Wavelength-specific light attenuation, chlorophyll concentration and phytoplankton communities in Puget Sound

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Running Head: Light response to phytoplankton

Non-Technical Summary

Little is known about the effect light quality has on phytoplankton. I hope to add to this knowledge base with this research project by examining the links between changes in the intensity of red and blue wavelengths of light and phytoplankton community structure as well as chlorophyll *a* concentration. I took water samples and light profiles at diverse locations in Puget Sound: Dabob Bay (Hood Canal), Elliot Bay (Main Basin), the mouth of the Skagit River (Whidbey Basin), and the Strait of Juan de Fuca. I analyzed the chlorophyll *a* concentration at the surface and chlorophyll max, as well as counting the samples for phytoplankton. Each location had different relative intensities of red versus blue wavelengths of light, and they also had marked differences in phytoplankton community structure and chlorophyll concentrations. Larger cells and greater chlorophyll concentrations seem to be related to light regimes in which red light intensity is much greater than blue, but other locations where red light is similar in intensity to blue or where blue light intensity is greater are dominated by smaller cells with a lower chlorophyll concentration. Extreme differences in the intensities of red and blue light also seem to be related to increased total concentrations of phytoplankton cells.

Acknowledgments

I would like to thank Leon Delwiche and Miles Logsdon for their help with building the light sensors. I would also like to thank Megan Bernhardt for training me to work with the microscopes, prepare slides and identify phytoplankton; and I thank Evelyn Lessard for allowing me to use her lab and equipment for my project.

Abstract

This study evaluates the link between red and blue light intensity, chlorophyll concentration and phytoplankton community structure. Light profiles and water samples were taken at thirteen stations in Puget Sound including Dabob Bay (Hood Canal), Elliot Bay (Main Basin), the Strait of Juan de Fuca and the Skagit River Mouth (Whidbey Basin). Water samples from the surface and chlorophyll max depths were used for phytoplankton counts and chlorophyll *a* analysis. Light quality was measured using red and blue bandpass filters placed over Hobo sensors and attached to a CTD (conductivity, temperature and depth sensor package). A normalized difference ratio between the blue and red light intensity (BR Ratio) was used to compare the light results with positive values indicating a higher proportion of blue to red light and negative values indicating a higher proportion of red to blue. There are many differences between the stations; each location had different light quality regimes based the BR Ratio. Dabob Bay had the highest chlorophyll concentrations with a surface average of 12.93 mg m^{-3}. The Dabob Bay stations also had the highest concentrations of cells (6.35 $x10^{6}$ cells l^{-1} for HC4 surface) and were comprised of mainly larger cells (greater than 50% at all stations) such as Chaetoceros sp. and other diatoms. The BR Ratio was negatively correlated to chlorophyll *a* concentration and the percentage of large cells in the phytoplankton community. The BR Ratio was positively correlated to the percentage of small cells. The results indicate that there is a strong effect on light quality by the phytoplankton in the water column that may affect our current views on the attenuation of light in surface waters.

Introduction

Little is known about how light quality affects and is affected by phytoplankton. Phytoplankton composition affects the absorbance and reflectance properties of the water and

therefore affects attenuation of light. If light levels are high at depth, there may be a shift in phytoplankton composition towards taxa that are less photoinhibited or that utilize red wavelengths of light more effectively. This study addresses the affect red light has on phytoplankton. When the attenuation of the red wavelengths (~650 nm) is low allowing more red light to reach phytoplankton, the dominant taxa of the phytoplankton communities and chlorophyll concentrations should be different, in terms of cell concentrations and cell types, than in areas where the attenuation of the red wavelengths is high.

Many different substances contribute to the attenuation of light in seawater. Pure water itself attenuates light. For PAR, longer wavelengths (red) are attenuated by water more strongly than shorter wavelengths (blue). However, other substances affect the absorption of light in water. Pigments, colored dissolved organic matter (CDOM) and suspended solids all change the properties of the water and reflect and absorb different wavelengths of light. For example, many dissolved organic substances absorb strongly in the ultraviolet (UV) and blue wavelengths (Hoepffner 1992).

Different types of phytoplankton react to light differently. In general diatoms have an optimal light level for growth that is higher than dinoflagellates, and because of this diatoms tend to have higher concentrations of chlorophyll (Miller 2004). Phytoplankton can also acclimate to changing light levels by either increasing pigment concentrations when light levels are too low, or by increasing their growth rate at high irradiances to take advantage of the extra energy available (Finkel 2006).

Because chlorophyll *a* and *c* absorb red light, changes in the amount of irradiance of the wavelengths at 665 nm could affect chlorophyll concentration, but the type of changes may depend on the phytoplankton present in the water.

Light is a necessary requirement for phytoplankton growth by providing the energy for use in photosynthesis; however, too much light, or light of the wrong wavelengths (mainly UV) can damage phytoplankton or cause photorespiration, where phytoplankton respire even though they have the nutrients and light to perform photosynthesis and cell growth is negatively affected (Miller 2004; Leu 2006). Light controls whether phytoplankton will maximize growth or maximize production of light harvesting components (Korbee 2005). Light intensity is not directly linked to chlorophyll concentration due to other factors including nutrient availability and the different responses by different types of phytoplankton. Downwelling light intensity decreases exponentially with depth; however, chlorophyll concentration does not. Chlorophyll concentration can have a peak at depth or can be nearly constant from the surface to a depth of several tens of meters (Miller 2004).

Red wavelengths of light in general have a shallow penetration depth and typically do not penetrate deeper than a few meters because of the absorption affect of water which acts preferentially on the longer wavelengths (Richardson 1996). As a result, PAR at depth is composed primarily of the blue wavelengths of light. The effect of changes in the intensity of red light on phytoplankton is uncertain, and it may differ for different phytoplankton taxa (Miller 2004; Martin 2006). A change in penetration depth of red light should be able to increase or decrease chlorophyll *a* concentrations because this pigment has an absorption peak in the red wavelengths that is secondary to the blue absorption peak.

