Diatom diversity and primary production in Barkley Sound and Effingham Inlet, Canada based on light microscopy.

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Non-technical summary

Phytoplankton along continental margins play a large role in the global carbon cycle and contribute approximately 15% to the planet's net annual primary production. The role of diatoms, a significant phytoplankton group, is of special interest because they are the major players in biogeochemical cycles. Diatoms are especially important in carbon sequestration because they are the main transporters of carbon to the sediment. They are also the main primary producers. However, the scientific community has yet to agree on how diversity and production are related, and how that relationship might vary due to global climate change. The unique and complex system found in Barkley Sound and Effingham Inlet provide an excellent opportunity to study the ecological relationship between diversity and primary production. By using light microscopy and incubations, diversity and primary production values were determined for Barkley Sound and Effingham Inlet. To resolve environmental conditions that could have influenced production, CTD hydrocasts were deployed and nutrient samples were collected. It was concluded that *Skeletonema sp*. experienced an early bloom in March even though conditions were not optimal. It was also concluded that in Barkley Sound and Effingham Inlet there was not a relationship between primary production and diatom diversity.

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Abstract

In view of global climate change it is important to understand the ecological relationship between diversity and production. The goal of this investigation was to determine the relationship between community phytoplankton diversity and primary production in Barkley Sound and Effingham Inlet in March 2010. Samples were collected at the chlorophyll-a maximum depth and light microscopy was used to identify and quantify abundances and diversity of the main diatom genera present. *Skeletonema sp*. was the dominant diatoms at all stations, with proportions between 80 – 97%. Shannon diversity values were low for all stations, and chlorophyll-a (Chl-a) concentrations were low as well (0.37 - 1.74 μg/L). Primary production experiments were performed with on shore incubations and measured with an Ocean Optics oxygen electrode. Primary production values were between 1.7 – 4.0 mg O2/L/day. In addition to identifying phytoplankton and calculating production, other environmental factors that influence primary production were investigated, such as nutrients. It was concluded that *Skeletonema sp*. experienced an early bloom in March even though conditions were not optimal. No relationship between production and diversity was found in Effingham Inlet, but there was a negative correlation found in Barkley Sound.

Introduction

In recent years, there has been increased interest in processes that are influenced by phytoplankton communities, such as carbon dioxide (CO_2) uptake, primary production, chemical fluxes, and global climate change (Harris et al. 2009; Pemberton et al. 2004; Taylor et al. 1996). Primary production is a key process in environmental cycles, and has been studied extensively with respect to photosynthesis, nutrients, light, and mixing (Agard et al. 1996; Duarte et al. 2006). However, relatively few studies have included phytoplankton community diversity in their analysis of primary production dynamics. Given the changes in diversity that could be caused by global climate change it is imperative to study the relationship between production and community diversity. Characterizing phytoplankton community diversity will help develop the understanding of ecosystem functioning, as diversity may play a major role in the dynamics of primary production (Jouenne and Lefebvre 2007; Duarte et al. 2006). The role of diatoms, a significant phtyoplankton group, is of special interest because they are the major players in biogeochemical cycles (Wawrick et al 2003).

Diatoms are a significant taxonomic group in the global carbon cycle and contribute approximately 15% to the planet's net annual primary production (Muller-Karger et al. 2005). Diatoms are important in carbon sequestration, as they are the main transporters of carbon to sediment (Wawrik et al. 2003). During spring blooms, diatoms often compose more than 90% of total biomass, and they are the fundamental link to higher trophic levels. Therefore, it is important to understand how changes in diatom diversity could affect the dynamics of primary production and CO_2 uptake in the oceans (Duarte et al 2006; Ward 2008).

Diatoms are especially important in temperate and polar latitudes, as well as along coasts where nutrients are abundant from runoff and upwelling (Thomas et al. 2004). The west coast of Vancouver Island is a very productive area (Harris et al. 2009). Barkley Sound is a large, open embayment located on the southwestern side of Vancouver Island.

