

## **Diatom and dinoflagellate abundance and ecology in waters off the Hawaiian Coast**

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## **Non-Technical Abstract**

Diatoms and dinoflagellates are two groups of microscopic algae known as phytoplankton. Phytoplankton get their energy from the sun through photosynthesis, the same way land plants do. This means that they “breathe in” carbon dioxide (CO<sub>2</sub>), much like land plants do, and store it in their bodies. When they die, their bodies sink to the bottom of the ocean where they eventually become rocks. This process of CO<sub>2</sub> being taken from the air and taken to the ocean floor is known as the biological pump. As you may know, CO<sub>2</sub> is a green house gas that plays an instrumental role in climate change, so tracking the way it leaves the atmosphere is important. My study is designed to further our understanding of what types of diatoms and dinoflagellates live in the waters around Hawaii and how their numbers differ between locations. I collected and counted hundreds of phytoplankton along with taking samples of the concentrations of key nutrients that allow phytoplankton to grow. Unexpectedly, the number of phytoplankton at a given location did not match the amount of nutrients. This is likely because there are other types of phytoplankton, such as bacteria, who use the same resources as diatoms and dinoflagellates. It is also possible that some dinoflagellates are getting their energy from eating food instead of from the sun, because there are several species that have the ability to do both. Future research is needed to test both of these possibilities.

## **Abstract**

Diatoms and dinoflagellates are two groups of phytoplankton important in carbon sequestration, which affects climate change, and harmful algal blooms (HABs). Understanding their community ecology is important to our ability to make predictions about these issues. This study uses phytoplankton abundance and biomass at seven locations around the Hawaiian Islands to determine if diatoms and dinoflagellates correlate with nutrient abundances. The results suggest that there is little relation between  $[PO_4]$ ,  $[NO_3]$ ,  $[NO_2]$ ,  $[NH_4]$  and  $[Si(OH)_4]$  and phytoplankton biomass. However, diatom and dinoflagellate biomass does not correlate strongly with chlorophyll either, suggesting that other groups of phytoplankton are a major constituent of the surface primary producers. The presence of the cyanobacterium *Trichodesmium* was noted at all sites, supporting this hypothesis. Another possibility is that some diatoms and dinoflagellates are not behaving as autotrophs, explaining why they do not correlate with chlorophyll, and that the actual photosynthesizers are more closely related to nutrients.

## **Introduction**

Diatoms are phytoplankton that dominate planktonic communities when certain nutrients are present (Egge and Aksens 1992). Therefore, they play a major role in the natural processes phytoplankton take part in. One such process is the biological pump, which is driven by photosynthesis occurring in the surface ocean's phytoplankton. Photosynthesis helps remove carbon from the atmosphere. The carbon remains stored in the phytoplankton's tissues until they are consumed or die. Once they die, they sink out of the surface ocean, and towards the bottom, carrying the carbon with them. The transfer of carbon from surface waters to depth by means of organically derived matter is an important part of the biological pump (Longhurst and Harrison

1989). The process of removing carbon from the atmosphere and storing it in sediments is known as carbon sequestration, and has important implications for climate change.