Chlorophyll *a* is the primary pigment in phytoplankton that absorbs light for use in photosynthesis. Chlorophyll *a* absorption peaks in the visible light between 400 and 475 nm and around 665 nm (Roesler 1995; Ruddick 2006); it reflects most of the light with wavelengths between 475-550 nm (Miller 2004). Chlorophyll *c*, a major secondary pigment, peaks in

absorption at 475 and 650 nm, but reflects light from about 500-550 nm (Miller 2004). Phytoplankton can change the amount of chlorophyll in the cell in response to the amount of light the organism receives. Chlorophyll *a* concentration generally increases when the cells are not receiving enough light to grow, such as in low light conditions with ample nutrient concentrations.

The attenuation of red light changes by location as the concentrations of dissolved or suspended materials in the water changes is not understood. It is also unclear what effect these changes in attenuation will have on chlorophyll concentration. A study on the effect of red and blue light on the macroalgae *Porphyra sp.* found that blue light increased the concentration of photosynthetic pigments while red light increased cell division and cell storage (Korbee 2005).

This study aims to clarify the relationship between light quality and phytoplankton by measuring chlorophyll *a* concentration and examining the phytoplankton community through cell counts. By better understanding the relationships between changes in red and blue light intensity and phytoplankton, we can better understand the phytoplankton community through in situ measurements of these wavelengths of light. These relationships can be used for new creating new remote sensing models for phytoplankton communities that may be able to determine the types of cells present in the water column (diatoms, dinoflagellates, cyanobacteria etc.) based on how these cells absorb and reflect light. Current models rely on Ocean Color algorithms and can only be used to estimate chlorophyll *a* concentrations. Chlorophyll *a* estimations are not very useful for coastal areas because the use of shorter (blue and green) wavelengths requires a pixel size of 1 km. Future algorithms based on the water-leaving radiance of red wavelengths could have a pixel size of 250 m, which would allow for better estimations and for spatial patterns to be seen in coastal areas such as Puget Sound. In order to develop better algorithms for estimating

chlorophyll *a*, more data on the relationship between red light and phytoplankton is needed.

Methods

Sampling for this project was done at four locations in Puget Sound (Fig. 1) using the *R/V Thomas G. Thompson* from March 19 to March 23. To take advantage of optimal light availability, sampling took place between 1030 and 1500 local time. At two locations (Main Basin and Dabob Bay) four stations were sampled (Table 1). At a third location (Skagit River area) three stations were sampled. In the Strait of Juan de Fuca two stations were sampled.

Onset Hobo light and temperature sensor pendants were used to acquire the wavelength specific light intensity. The sensor pendants were modified into a sensor package of two sensors taken out of their casings and with band pass filters placed over the light sensor. The sensor casings were then attached to and deployed using the CTD cage about 2 m above the CTD sensors and Niskin bottles. Because of the placement of the light sensors, the light intensity measured was the light reaching the water sampled for chlorophyll and phytoplankton. A normalized difference ratio (Eq 1) was used to compare the intensities of the red and blue wavelengths between stations.

Equation 1: BR Ratio = (Blue-Red) / (Blue+Red)

This ratio accounts for the differences in incident light at each station due to cloud cover and time of day.

Water samples were collected from the surface water and at the depth of the chlorophyll maximum (as determined by CTD fluorescence) using the 10 L Niskin bottles on the CTD. Most stations lacked a clear fluorescence peak, or had a peak at the surface, so for these stations the depth just above where the fluorescence began to decline was chosen for the chlorophyll max depth samples. For each station, these water samples were used for a chlorophyll *a* analysis

following the technique described by Newton (unpubl.). In addition, two samples at both depths were taken from the ends of each transect and from the first Strait of Juan de Fuca station for a phytoplankton count. The phytoplankton samples were divided into treatments with Lugol's Iodine for settling counts and gluteraldehyde for epifluorescent counts.

For the chlorophyll analysis, three 100 ml samples were collected for each depth and analyzed for chlorophyll *a* concentration following the sonication and fluorometer procedures of Newton (unpubl.) to quantify the changes in chlorophyll concentration between stations and depths. Water was filtered through 0.7 micron GF/F filters, and filters were treated with 10 ml of 90% acetone. These filters were then sonicated using a Model CV26 sonicator by Sonics and Materials Inc. and a Model VC502 High Intensity Ultrasonic Processor and centrifuged using an IEC Centram CL2 from International Equipment Company. A sub sample of 10 ml was inserted into a Turner Designs TD-700 fluorometer, and in the case of the Dabob Bay samples, diluted by a factor of 10. At two stations on each end of a transect and at the first Juan de Fuca station, an additional 1 L sample was taken for both depths and used for the phytoplankton counts.

For the phytoplankton counts, the gluteraldehyde treated samples were stained using DAPI and Proflavin stains following the procedures provided by Lessard and Bernhardt (pers. comm.). The gluteraldehyde samples were split into two and filtered onto a 0.2 micron black filter and a 0.8 micron black filter. These samples were counted in a dark lab under a Zeiss Standard Epi-fluorescent microscope for autotrophic organisms. Cyanobacteria, picoeukaryotes, and autotrophic flagellates were counted at 1000x and cryptophytes and autotrophic dinoflagellates were counted at 400x. The Lugol's treated samples were poured into settling chambers and counted for larger autotrophs (diatoms, autotrophic ciliates and larger autotrophic dinoflagellates) under a Zeiss Axiovert 35 Inverted microscope at 400x. The program used for

counting the Lugol's samples was the Microbiota software with a Summa Sketch Digitizing Tablet.