Effingham Inlet is a smaller fjord that opens into Imperial Eagle Channel on the eastern side of Barkley Sound (Fig. 1).

Fig 1. Map of Barkley Sound and Effingham Inlet with stations labeled. TC4 is located between TC1 and TC2 in the yellow oval.

Despite the importance of diatoms and their role in primary production little is known about how diversity relates to primary production in these areas. Other studies have investigated the spatial and temporal distributions of microplankton on the southwestern side of Vancouver Island, but not in conjunction with primary productivity (Taylor and Haigh 1996; Mackas and Sefton; 1982, Harris et al. 2009).

The primary goal of this research is to determine the diversity of the natural phytoplankton communities on the southwestern coast of Vancouver Island by using light microscopy. Barkley Sound and Effingham Inlet were chosen because it is expected they would have very different physical and chemical properties. The second goal is to relate the phytoplankton species present in the water column to primary production. It was anticipated that in Barkley Sound and Effingham Inlet, phytoplankton community diversity and primary production would co-vary as the availability of dissolved inorganic nutrients (nitrate, phosphate, and silicate) varied at each station. Specifically, it was expected that the lowest diversity would be found at the location with the highest primary production.

Methods

Field Methods

Seven stations were sampled on March 21 and 22, 2010 using R/V Barkley Star and R/V Alta, in Barkley Sound and Effingham Inlet. Water samples were collected at the chlorophyll-a maximum from a 5 L Niskin sampling bottle. Three stations were located in Effingham Inlet and four in Barkley Sound. The station TC4 is located between TC1 and TC2, but the latitude and longitude are unknown. In addition, due to CTD malfunctions, there was no hydrocast at this station. At each station, 100 ml were sub-sampled, fixed with 5% formalin, and stored in an 8 oz jar. Water from a 20 µm mesh net tow was also collected for identifying phytoplankton. Temperature and conductivity was measured with the shipboard SeaBird Electronics CTD, and a sub-sample of water was collected for nutrients (nitrate, phosphate, and silica). The Marine Chemistry Lab at the School of Oceanography analyzed these samples.

Laboratory Methods

Cells in phytoplankton samples were allowed to settle in 100 ml counting chambers for at least 24 hours. The top 90 ml were removed and 2 ml of the remaining 10 ml concentrate were used for species identification and calculating abundances. Cells were enumerated with one transect across the slide at 25x magnification. Cells were identified to the genus level. All counts were then converted to cells per liter (Horner et al. 2005). Diatom diversity was calculated using the Shannon-Wiener function for each sampling occasion.

Primary production was measured in 48-hour incubations using 130 ml bottles and water from the chlorophyll maximum. Incubations were held in mesh bags that simulated the light level at the depth at which the water was collected. Incubations took place in an acrylic water bath with a surface freshwater pump at the shore lab. Each bottle had an Ocean Optics oxygen pad placed inside, by which an Ocean Optics NeoFox microelectrode probe read oxygen levels. The probe was calibrated with 0% and 100% dissolved oxygen solutions. Because of a limited supply of NeoFox oxygen pads, primary productivity was measured with duplicates at only two stations to determine averages (Spencer 2010).

Results

Hydrography was investigated to determine other factors that could have influenced primary production. In general, temperatures in the surface layer (2-5m) throughout Barkley Sound were between 8.7 – 9.0 ºC, and in Effingham Inlet, these values ranged from 8.6 – 8.9 ºC. Chlorophyll-a (chl-a) max depths were between 2-5 m and chl-a concentrations were 0.37 - 1.74 μg/L (Fig. 2) throughout the Sound. This data was used as a proxy for phytoplankton biomass.