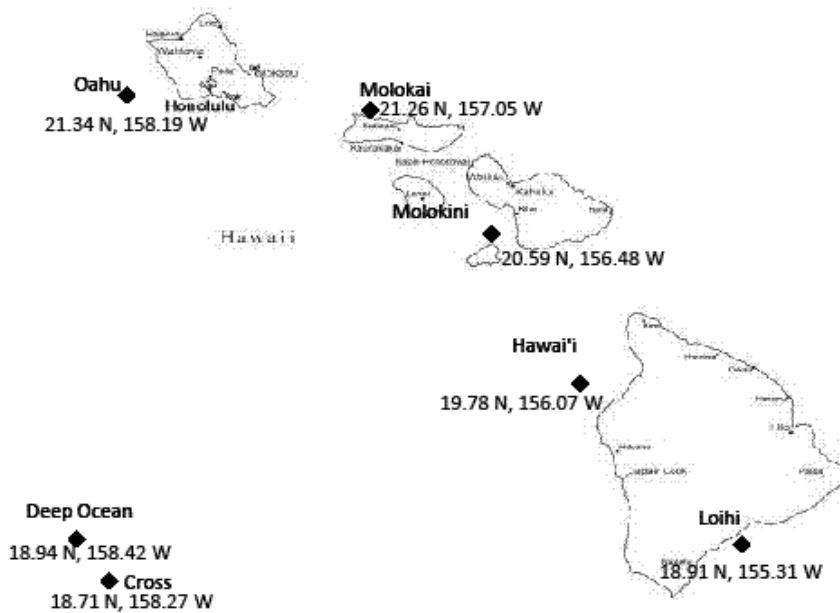
Of course, different groups of phytoplankton are more likely to sink to their mass and the shapes of specialized appendages used to help them float (Eppley et. al, 1967). Diatoms sink easily because they are negatively buoyant due to their silica shells (Horner 2002). However, as previously noted, diatoms require certain amounts of silica and nitrate to grow, often different than those of other plankton. For instance, Egge and Aksens 1992 study compared treatments of phytoplankton with different levels of silica in the water, to determine its impact upon species composition. They found that diatoms dominated over other groups only at higher silica concentrations. Other studies have shown a similar relationship between diatoms and other nutrients, such as iron and nitrate (Takeda 1998, Eppley and Renger 2008). Nutrient levels are high in coastal environments, where upwelling and run-off from land boost nitrogen and micronutrients. Open ocean environments are characterized by low nutrient levels. However, features, such as islands, exhibit characteristics of both coastal and open ocean environments. The Hawaiian Islands are a prime example, due to their location in the middle of the oligotrophic Pacific, and their high population and agriculture, implying high nutrients. Thus, it is difficult to predict the community composition of phytoplankton in the waters around Hawaii, along with the effects they have on carbon cycling. Consequently, it is important to know the abundance and species of diatoms in the waters around Hawaii to form an accurate understanding of the biological pump there and in similar cases.

Diatoms and dinoflagellates are also known to produce Harmful Algal Blooms (HABs) by releasing toxins into the water. HABs cause both health and economic concerns due to toxins accumulating in fish and shellfish that humans then consume. In Hawaii, the economic cost of

HABs is estimated at about \$3 million due to ciguatera poisoning in otherwise marketable fish (Hoagland et al. 2002). At least one dinoflagellate found in Hawaii, *Cambierdiscus toxicus*, is linked with ciguatera poisoning (Shimizu et al, 1982). Therefore, it is necessary to know which potentially harmful phytoplankton species are present in coastal waters and where they are, if we want to know how to minimize illness and economic loss due to diatoms and dinoflagellates. Consequently, the purpose of this research is to determine the diatom and dinoflagellate abundance along with the species present in near-shore waters off Hawaii.

## **Methods**

Data was collected at thirteen waypoints around the Hawaiian Islands from aboard the *R/V Thomas G. Thompson* from 27 December 2010 through 4 January 2011 at the stations shown in Figure 1. At each site, a plankton net tow was performed using a net with 25  $\mu\text{m}$  mesh. The net was lowered to the chlorophyll maximum, determined using fluorometer data attached to a conductivity-temperature-depth (CTD), and towed vertically to the surface. Each net sample was washed into a jar and preserved with 2% formalin solution. A niskin rosette attached to a CTD was also used at each station. A niskin was closed at the chlorophyll maximum and one liter collected and preserved with 15 mL of 2% formalin solution. All samples were processed on land following the cruise.



**Figure 1: The seven locations sampled labeled by name, longitude and latitude. Map source: <http://www.mapresources.com/catalogsearch/result/?dir=desc&cat=40&order=cost&q=Hawaii>**

To perform cell counts from the net tows, a Palmer-Maloney slide was utilized using the technique outlined in Horner 2002. The area of one 0.1 mL slide was analyzed by cataloguing the genus' present and the number of each. At least 200 cells were counted from each slide and no more than 500.