Results

The Dabob Bay stations were consistently different from the other stations in terms of light quality, chlorophyll *a* concentration and phytoplankton community structure. These stations were characterized by extremely low BR Ratios (between -1 and -0.75) (Fig. 2), chlorophyll *a* concentrations in excess of 10 mg m^{-3} (Fig. 3) and were dominated by large diatom cells (Fig. 4). The Skagit River mouth stations were also different, having slightly higher chlorophyll *a* concentrations (average greater than 1.9 mg m^{-3}) than the Main Basin and Strait of Juan de Fuca stations, and having a higher proportion of medium sized cells than the other stations; however, no light quality data was available for the Skagit stations due to technical difficulties. The Main Basin and Strait stations were similar except that the quality of light was different (slightly negative BR Ratios in the Main Basin and positive BR Ratios in the Straits) and the Strait stations had slightly higher cell concentrations.

As expected, light intensity decreased at depth (Fig. 5). The Main Basin samples and the Dabob Bay samples showed the unexpected trend of a higher average red intensity than the blue and green wavelengths. The Strait of Juan de Fuca samples typically had a higher average blue intensity than red or green. At each station in Dabob Bay, the intensity of blue wavelengths of light was zero at the chlorophyll max depth of 5 m.

Results for the BR Ratio are summarized in Table 3. The Dabob Bay and Main Basin stations had negative values of the BR Ratio indicating more red light than blue (Fig. 6). The Juan de Fuca stations all had positive values, more blue light than red. Dabob Bay also a trend to more negative values at depth than at the surface, with the extreme value of -1 at several

locations indicating an undetectable amount of blue light. The other stations (Main Basin and Strait) tended to be more positive at the chlorophyll max depth than at the surface.

The Strait of Juan de Fuca had the lowest concentration of chlorophyll *a* (Fig. 3). The surface samples in the Straits had an average of 0.64 mg m^{-3} of chlorophyll *a* (Table 2). The highest concentrations of chlorophyll *a* were found in Dabob Bay (12.83 mg m^{-3} at surface). The second highest chlorophyll *a* values were found near the Skagit River (2.52 mg m^{-3} at surface). In the Dabob Bay samples, higher chlorophyll *a* concentrations were found in surface waters than at the chlorophyll max (5 m in Dabob Bay). At other stations the chlorophyll *a* concentrations were much lower. For high concentrations of chlorophyll *a*, there was a correspondingly higher standard deviation.

Phytoplankton cell concentrations were compared for Dabob Bay, Strait of Juan de Fuca and the Main Basin stations (Fig. 4). Dabob Bay had the highest concentrations of cells at both the surface and the fluorescence max depth. Station HC4 had the highest concentration of cells in the surface waters 6.35×10^6 cells l^{-1}. In Dabob Bay, the surface stations had higher cell concentrations than at depth, but the reverse was true for the other stations in which the chlorophyll maximum depth had higher concentrations than the surface waters. Dabob Bay had the highest concentrations (greater than 2.14×10^6 cells l^{-1}) of large cells (diatoms and large autotrophic dinoflagellates greater than 20 μm). The Strait of Juan de Fuca station had the highest concentration (5.22×10^6 cells l^{-1}) of the smallest cells (picoeukaryotes, cyanobacteria 2 μm and autotrophic flagellates less than 10 μm). The highest concentration of medium sized cells (cryptophytes and small autotrophic dinoflagellates between 10 and 20 μm) was found at the Whidbey Basin stations (1.28×10^6 cells l^{-1}).

Comparing the cell abundance as a percent of the total concentration of cells for each

station revealed the dominant group of autotrophic organisms (typically small cells) in the water samples (Fig. 6). For the Dabob Bay samples, large cells comprised the majority of the cell counts (ranging between 60-70% of the total depending on the station) most of which were diatoms. The Main Basin samples were dominated by smaller cells (85-90%) mainly picoeukaryotes and cyanobacteria. About 90% of the Juan de Fuca samples were small cells. The Skagit River Mouth samples were 60-80% small cells; however, these stations also had a higher proportion of medium sized cells than elsewhere (10-30%).

Discussion

The results of this study indicate that there are links between the quality of light at a station and the chlorophyll concentrations and community structure of phytoplankton. This can be seen by breaking down the stations into different light types based on the BR Ratio. The first light condition (Category I) is where the intensity of blue light far exceeds that of red at depth, such as at the Juan de Fuca stations (BR Ratio is positive). This case is typical of open ocean waters since longer wavelengths of light (red) do not penetrate the water column as deeply as shorter wavelengths (blue) (Martin 2006), which is why in deep water only blue light is visible. The second case (Category II) occurred at the Main Basin stations with a BR Ratio closer to zero indicating that the intensities are nearly equal for the blue and red light.. The third light condition (Category III) is where the intensity of red light is much greater than the intensity of blue light (BR Ratio is negative). The Dabob Bay stations fall into this category. Because of how water attenuates the different wavelengths of light, having a higher intensity of red light compared to blue was not expected.

The presence of Category III waters was unexpected. At the Dabob Bay stations, blue light was not detected by a 5 m depth, while the red and green light intensities were still

measurable (Fig. 5). Part of this may be due to instrument sensitivity, but a positive BR Ratio was measured in the Strait of Juan de Fuca stations by the same sensors assigned to one filter for the duration of the cruise. This means that even if the blue light intensity was not zero in Dabob Bay, it was still much lower than measured at the rest of the stations, so the decrease in the BR Ratio found for the Dabob Bay stations was real.