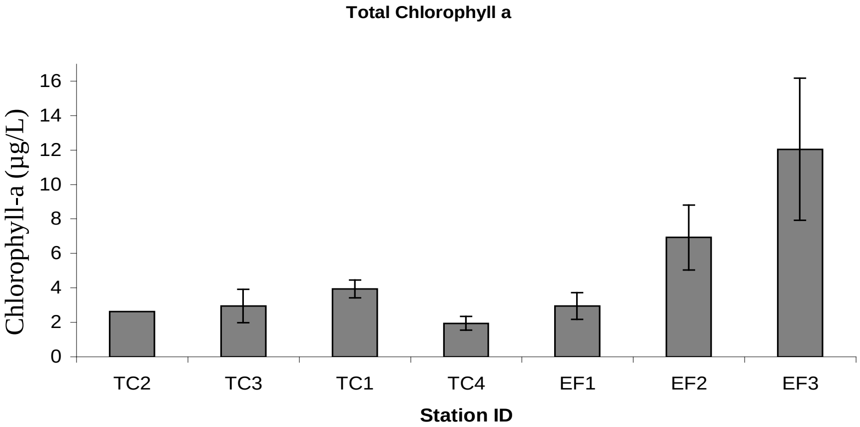
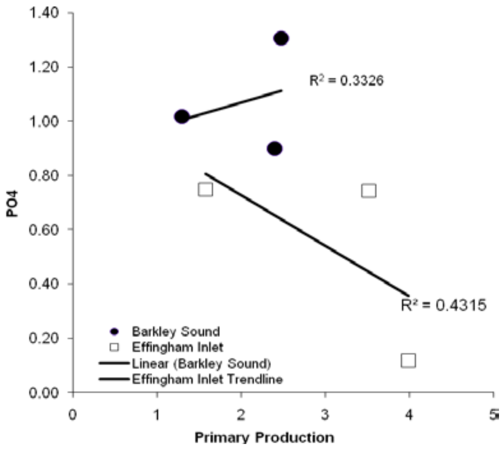
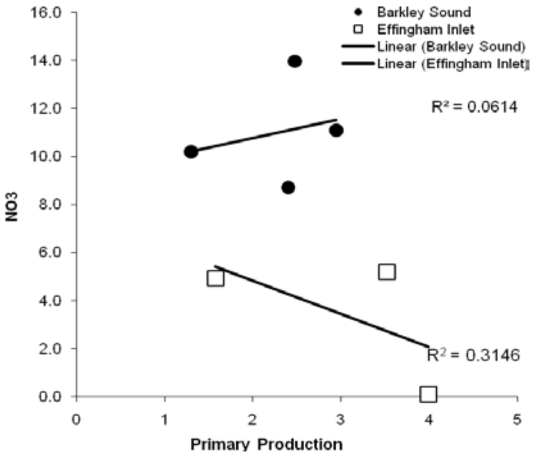


Fig 3. Total chlorophyll-a at each station with error bars determined from triplicate measurements.

Nitrate concentrations in surface waters ranged between 0.10 and 13.97 µM, phosphate ranged between 0.74 – 1.31, and silicic acid were 5.86 – 31.82 µM. These figures are plotted against production to determine if relationship existed between the two (Fig 3).





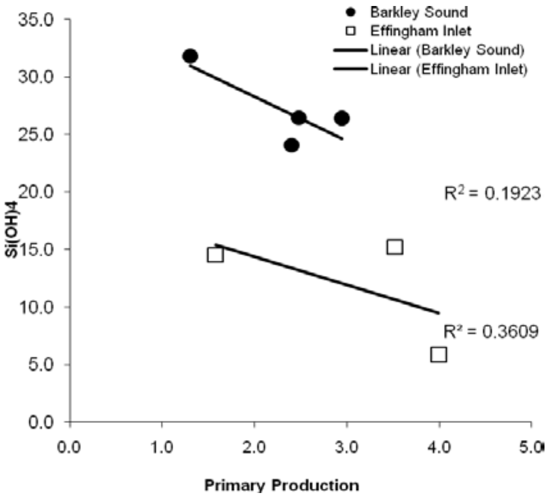


Fig. 3. Nutrients measured were nitrate, phosphate, and silicic acid. Station EF1, located at the head of Effingham Inlet, had lowest nutrient values.