Biomass was calculated using basic geometric shapes as suggested in Menden-Deuer and Lessard (2000). Two measurements were taken for each cell, length and girth. Length was defined as the line from the most distant points on the longer side (if applicable) and girth as the furthest distance on the shorter side (Table 1). Carbon was calculated using the Carbon to Volume ratio described in Menden-Deuer and Lessard 200 using the formula:

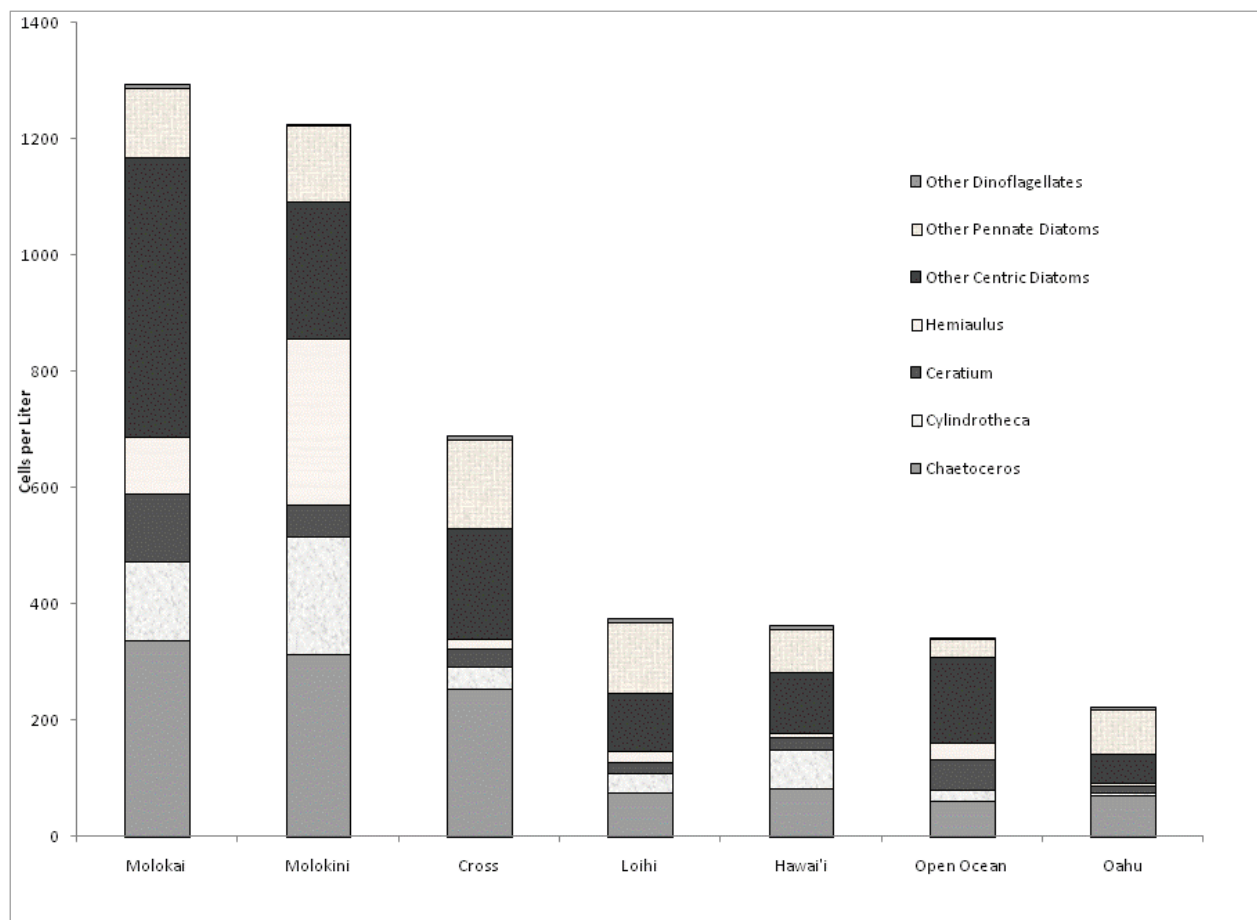
$$\log \text{pgC} = \log a + b * \log V$$

where V is the volume and a and b are constants specific to diatoms and dinoflagellates.

Chlorophyll was collected at depths ranging from the surface to below the chlorophyll maximum at each station. At each depth, three 0.136L samples were taken and processed using the spectrophotometric method as outlined in Lorenzen 1967. The results were then averaged and used to calibrate the CTD fluorometer results. Nutrient samples were collected at each station and analyzed by the University of Washington Marine Chemistry Lab for  $[\text{PO}_4]$ ,  $[\text{NO}_3]$ ,  $[\text{NO}_2]$ ,  $[\text{NH}_4]$  and  $[\text{Si}(\text{OH})_4]$ .