A possible explanation for the decrease in the BR Ratio at Dabob Bay is the phytoplankton community found there. Algae in the water column alter the light penetrating the surface of the water depending on the type of algae present and their accessory pigments (Atlas 1980). The cells can reduce the incoming light from a wide range of wavelengths (blue to red) to narrow bands of wavelengths (blue, green, and blue-green depending on cell types) (Atlas 1980). Chlorophyll *a* has a large effect as well; it absorbs blue wavelengths of light and re-emits the light in the red spectrum through fluorescence (Richardson 1996; Martin 2006). Since the light sensors were passing through the phytoplankton in the water column, this red signal was measurable by the sensors despite the fact that the red light is quickly attenuated by water (Richardson 1996). The high chlorophyll *a* concentrations found in Dabob Bay are therefore responsible for the negative BR Ratio values and the shift to a more negative value at depth

The Dabob Bay stations showed a decrease in the BR Ratio with depth. At the other stations, the BR Ratio increased as red light was attenuated more quickly than blue (Fig. 2). Chlorophyll *a* concentrations may explain these differences. The BR Ratio averaged over one meter depth at the surface and at the chlorophyll maximum depth is negatively related to the chlorophyll *a* concentration measured at those depths (Fig. 7). The strong relationship between the BR Ratio and chlorophyll *a* concentration is logarithmic with an R^2 value of 0.7096. This strong correlation indicates that the attenuation of blue light and the fluorescence of red light by

chlorophyll *a* mostly explain the BR Ratio values seen for each category.

The types of cells found at a station also have implications for the BR Ratio. As the composition of the phytoplankton community shifts towards smaller cells, the BR Ratio increases, and as the composition shifts to larger cells, the BR Ratio decreases (Fig. 8). The BR Ratio increased linearly as the percentage of small cells in the phytoplankton population increased (R^2 was 0.6087); however, the BR ratio decreased logarithmically as the percentage of large cells increased (R^2 of 0.7269). The percentage of medium sized cells did not vary even though the BR Ratio did. Based on the Dabob Bay and Strait stations, higher concentrations of cells seems to be more closely associated with extreme values of the BR Ratio (near positive or negative 1) while lower concentrations, such as those found in the Main Basin, may be more associated with values near zero (Fig. 4).

Conclusions

Phytoplankton clearly affect the light quality of the water column. Each location sampled had a different light quality ranging from blue light dominated (Category I) to red light dominated (Category III). The shifts from blue to red light dominance can be explained by the attenuation and fluorescence by chlorophyll *a*. Phytoplankton community composition also has a role in changing the quality of light penetrating through the surface waters. Higher concentrations of large cells (diatoms mainly) result in a highly negative BR Ratio, while higher concentrations of smaller cells result in a positive BR Ratio.

Further studies could examine how individual taxa of phytoplankton affect light quality, or what role heterotrophic cells may have. These categorizations of light quality may also be applicable elsewhere in Puget Sound and other coastal regions. Light conditions in the water column may be different than what is typically expected based on water-leaving radiance as

measured by hand held spectroscopy or satellites, which may not have detected the shifts in light quality found in this study. The relationships found in this study may pave the way for new measurements based on the relative intensities of blue and red light to be used for in situ estimations of phytoplankton community structure, and may also assist in the use of red light in the remote sensing of algal blooms.

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Table 1: Stations sampled for this study. M stands for the Main Basin transect, S refers to the Skagit River delta transect, JDF stands for the Strait of Juan de Fuca stations and HC stands for the Dabob Bay transect in Hood Canal.

Station ID	Latitude (deg)	Longitude (deg)
M4	47 37.121	122 26.494
M3	47 37.886	122 26.51
M2	47 38.645	122 26.454
M1	47 39.393	122 26.510
S1	48 16.152	122 31.118
S2	48 15.579	122 32.024
S3	48 14.953	122 33.012
JDF1	48 15.717	123 20.215
JDF2	48 12.454	123 32.582
HC4	47 39.841	122 52.732
HC3	47 40.552	122 52.606
HC2	47 41.275	122 52.395
HC1	47 42.014	122 52.173

Table 2: Average Chlorophyll *a* Concentrations for each station averaged over a 1 meter interval at the surface (0 m) and at the fluorescence maximum depth. Hood Canal had the highest average chlorophyll *a* values while the Strait of Juan de Fuca stations had the least.

Transect Location	Depth in m	Average Chlorophyll *a* mg m^{-3}
Hood Canal (Dabob Bay)	0	12.83
	5	9.73
Main Basin	0	1.41
	5	1.31
Strait of Juan de Fuca	0	0.64
	10	0.66
Skagit River Mouth	0	2.52
	5 & 10	1.93

Table 3: Average normalized ratios of Blue to Red for each station. The BR Ratio is calculated using the equation BR=(Blue-Red)/(Blue+Red), so high values indicate greater blue intensity than red, near zero values mean that blue and red light were at nearly equal intensity and negative values indicate that red light intensity was greater than blue. Hood Canal had very negative values, while the Strait of Juan de Fuca stations had very positive values. The Main Basin values were negative, but were also close to zero.

Station ID	Average BR Ratio
HC1S	-0.838
HC1D	-1.000
HC2S	-0.836
HC2D	-1.000
HC3S	-0.754
HC3D	-1.000
HC4S	-0.803
HC4D	-1.000
M1S	-0.298
M1D	-0.284
M2S	-0.430
M2D	0.000
M3S	-0.184
M3D	-0.292
M4S	-0.410
M4D	-0.298
JDF1S	0.692
JDF1D	0.742
JDF2S	0.707
JDF2D	1.000

Figures

Fig. 1: Map of sampling locations in Puget Sound and surrounding areas. The northernmost station is station 1 with numbering increasing southward for the transects at Skagit River Mouth (S), Main Basin (M), and Dabob Bay (HC). The Juan de Fuca stations (JDF) are labeled individually.

Fig. 2: The ratio of blue to red light intensity was calculated using the normalizing equation of (blue-red)/(blue+red). The Hood Canal stations (HC) showed the most negative values with decreases at the chlorophyll max while the Juan de Fuca stations (JDF) had the most positive values that increased at the chlorophyll max.

Fig. 3: Average chlorophyll *a* concentrations and the average phaeopigment concentrations. Error bars are standard deviations for the samples. D refers to at the "depth" of the chlorophyll max while S refers to "surface" samples. These concentrations were lowest at the Strait of Juan de Fuca stations (JDF 1&2) and highest in Dabob Bay (HC 1-4). The second highest chlorophyll *a* values were found near the Skagit River (S 1-3). In the Hood Canal samples, higher chlorophyll a concentrations were found in surface waters than at the chlorophyll max which was taken to be 5 m depth for these stations.