Fourteen diatom genera were identified during the two day sampling period. Diatom community diversity revealed that centric diatom, *Skeletonema sp*. dominated all samples with proportions between 80 and 97% (Table 1, Fig. 4). To a much lesser extent *Thalassiosira spp*., *Thalassionema spp*., and *Cyldrotheca spp*. were also present in all samples. Other genera were present in very small proportions (>0.01%), and not all genera were found at each station.

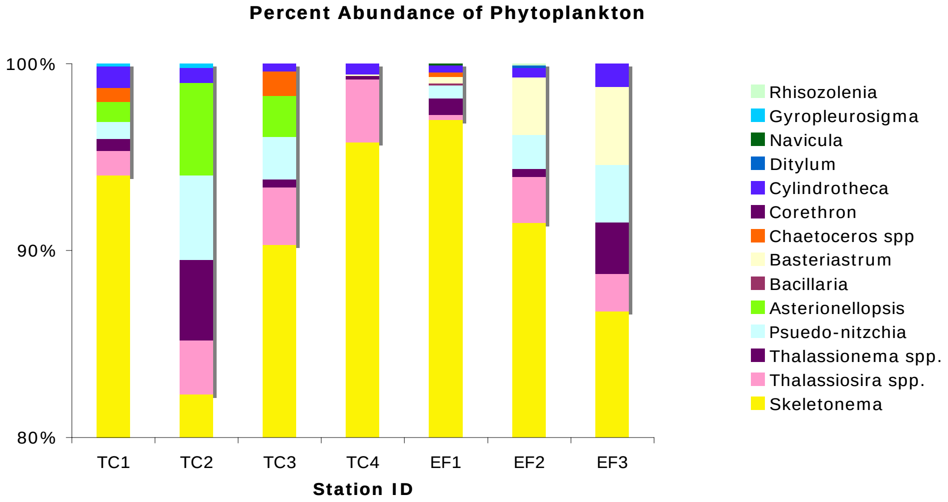


Fig 4. Percent contribution of each phytoplankton genera at stations in Barkley Sound and Effingham Inlet. All stations were dominated by *Skeletonema sp.*

Table 1 The number of genera present and the Shannon Index givin for each station

Phytoplankton Taxa	TC1	TC2	TC3	TC4	EF1	EF2	EF3
Total # genera	8	7	7	5	9	8	6
Total abundances	8.4E+05	5.4E+05	1.6E+06	5.9E+05	6.3E+05	1.1E+06	1.1E+06
Shannon Index	**0.34**	**0.74**	**0.47**	**0.20**	**0.19**	**0.42**	**0.60**

A PC-ORD4 cluster analysis was also used to create a dendogram to show station similarities based on species composition. By using 50% of the information, stations sorted into three groups as shown in Figure 6.

