

**Phytoplankton community structure in the Tahsis River and Zeballos
River plumes**

Senior Thesis
May 30, 2015

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Abstract

Rivers provide necessary nutrients for primary production in estuaries and along the coast. Salinity, light, temperature, and turbidity play a large role in shaping plume communities. By looking at the community structure throughout varying salinities in the Tahsis and Zeballos River plumes we can see the role salinity plays in shaping the plume community structure. Sampling throughout the plume allows for identification unique communities in the plumes changing salinity. Biomass at the mouth of both rivers was lower than at stations in higher salinities. This suggests that although nutrients are higher in the rivers, lower salinities are less conducive to phytoplankton growth. Nutrient input from rivers provides for a high biomass further out in the plume, where nutrients remain higher in the plume than in surrounding waters, and where turbidity has begun to decrease.

Introduction

River inputs can have a large impact on coastal ecosystems. In general, river plumes support a higher phytoplankton biomass than surrounding waters with nutrients, light, salinity, temperature, and turbidity playing a role in shaping the community composition (Goes et al. 2014). Unique communities develop throughout the plume as changing conditions allow for a greater species abundance and diversity (Clifford et al. 1991). The highest biomass is generally found in the intermediate salinities (15 to 25), where the nutrients remain high and the turbidity has decreased, allowing for the growth rate of both small and large species to be high (Dagg et al. 2004). In rivers with higher discharge rates, the turbidity levels are higher and the plume is larger, leading to the greatest biomass being further off shore (Novoa et al 2015).

Due to changing conditions throughout river plumes, species diversity and abundance are not constant. Chakraborty et al. (2015) looked at the community structure of a river influenced

region of the Gulf of Mexico. The study found that regions of higher freshwater were dominated by diatoms, cryptophytes, cyanobacteria, and chlorophytes, while the higher salinity marine waters were dominated by cyanobacteria, haptophytes, and prochlorophytes. Estuarine regions that fell in between the freshwater and marine water were dominated mostly by cyanobacteria and diatoms (Chakraborty et al. 2015).

A large volume output associated with the Fraser River creates a river plume in the Strait of Georgia in British Columbia. Research completed by Clifford et al. looked at the nutrient and phytoplankton dynamics of the river's plume. The study found that productivity was highest in the outer plume, and that the dominant species were different than the species that made up the bulk of the biomass. All nutrient concentrations were generally higher in the plume than in the river itself, except for silicate, suggesting that low river productivity was due to low nutrients levels, rather than light limitation. Because this study was conducted in late July, discharge from the Fraser River was at its low point, nutrients in the plume were supplied by entrainment as opposed to the river itself. (Clifford et al. 1991). This study provided the impetus for my research.

The vast diversity of phytoplankton species contributes to the development of unique niches; these niches play a large role in shaping phytoplankton communities. Communities in river plumes are often structured along a salinity gradient (Watanabe et al. 2013). This study looked at phytoplankton communities in the Tahsis River and Zeballos River plumes and describes how the phytoplankton communities vary throughout the plume, in relation to salinity and to one another. Biomass assessments, using an estimation of the organic carbon content of each species, was used to determine not only the most abundant species but also which species make up the greatest biomass in each plume. Because plumes support communities that may not

be able to exist without their presence, understanding the role plumes play in shaping phytoplankton communities is important. Understanding how the salinity gradient in the plume affects phytoplankton distribution allows us to gain a greater understanding of the role river plumes play in primary production.

Methods

The Tahsis River plume, in Tahsis Inlet, and Zeballos River plume, in Esperanza Inlet, were sampled between December 13th and 20th, 2014 with the use of a small boat launch off the R/V Thomas G. Thompson. The plumes were located using a YSI Data Sonde 556, which can read both temperature and salinity, which are needed to track the plumes. River plumes were defined as having a salinity below 30.

Once a plume was located three sampling sites were determined based upon salinity. Sites were target for surface salinities of zero, 15, and 25. See Table 1 for full site details. At each of the sites a 1L Niskin bottle was used to collect a water sample. The first sample was taken at the surface. The sample was collected at a depth that was selected by the point at which the secchi disk was no longer visible. The secchi disk provided an estimate for the depth to which light penetrates, because phytoplankton require light the secchi disk provides a mean to estimate where the phytoplankton are no longer able to survive. Water samples were used for nutrient, phytoplankton, and chlorophyll analysis. Phytoplankton samples taken were fixed with 2% formalin and stored for analysis back at the University of Washington.