Fig. 4: Cell concentrations were compared for stations at the ends of transects and for the first Juan de Fuca station (JDF1) with the concentrations separated into rough cell sizes (Large: ciliates, dinoflagellates, and diatoms; Medium: small autotrophic dinoflagellates and cryptophytes; Small: autotrophic flagellates, picoeukaryotes, and cyanobacteria). HC4 (Hood Canal) had the highest concentrations of cells at both the surface (S) and the chlorophyll max depth (D). Station M4S (Main Basin: Surface) had the smallest concentration of cells. In Hood Canal, the surface stations had higher concentrations than at depth, but for the other stations the

fluorescence max depth had higher concentrations than the surface waters. The Hood Canal stations also had higher concentrations of large cells than were found at the other stations while the Juan de Fuca stations had the highest concentrations of small cells. The Skagit River Mouth stations had the highest concentrations of medium sized cells.

Fig. 5: Average light intensity for the red, blue and green wavelengths is compared by depth and station. M represents the Main Basin samples, JDF the Juan de Fuca samples, and HC the Hood Canal samples. D refers to at the "depth" of the chlorophyll max while S refers to "surface" samples. The values measured were averaged at one meter intervals at the surface (taken as the top 2.5 m) and at the chlorophyll max (4.5-5.5 m or 9.5-10.5 m). Light intensity decreased at depth for each wavelength. The Main Basin samples and the Dabob Bay samples showed a higher average red intensity than the blue and green wavelengths. The Strait of Juan de Fuca samples had a higher average blue intensity than red or green. In Dabob Bay, the intensity of blue wavelengths was zero by 5 m depth.

Fig. 6: A comparison of the cell abundance as a percent of the total concentration of cells showed sharp differences between the Hood Canal (HC) stations and the other stations. D refers to the samples taken at the fluorescence max depth and S refers to the surface depth. In the Hood Canal samples, larger cells of mostly diatoms dominated the counts (60-70% of the total). At the other stations, smaller cells mainly picoeukaryotes and cyanobacteria were the biggest contribution to the counts (60-90%). The Skagit River Mouth stations had the highest proportion of medium sized cells consisting mainly of cryptophytes (10-40%).

Figure 7: Comparison of the BR Ratio and the chlorophyll *a* concentration. Average BR Ratios over 1 m were determined at the depths of the chlorophyll samples. The logarithmic regression shows a strong negative trend between the BR Ratio and the chlorophyll *a* concentration. The R^2

value was 0.7096.

Figure 8: Comparison of the BR Ratio and the percentage of a cell range (small, medium, large) in the phytoplankton population. Average BR Ratios were determined over 1 m at the depth of the phytoplankton samples. The percentage of small cells had a positive linear relationship with the BR Ratio (R^2 value 0.6087). The percentage of medium cells did not vary, and the percentage of large cells had a negative logarithmic regression (; R^2 value 0.7269).