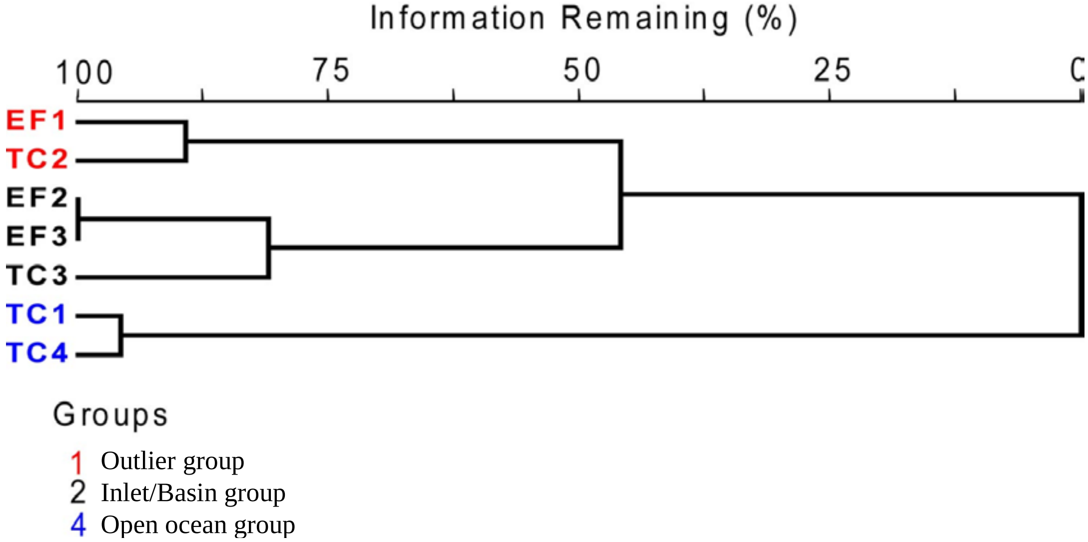


Fig. 6. A cluster dendogram displaying stations clustered based on similarity from species composition. The scale is the amount of information used to create branches.

Shannon-Weiner Diversity Index values (Table 1) were generally low (0.20–0.70). Production values for Effingham Inlet were 1.6 - 4.0 mg O_2/L/day. The highest productivity was found at the station located at the head of the inlet, EF1. In Barkley Sound, production ranged from 1.7 – 3.0 mg O_2/L/day and in Effingham Inlet production was 1.6 – 4.0 mg O_2/L/day. Production and diversity were plotted against each other to determine if a relationship existed between the two (Fig. 6) (Rombeau 2010). R^2 values for Effingham Inlet and Barkley Sound were 0.0627 and 0.9528 respectively.

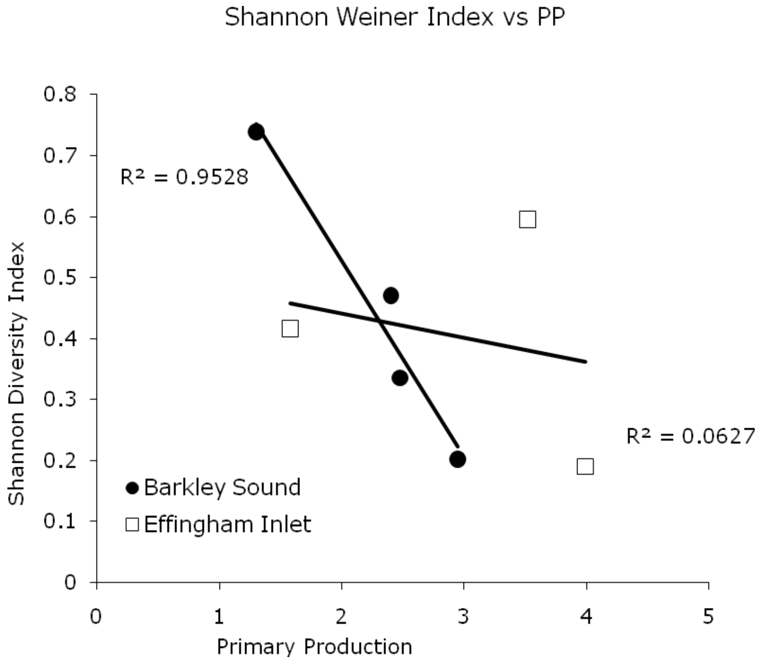


Fig. 7. Shannon Diversity Index vs. production at all stations. A correlation exists between in Barkley Sound but not Effingham Inlet.

Chl-a and production were also plotted against each other to see if a relationship existed between the two. The R^2 values for Effingham Inlet and Barkley Sound were 0.0081 and 0.7425 respectively.