On Board the Thompson

Immediately, upon returning on board the Thompson, chlorophyll samples were analyzed (Strickland and Parson, 1972). Each sample was extracted from 250mL to a volume of 10mL. The filters were placed in an ice bath for seven minutes while a sonicator probe was partially

submerged in the ice bath. Following at least a ten minutes rest in a cold dark room, the samples were placed in a centrifuge for five minutes. Samples were then read using a Turner TD-700 fluorometer, which was calibrated every 8 samples.

Post Cruise

Upon arrival back at the University of Washington nutrients samples were delivered to the Marine Chemistry Lab for analysis. Phytoplankton samples taken via Niskin bottles were analyzed using an inverted microscope. Each sample collected was approximately 800mL, the samples were settled and excess water was removed to reduce the sample volume to 50mL. They were then settled in 50mL sedimentation chambers to produce a 0.1mL Palmer-Maloney slide. The entire area of each slide was examined and species were identified, counted, and measured. The biomass of each species was determined using the equation from Strathmann (1967), which provides a method for estimating the organic carbon content of each cell. For each species cell dimensions were used to estimate mean cell volume. The cell volume was then used to estimate the organic carbon content of the cell.

Results

River Plume Salinity

In both the Tahsis and Zeballos River plumes, salinity readings in the surface layer were zero. However, salinity levels at depth in the rivers varied greatly. The Tahsis River had higher salinity at depth throughout the plume. In Esperanza Inlet other rivers in addition to the Zeballos River provided freshwater mid inlet (Figure 1 & 2).

Nutrients

In the Tahsis River plume surface levels of NH_4 generally increased away from the river mouth, but peaked at the second station with a maximum reading of $0.64 \mu\text{M}$ (microMolar) at the

surface. At depth NH_4 decreased away from the river mouth, reaching a minimum of $0.25 \mu\text{M}$ at the second station. In the Zeballos River NH_4 increased away from the river at the surface, but at depth generally decreased away from the river reaching a minimum at the second station. NH_4 in the Zeballos River at depth was almost five times the levels in the Tahsis River at depth. In the Tahsis River and Zeballos River plumes, NO_2 increased away from the river at surface and at depth. Initial surface readings of NO_2 in both rivers were zero.

In the Tahsis River plume NO_3 levels peaked at the second station, both at surface, and at depth. NO_3 reached a maximum of $10.13 \mu\text{M}$ at the surface, and at depth reached a maximum of $12.24 \mu\text{M}$. In the Zeballos River plume NO_3 generally increased away from the river with levels higher at depth than at the surface. At the surface NO_3 increased from $2.35 \mu\text{M}$ in the river to $6.34 \mu\text{M}$ at the last station, and at depth increased from $3.19 \mu\text{M}$ to $10.72 \mu\text{M}$, with a peak of $11.56 \mu\text{M}$ at the second station. In both river plumes the surface level of PO_4 in the rivers were zero and PO_4 levels in general increased away from the river mouth (Fig 3).

In both river plumes $\text{Si}(\text{OH})_4$ levels decreased away from the river, and were higher at the surface than at depth. In general $\text{Si}(\text{OH})_4$ levels were higher in the Tahsis River plume.

Chlorophyll

In the Tahsis River and the Zeballos River plumes chlorophyll levels increased away from the river. Chlorophyll levels in the Tahsis River plume were in general higher than those in the Zeballos River plume (Fig 4).

Phytoplankton Composition

Species composition varied greatly between stations and between depths. In the Tahsis River plume the greatest diversity was found at depth at the first station, where 17 different species were observed. In the Zeballos River plume the greatest diversity was observed at the surface at station two, and at the third station at depth. At both stations 12 species were observed.

The most common species seen throughout both plumes were *Chlamydomonas*. These diatoms dominated at depth at Tahsis River station three, making up almost 50% of the cells at the station. In both river plumes diatoms were common throughout all of the plume, with algae and flagellates increasing away from the river. In both plumes freshwater species can be found everywhere, however marine species dominated as salinity increased. (Fig. 5 & 6)

Abundance

In the Tahsis River plume, abundance at the surface peaked at the second station and abundance at depth peaked at the last station. In the Zeballos River plume, abundance increased away from the river with counts of phytoplankton being higher at the surface than at depth at each station. Between the two river plumes abundance was highest at depth, at station three, in the Tahsis River plume (Fig. 7).