Figure 1: Map of Sampling Locations

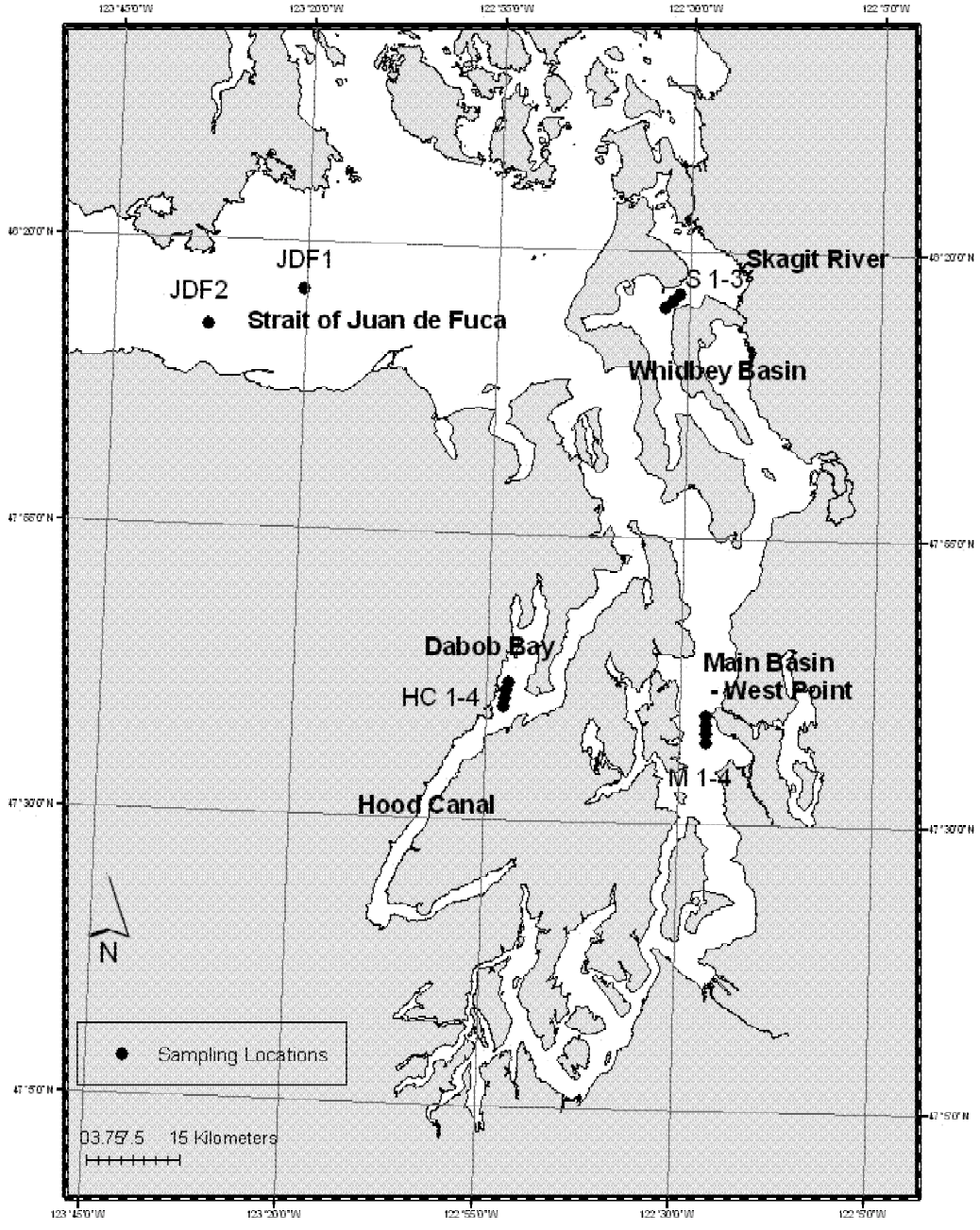

Figure 2: Normalized Light Intensity

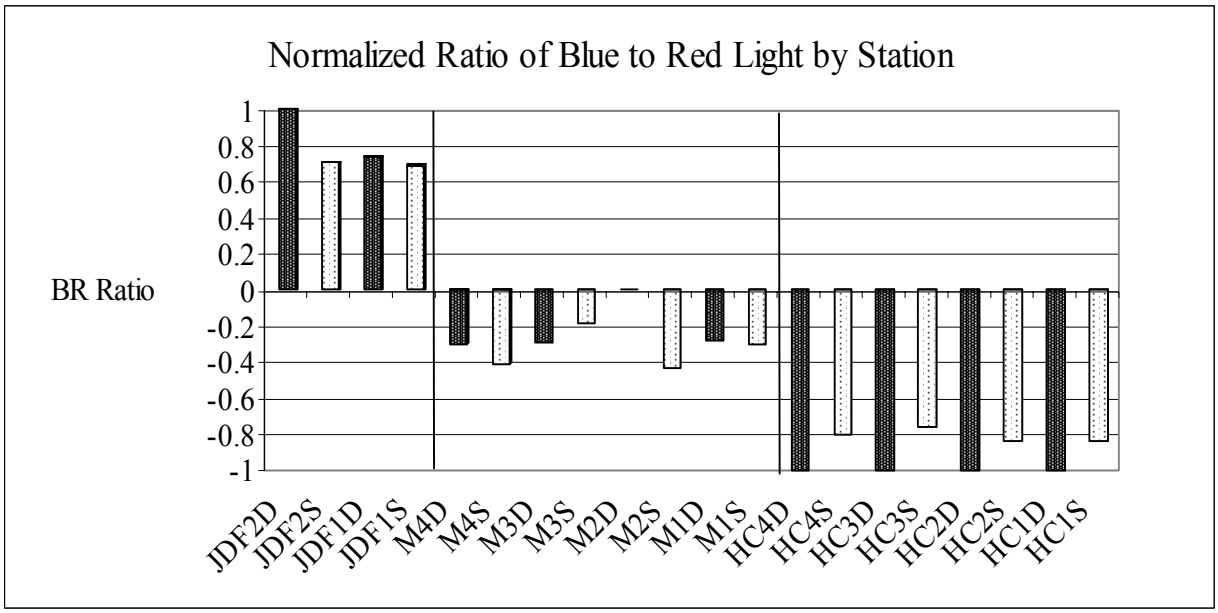

Figure 3: Chlorophyll Concentration