## Results

The number of cells varied from 222 to 1295 cells per liter of seawater (Figure 2). The highest cell abundances were at Molokai and Molokini, while the lowest were at the open ocean station, and Oahu. The diatom genus *Chaetoceros* made up the greatest abundance at all sites, followed by *Cylindrotheca* at all but two. The diatom *Hemiaulus* and the dinoflagellate *Ceratium* were also heavily present (Figure 2). At all sites centric diatoms made up more than half the population. The Shannon-Weiner diversity index was calculated for all sites, with H values ranging from 1.78 to 2.22 (Shannon 1948). The equability ranged from 0.71 at Molokai to 0.80 at both Loihi and Open Ocean. This range is on the higher end of species richness and evenness of distribution considering that the highest possible equability is one.



**Figure 2: The abundance of cells at each station for the four most common genus' of diatoms and dinoflagellates and for other types.**

The total volume of cells and consequent mass of carbon stored in the phytoplankton followed the same trend as the overall cell abundance, with the exception that station Oahu had greater biomass than Open Ocean due to larger cells. Carbon ranged over an order of magnitude from 0.0507  $\mu\text{g}$  at Open Ocean to 0.781  $\mu\text{g}$  at Molokini.

Nutrient data from the chlorophyll maximum did not correlate with cell abundance for  $[\text{PO}_4]$ ,  $[\text{NO}_3]$ ,  $[\text{NO}_2]$ ,  $[\text{NH}_4]$  or  $[\text{Si}(\text{OH})_4]$ . The  $R^2$  values ranged from 0.0023 to 0.21, meaning none of the nutrients sampled contributed to more than a quarter of the variation in biomass. There were also spikes in  $[\text{Si}(\text{OH})_4]$  and  $[\text{NO}_3]$  at sites with lower abundance. The chlorophyll

readings did not correlate either, with an  $R^2$  value of 0.33 when compared to the biomass (Figure 3).

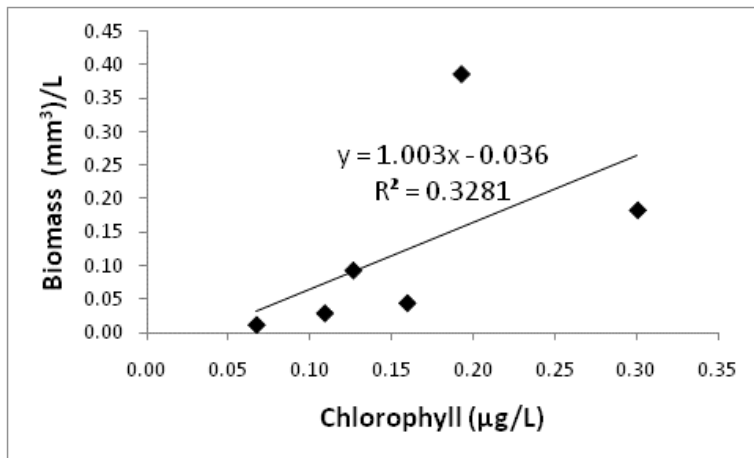


Figure 3: Chlorophyll and biomass comparison with treadmill.

## Discussion

The general trend of stations nearer to shore having higher cell abundance than those removed from shore seems to be true of this data. One notable exception is the station Oahu. This sample was the only one not preserved immediately after being collected – instead it was left alive overnight – and is potentially lacking in phytoplankton cells due to grazing by predators that were observed in the sample. Oddly enough, this site had the highest concentration of  $[\text{Si}(\text{OH})_4]$ , a nutrient essential to diatoms. However, the chlorophyll measured at the chlorophyll maximum is only  $0.11 \mu\text{g/L}$ , the second lowest value, after the Open Ocean station, which suggests there is actually a low concentration of phytoplankton at this site. It is possible then, that a micronutrient or one of the other nutrients sampled is limiting growth of phytoplankton at this location. One such nutrient that has previously been found to limit diatom growth in the tropical Pacific is iron, which was not tested for (Hutchins and Bruland, 1998). Future research might include testing for iron.