Biomass

In the Tahsis River plume the biomass of phytoplankton at the surface decreased away from the river. At depth biomass generally decreased away from the river, but peaked at the final station reaching 0.2 ug C. This was the highest level found between all of the stations in both plumes. In the Zeballos River plume the biomass of phytoplankton increased outward from the river at both depths (Fig. 8).

Discussion

Multiple river sources in each inlet make both plumes complex. Stations further down the inlets have plumes comprised of multiple river sources, therefore combining nutrient sources. Previous research suggested that biomass would either be greatest in intermediate salinities, where both marine and freshwater species could survive, or would be highest in the outer plume where entrainment has brought nutrients up from depth (Clifford et al. 1991). In the Zeballos

River, the highest biomass was seen in the outer plume and is likely due to entrainment of deeper water. NO_3 , NO_2 , and PO_4 increased away from the river mouth, and the increase of NO_3 allowed for an increase in primary production. This dynamic suggest that entrainment and not the river is supporting life in the plume. Because of this biomass is likely to be greatest at plume boundaries and not in the center of the plume itself. When river discharge rates decrease entrainment is likely to increase as the plume spreads out more, creating more boundary regions where entrainment can occur. Seasonal changes in river discharge affect associated variable such as nutrient ratios, light, and stratification (Chakraborty et al. 2015). Seasonal changes likely lead to differences in community structure throughout the year.

The overall phytoplankton composition of both plumes was similar. Freshwater phytoplankton were found throughout both river plumes, strong currents from the outflowing river water dispersed the freshwater phytoplankton throughout the plume. Many freshwater phytoplankton have some form of osmoregulation that allows them to survive in freshwater; as they are pushed into higher salinities they are still able to survive; although growth rates, reproduction, and survival may be decreased. Because freshwater phytoplankton can survive in high salinities they were seen throughout the plume. Incoming tides likely mixed marine phytoplankton into low salinities. In low salinities marine phytoplankton cannot survive for long due to the fact that they lack efficient mechanisms for osmoregulation. In low salinities water continues to move into the body of the cell until it bursts (Heine-Fuster et al. 2010).

The change in community structure throughout the plume suggests that salinity plays a large role in shaping which phytoplankton were in the plume. Diatoms were the most diverse and dominant phytoplankton in both plumes. Research suggested that diatoms would be most prevalent in the lower salinities and then as salinity increased the community would move to

cyanobacteria, haptophytes, and prochlorophytes (Chakraborty et al. 2015). Diatoms were found to be the most abundant in the low salinities, but flagellates and algae were more prevalent in the higher salinities. Dinoflagellates were mostly marine species and were therefore found mostly in the high salinities, except for the genus *Euglena*. Species in the genus *Euglena* prefer high nitrogen waters and were therefore consequently seen in highest abundance at stations with NO_3 levels above $10\mu\text{M}$. Algal species were also more prevalent in higher salinities where PO_4 and nitrogen had increased. Because salinity plays such a large role in the plume, the community likely changes through out the year when discharge and rainfall decrease, leading to a more saline plume.

The highest biomass was found at depth of the third station, which was furthest from the river, in the Tahsis Inlet plume. More than half of the biomass at this station was from an alga in the genus *Chlamydomonas*. This genus is generally found in habitats that are rich in ammonium salt (John et al. 2002). In contrast to the findings of John et al. (2002) NH_4 at this station was not the highest in the plume. The highest NH_4 levels were seen at depth at the station located in the river mouth, however there was a very small population of the genus at this station.

Chlamydomonas prefer stagnant and low velocity waters; the high discharge rates in the river may have made the habitat unfavorable for a large population. In the Zeballos River plume the highest biomass was also seen at depth of the third station and was comprised mostly of *Chlamydomonas*. NH_4 at this station was not as high as it was in the Tahsis River and is likely why the population was not as large as in the Tahsis River plume.

The stations with the highest biomass in each plume were not necessarily the stations with the highest diversity. In the Tahsis River plume the greatest diversity was seen at depth at the first station, however biomass here was less than half that of the station with the highest

biomass. Incoming tides pushed high salinity water into the river mouth resulting in a higher salinity at depth. This allowed for marine phytoplankton to survive in the river. Turbulence in the river pushed freshwater phytoplankton to depth creating a diverse community composed of seventeen different genera of phytoplankton. Turbulence in rivers can often cause a decrease in biomass because phytoplankton are pushed out of the photic zone, however because the Tahsis River is only 2.5 meters deep, the phytoplankton were not pushed deep enough to leave the photic zone (Cloern et al. 1987). In the Zeballos River plume the greatest diversity was seen at two different stations. At both stations twelve different genera of phytoplankton were seen. Neither of these stations had the highest biomass, nor did they have unusually high nutrient levels. Research suggests that the greatest diversity in phytoplankton can be seen in regions of intermediate productivity levels (Vallina et al. 2014). Vallina et al. suggested that this community dynamic is due to selective feeding by predators. Findings in the Zeballos River plume support the theory that intermediate productivity supports higher biodiversity, however without studying other trophic levels it is unknown if selective predation is responsible for this dynamic.

