					
P-value		0.8	3		
= 0.05					

Table 3. A comparison of the diversity of each sites using the Shannon-Wiener Diversity Index.

Sites	Н	Evenness
BAO	1.88	0.86
BAA	1.77	0.91S
TWI	1.61	0.83
ART	1.89	0.86

Table 4. The average pH level across each site based on water analysis.

Sites	Average pH
BAO	7.9
BAA	8.1
TWI	8.1
ART	8.2

A portion from the sample bottles of each site was closely observed under the microscope. Table 1 shows the phyla present among the sites and the quantity of the individuals was also noted. It can be seen that the site BAO, which was the site where the  $CO_2$  vent was located, contains the smallest number of organisms. The remaining sites were considerably larger in number.

Although there was an increase in number of individuals from the non-CO<sub>2</sub> venting reef sites compared to the CO<sub>2</sub> venting reef site, they were not significant enough to indicate a relation between the acidification of the water to the decline in number of organisms living there, as shown in the Pearson Correlation Coefficient value in Table 2. This may be due to the fact that the pH level across the reef sites remain to be basic (Table 4). The pH level in the CO<sub>2</sub> venting reef site has not reached the threshold level wherein it would have an immediate or observable effect on its inhabitants, particularly in the decline in numbers of invertebrates. The pH value from each reef site may be in relation to the season in which the ARMS units was retrieved. High pH level in the water, turning the seawater basic, is experienced during the summer due to the change in water temperature and an increase in gross photosynthesis as well as a decrease in respiration from the surrounding (Kerrison et al., 2011). A previous study of Garrard et al. (2014) compares the invertebrate community between two distinct pH zone: pH 8.1 and pH 7.8, almost similar to the pH recorded from the ARMS sites. The study showed that many of the invertebrates were able to thrive even in acidic waters in nearshore habitat (Garrard et al., 2014).

The Shannon-Wiener Diversity Index is a common tool to check for the diversity of a community. The higher the H value is, the more diverse a certain community. (Lee, 2011). Among the sites studied, ART has the highest H value, making it the most diverse among the four. BAA, which is the marine protected area, obtained the



lowest diversity. This may be due to the number of individuals living in each reef site, which is the species richness or due to number of individuals of different species found across the reef sites, which is the species evenness (Lee, 2011). There were more individual of different species found in ART than in BAA, which contributed to the low H value depicting a low species diversity. Although there was no significant difference in the diversity found among the reef sites.

### 4. CONLCUSION

No significant difference in the abundance and diversity of individuals was found between each sites. An increase in the area studied and an increase in the number of samples may help assess more the effect of acidification for the study. Sampling during the wet season (June throughout November) to see if there are differences in the  $CO_2$  concentration in the water.

#### 5. ACKNOWLEDGEMENT

The ARMS project was a collaboration between international organizations, which are the Smithsonian Institution (SI), National Oceanic and Atmospheric Administration (NOAA), Sand Diego State University (SDSU), and the Indonesian Biodiversity Research Centre (IBRC) and local universities and organizations, which are the De La Salle University (DLSU), University of the Philippines Marine Science Institute (UP-MSI), Philippine National Museum of Natural History (PNMNH), and National Fisheries Research and Development Institute (NFRDI). This paper makes use of samples provided by SI and water analysis data given by NOAA, who were funded by the NOAA Coral Reef Conservation Program. We would like to thank Glenn Oyong for letting us use the Molecular Biology Laboratory.

### 6. REFERENCES

Duarte, C. M., Hendriks, I. E., Moore, T. S., Olsen, Y. S., Steckbauer, A., Ramajo, L., Carstensen, J., Trotter, J. A., & McCulloch, M. (2013). Is ocean acidification an open-ocean syndrome? Understanding anthropogenic impacts on seawater pH. *Estuaries and Coasts*, 36(2), 221-236. doi: 10.1007/s12237-013-9594-3

- Garrard, S. L., Gambi, M. C., Scipione, M. B., Patti, F. P., Lorenti, M., Zupo, V., Paterson, D. M., & Buia, M. C. (2014). Indirect effects may buffer negative responses of seagrass invertebrate communities to ocean acidifiaction. Journal of Experimental Marine Biology and Ecology, 461, 31-38. doi: 10.1016/j.jembe.2014.07.011
- Gazeau, F., Parker, L. M., Corneau, S., Gattuso,
  J., O'Connor, W. A., Martin, S., Portner, H., &
  Ross, P. M. (2013). Impacts of ocean
  acidification on marine shelled molluscs.
  Marine Biology, 160(8), 2207-2245. doi:
  10.1007/s00227-013-2219-3
- Kerrison, P., Hall-Spencer, J. M., Suggett, D. J., Hepburn, L. J., & Steinke, M. (2011). Assessment of pH variability at a coastal CO<sub>2</sub> vent for ocean acidification studies [Electronic version]. Estuarine, Coastal and Shelf Science, 94(2), 129-137.
- Kroeker, K. J., Korda, R. L., Crim, R., Hendriks, I. E., Ramajo, L., Singh, G. S., Duarte, C. M., & Gattuso, J. (2013). Impacts of ocean acidification on marine organismes: Quantifying sensitivities and ineraction with warmning. *Global Change Biology*, 19(6), 1884-1896. doi: 10.1111/gcb.12179
- Lee, J. (2011). Shannon index Its development, implication, and effectiveness. Retrieved October 20, 2015 from https://math.dartmouth.edu/~m20f11/LeeProj. docx
- Milman, O. (2014, April 13). Entire marine food chain at risk from rising CO2 levels in water. Retrieved October 20, 2015 from http://www.theguardian.com/environment/201 4/apr/14/entire-marine-food-chain-at-riskfrom-rising-co2-levels-in-water.
- Sabine, C. L., Feely, R. A., Gruber, N., Key, R. M., Lee, K., Bullister, J. L., Wanninkhof, R., Wong, C. S., Wallace, D. R., Tilbrook, B., Millero, F. J., Peng, T., Kozyr, A., Ono, T., & Rios, A. F. (2004). The ocean sink for anthropogenic CO<sub>2</sub> [Electronic version]. Science, 305(5682), 367– 371.



Presented at the DLSU Research Congress 2016 De La Salle University, Manila, Philippines March 7-9, 2016