Another notable occurrence is that the stations at Loihi and Hawaii have similar biomass and cell abundance. These two sites are also situated near the coast of the island Hawai'i. Despite the apparent correlation in diatom and dinoflagellate biomass, chlorophyll concentrations at the Hawaii station are close to those at Molokini, much higher than the readings at Loihi. Thus it appears that the primary production at this station is aided by organisms other than diatoms and dinoflagellates. One possible such organism is the cyanobacterium *Trichodesmium*, which was present in all the samples, although the amount was never quantified. Based on qualitative observations, the sites differed in *Trichodesmium* abundance. Previous studies have observed *Trichodesmium* in the waters around Hawaii, where it can account for at least 18% of the chlorophyll a found in surface waters (Letelier and Karl 1996). *Trichodesmium* is most noted for its ability to fix nitrogen, so future research might include quantifying the bacteria's presence and comparing it to concentrations of different nitrogen compounds such as ammonia, nitrate and nitrite (Carpenter and Price, 1976).

Since total biomass did not appear to relate to any of the nutrients or chlorophyll, specific genus' were analyzed for correlations with different nutrients. One such genus was *Hemiaulus* because it requires less silica than most diatoms (Benitez-Nelson et al. 2007). Despite predictions of a negative correlation with the concentration of silica, it showed no trend at all.

Of the dinoflagellates identified, none are known for causing harmful algal blooms (HABs), determining that, at least at the time of this study, none were occurring at these locations. Despite this, further studies that repeat samples over time are needed to track blooms that can be potentially harmful and further research is needed. *Pseudo-nitzschia*, a diatom known to secrete the neurotoxin domoic acid and cause HABs was present at some sites (Marchetti et al., 2008).

## Conclusions

Based on the results of this study, diatoms and dinoflagellates appear to be less abundant at locations where there is no nearby land. However, there is a great deal of variations between the waters around specific islands, allowing for the possibility of many mechanisms controlling phytoplankton populations around Hawaii. It does not appear that nutrient concentrations (at least for those sampled) have a major impact on diatoms and dinoflagellates. Two possible reasons for this exist. First, it could be that other phytoplankton dominate the waters near Hawaii. This is supported by the presence of *Trichodesmium* in the samples. An alternate hypothesis, is that the diatoms and dinoflagellates identified are not all living as autotrophs. Dinoflagellates in particular are known to be mixotrophic or heterotrophic. Both these hypothesis are consistent with chlorophyll levels that do not match diatom and dinoflagellate trends. In sum, there is further research needed to determine the cause of variation between waters around the islands of Hawaii.

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## Appendix 1

The cells of each genus and total cells counted as well as the calculated cells per liter based of volume measurements.

	Cross	Hawaii	Loihi	Molokai	Molokini	Oahu	Open Ocean
<b>Diatoms</b>							
<b>Centric</b>							
<i>Bacteriastrum</i>	0	0	0	1	1	0	0
<i>Chaetoceros</i>	138	80	43	80	119	67	35
<i>Coscinodiscus</i>	0	0	1	0	0	0	3
<i>Guinardia</i>	0	0	8	5	66	0	24
<i>Odontella</i>	0	0	1	0	0	0	0
unidentified centric	103	102	48	108	23	46	60
<b>Pennate</b>							
<i>Cylindrotheca</i>	21	64	20	32	76	4	11
<i>Diatyliosolen</i>	43	18	37	11	23	33	0
<i>Ditylum</i>	0	0	1	0	2	0	2
<i>Hemiaulus</i>	10	5	10	23	108	4	16
<i>Navicula</i>	3	8	1	0	2	1	3
<i>Proboscia</i>	11	6	4	6	14	1	9
<i>Pseudo-nitzschia</i>	1	1	0	0	0	2	0
<i>Thalassionema</i>	0	3	0	0	0	2	0
unidentified pennate	25	35	27	11	8	33	4
<b>Dinoflagellates</b>							
<i>Ceratium</i>	16	21	11	28	21	11	31
<i>Dissodium</i>	0	1	0	1	0	0	0
<i>Proto-peridinium</i>	4	6	3	1	1	4	1
unidentified dino	0	0	1	0	0	0	0
<b>Total</b>	375	350	216	307	464	208	199
<b>Cells per Liter</b>	690.00	364	375.84	1295.54	1224.96	222.56	342.28