Discussion

Hydrology

Barkley Sound is a dynamic area influenced by the Vancouver Island Coastal Current (VICC). This current is driven by water that comes from the Straight of Juan de Fuca (Freeland et al. 1984), as well as the deeper California Undercurrent (Taylor et al. 1996). Upwelling favorable winds generally occur between March and September. This causes the VICC to bring nutrient laden water on the continental shelf and possibly into

Barkley Sound. Besides river input, these currents are possible nutrient sources for Barkley Sound (Taylor et al. 1996). In earlier studies (e.g. Harris et al. 2009; Taylor et al. 1996), nutrients on the continental shelf off Vancouver Island and Barkley Sound, were high. Nitrate was about 4.5 to 8.1 µM, silicate was about 12.6 to 18.4 µM, and phosphate was about 0.4 to 0.1 µM (Harris et al. 2009). During this study, nutrients were even higher in Barkley Sound (Fig. 3), which indicates that there might have been more upwelling events and that nutrients were not yet consumed by blooming phytoplankton. In Effingham Inlet, nutrients were lower, possibly due to less complicated circulation patterns from the morphology and sill of the inlet (Monk 2010).

Phytoplankton community composition

This is the first investigation in Barkley Sound and Effingham Inlet to study the diversity of phytoplankton communities in conjunction with primary production. Between Barkley Sound and Effingham Inlet, the number of genera at each station was similar. During the March sampling period, *Skeletonema sp*. dominated phytoplankton abundances at all stations. Even though this species is known to bloom during April in the Strait of Georgia, it has experienced blooms as early as March in coastal fjords (Taylor and Haigh 1996).

Previous studies suggest that *Skeletonema sp*. prefer semi-enclosed waters in temperate regions of the world (Taylor and Haigh 1996; Smayda 1957). In agreement with these previous studies, the diatom assemblage at each station consisted of 80 – 95% *Skeletonema sp.*, even at stations located closest to the ocean. This suggests that *Skeletonema sp.* were able to bloom early in the season. The higher nutrient values suggest that they were in an early stage of the bloom. *Skeletonema* is a hardy genus that can tolerate a wide range of environmental conditions, and blooms early in the spring to take advantage of the higher nutrients and light conditions (Strickland 1983). Chl-a values in Barkley Sound were generally lower than Effingham Inlet, but they a positive correlation with primary production. This implies that even though they had low biomass, the diatoms were growing. In Effingham Inlet there was no relationship between the production and chl-a, however the highest values of chl-a were found at EF3. EF3 is located at the interface between Effingham Inlet and Imperial Eagle Channel, and the mixing of these two water bodies might result in more productive waters.

The small abundances of *Chaetoceros spp*. and *Thalassiosira ssp*. present in the samples also suggest that Barkley Sound and Effingham Inlet were experiencing an early spring bloom. The species succession in areas like Barkley Sound is similar to other temperate water basins, like Puget Sound. Generally, they dominate later during the year and persist through the summer in these areas. Therefore, during the summer months, Barkley Sound and Effingham Inlet would be expected to have higher abundances of *Chaetoceros spp.* and *Thalassiosira spp.* and lower abundances of *Skeletonema sp*. (Strickland 1983).

A study of the diatom communities at each station was important to investigate in order to conclude if there were any important patterns observed in Barkley Sound and Effingham Inlet. A PC ORD4 cluster analysis was used to determine stations that were similar based on species composition and abundances. When 50% of the information remains in the analysis three groups can be distinguished using this statistical approach. As expected the stations located closest to the open ocean, TC1 and TC4, were grouped together. They have similar species compositions and similar production values (2.48 and