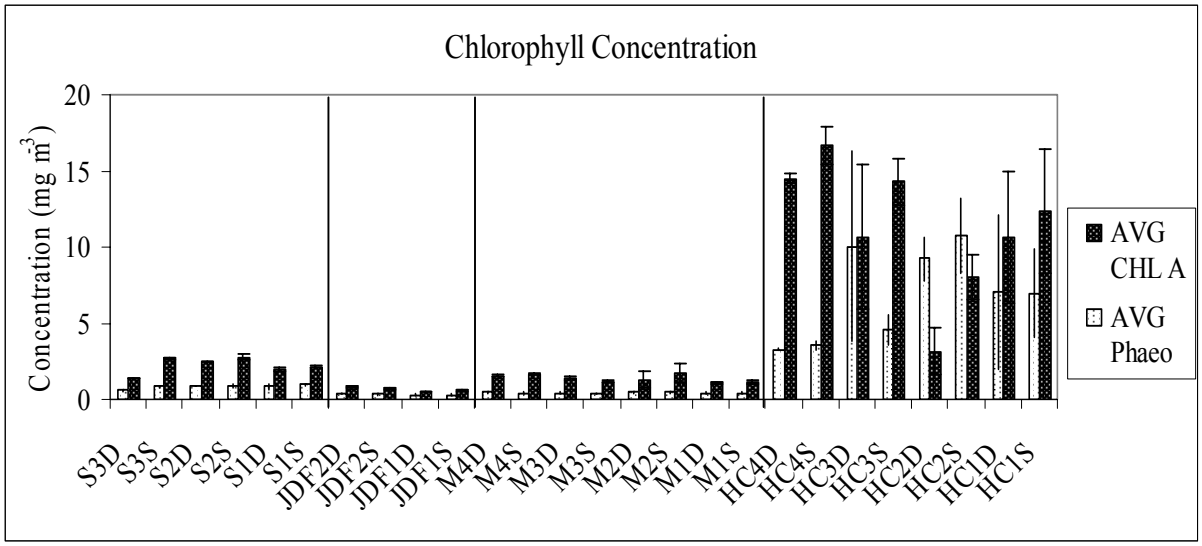

Figure 4: Autotrophic Cell Concentrations

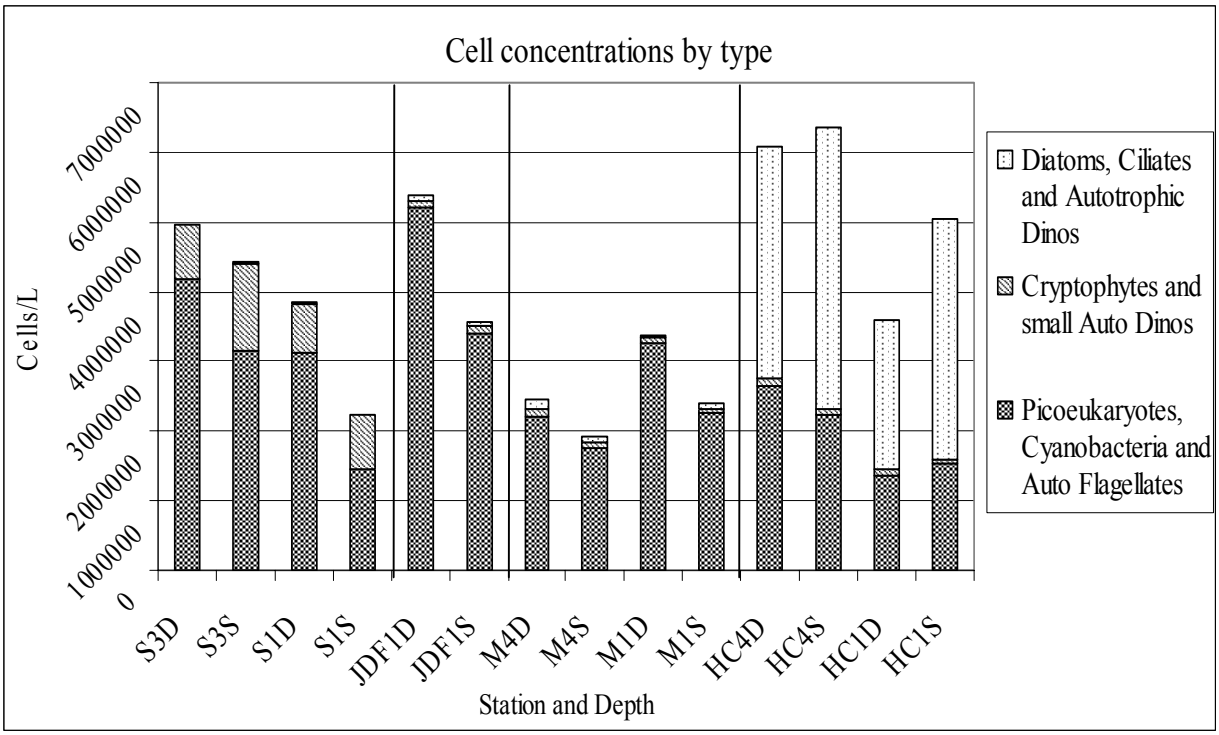

Figure 5: Average Light Intensity

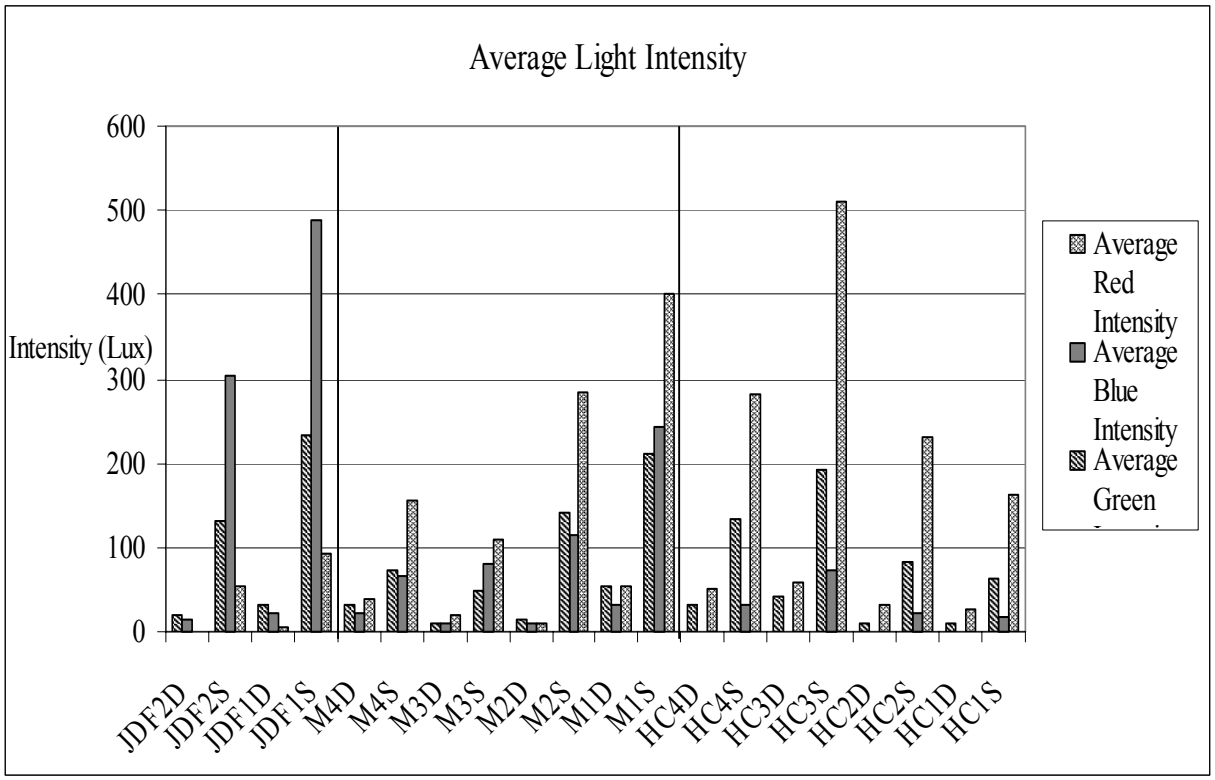

Figure 6: Autotrophic Cell Abundance