2.94 mg O_2/L/day), low to intermediate Shannon Diversity values, and similar nutrient concentrations. EF2, EF3, and TC3 were grouped together because they had 6-8 genera identified at each station, the highest abundances (> 1 million cells), and they had similar species compositions. These stations were located near the entrances of Effingham and Alberni Inlets into Barkley Sound, which implies they may have similar circulation patterns. However, it is interesting to observe that EF1 and TC2 group together. EF1 is located at the head of Effingham Inlet and TC2 is located in the southern portion of Barkley Sound. Their species compositions and abundances are dissimilar from each other as well as the other two groups. They have neither similar values of production nor nutrient concentrations. However, they have the same Shannon-Weiner Diversity value of 0.20, which was the lowest at all the stations. This grouping could be due to several factors. It is possible that an environmental variable that went unmeasured during the study affected the community composition at these two stations. Another reason for this grouping could be due to the random patchiness found in the marine environment. Organisms in the ocean are patchily distributed and these patterns are highly variable. This phenomenon arises from both physical and biological factors (Strickland 2010). These samples represent a snapshot of the community diversity which could easily vary temporally and spatially.

Production and Diversity

Shannon-Weiner Diversity Index, one of many diversity indices, was calculated for each station. It was chosen because it takes into account species richness and evenness (Duarte et al 2006). Because each station was dominated by *Skeletonema spp*. the diversity values were low, 0.19 - 0.74. During blooms, it is not uncommon for diatoms to compose 90% of the phytoplankton abundances (Duarte et al 2006). But it is unknown how the diversity of these diatom assemblages affects the production of blooms on SW Vancouver Island.

In terrestrial environments, observations indicate that total community productivity increases with increased diversity. However, in the ocean, production is due to phytoplankton and other unicellular organisms that grow rapidly, but have low biomass (Pemberton et al. 2004). In these environments, community productivity increases with decreasing diversity. In recent work, in a coastal lagoon in France, it was shown that the highest phytoplankton diversity was associated with decreases in biomass and production (Duarte et al. 2006). In the Celtic Sea, the highest rates of primary production corresponded with a phytoplankton community dominated by one diatom, *G. delicatula* (Pemberton et al. 2004). This correlation was also found in Barkley Sound, but not in Effingham Inlet. This suggests that these two areas behaved differently in terms of hydrography and nutrient condition (Monk 2010). In Effingham Inlet, at station EF1 located at the head of the inlet, the highest production rates co-occurred with the lowest diversity value. This station, as well as the stations in Barkley Sound, had data that agreed with other studies (Pemberton et al. 2005; Duarte et al. 2006). In the rest of Effingham Inlet, however, there was no relationship found between diversity and production.

There could be several reasons for this conclusion. First, the measurements of production might have been affected by the oxygen method used. There was no statistical way to measure the accuracy of the Ocean Optics oxygen probe because triplicate samples could not be performed. Therefore measurements were large approximations without standard deviations. Also, the phytoplankton identification is biased because sampling scheme was limited spatially and temporally. A more thorough sampling scheme that includes sampling on longer time scales would give a better picture of the phytoplankton community diversity.

Ultimately, a more detailed study is needed to measure all the components of the pelagic ecosystem, such as animal and microbial diversity, in order to accurately test the ecological hypothesis that relates phytoplankton diversity to productivity. For example, in the marine ecosystem, zooplankton grazers can limit the diversity of phytoplankton because of selective feeding. In addition, phytoplankton rely on the regenerated nutrients made available by bacteria (Strickland 1983). The diversity of these organisms is important to understand how diversity might affect primary production.

Conclusion

Barkley Sound and Effingham Inlet are complex coastal embayments that showed low diversity, but high abundances of diatoms, especially *Skeletonema sp*. Composition of diatoms consisted chiefly of the centric diatoms *Skeletonema sp*. and to a lesser extent *Thalassiosira spp*. and *Thalassionema spp.* There was a negative relationship between primary production and diatom diversity found in Barkley Sound, but not in Effingham Inlet. This suggests that the dynamics of primary production in Barkley Sound and Effingham Inlet behave differently, and are controlled more by environmental factors than by diversity. This paper emphasizes that studies attempting to determine the dynamics of primary production and how production affects climate change and other biogeochemical cycles, should not overlook evaluating phytoplankton diversity. (Agard et al. 1996; Duarte et al. 2006)

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