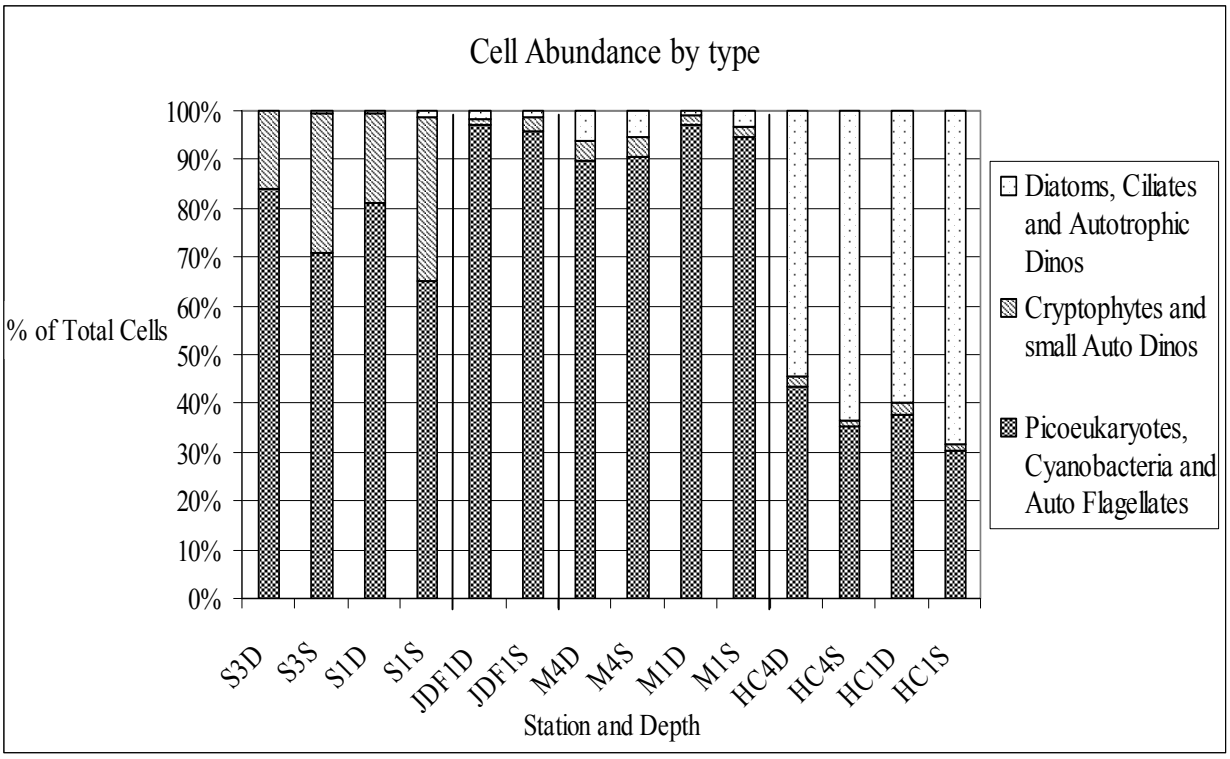

Figure 7: Comparison of BR Ration and Chlorophyll *a* Concentration

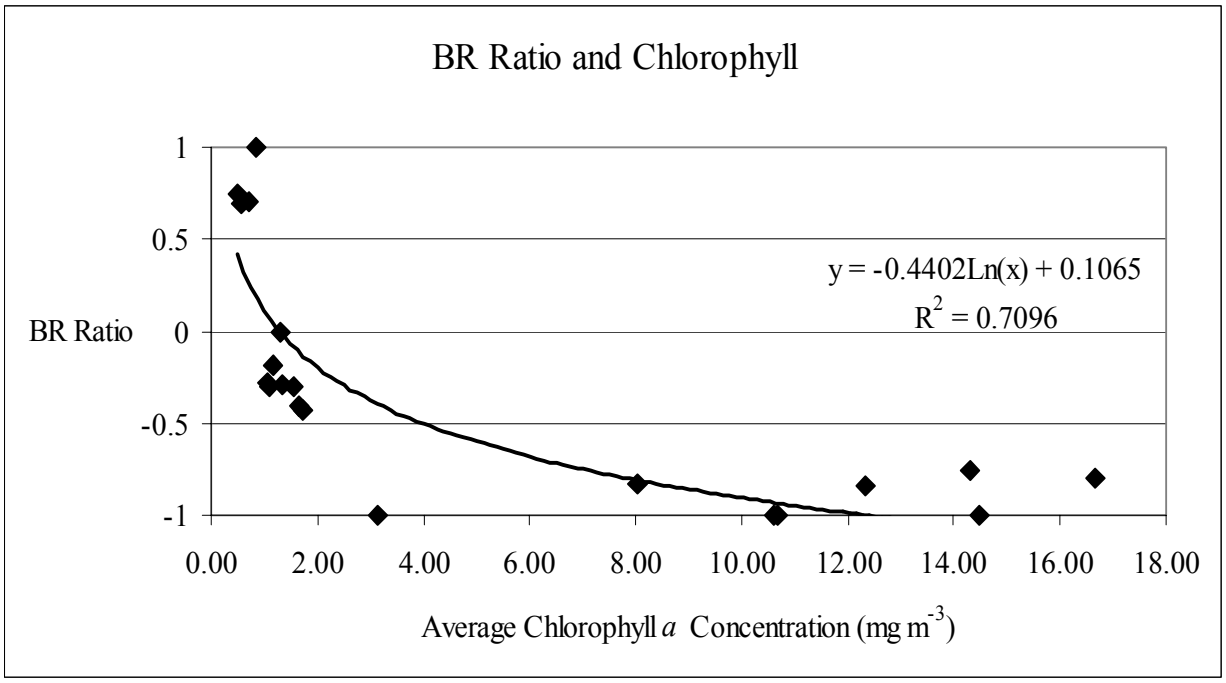

Figure 8: Comparison of BR Ratio and Cell Composition