Conclusion

In order to fully understand the role rivers and their plumes play in the structure of the phytoplankton community in Nootka Sound, more research needs to be done. River plumes should be studied during different seasons to detect changes in plume community dynamics throughout the year. Seasonal changes in river discharge, light, and nutrient availability could play a large role in shaping the phytoplankton community. Additional nutrients sources from multiple rivers can be beneficial in times of low nutrients, however they also increase the likelihood of eutrophication occurring (Pinckney et al. 2001). Eutrophication may lead to

harmful blooms that can cause fish kills and hypoxic conditions. Because phytoplankton represent the first trophic level in much of Nootka Sound, it is important to understand the factors that shape the community and therefore the higher trophic levels above.

Acknowledgements

I would like to thank Professors Julie Keister and Arthur Nowell, as well as Kathy Newell and my fellow classmate for all their support and input throughout the year. Your guidance and feedback have been paramount in my research this year.

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Figures

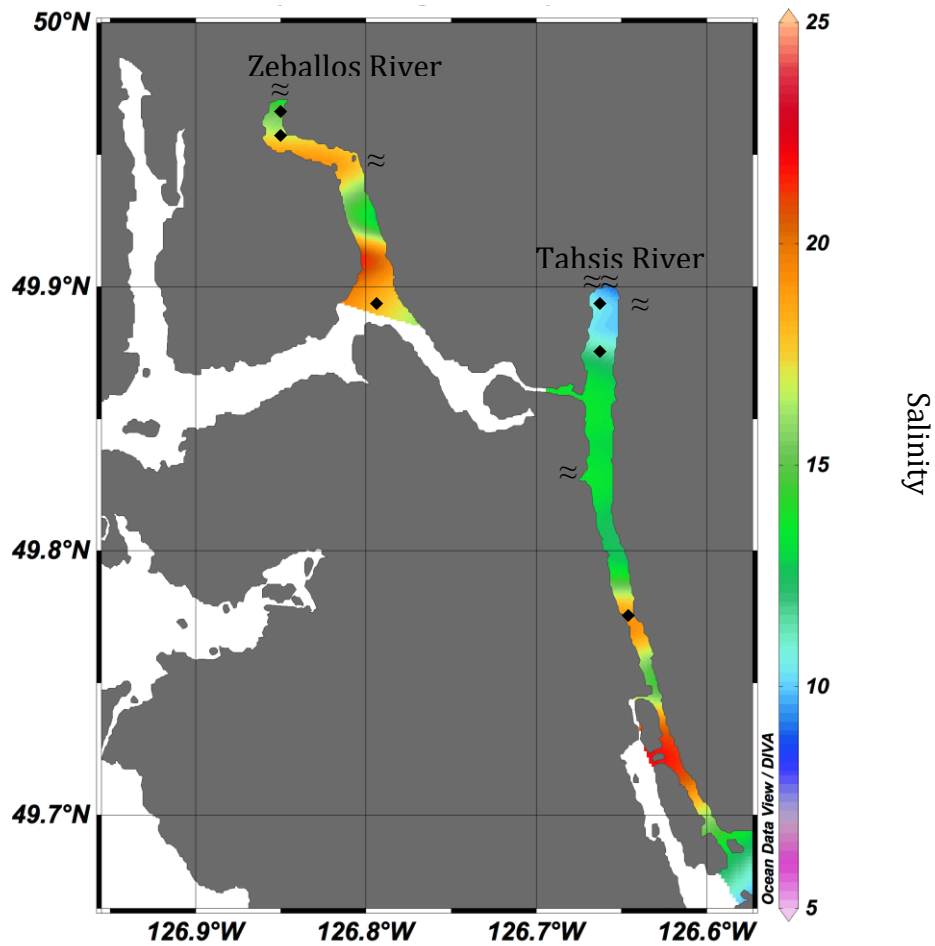


Figure 1. Surface salinity in Tahsis and Esperanza Inlets with rivers indicated by \approx , and stations indicated by \blacklozenge

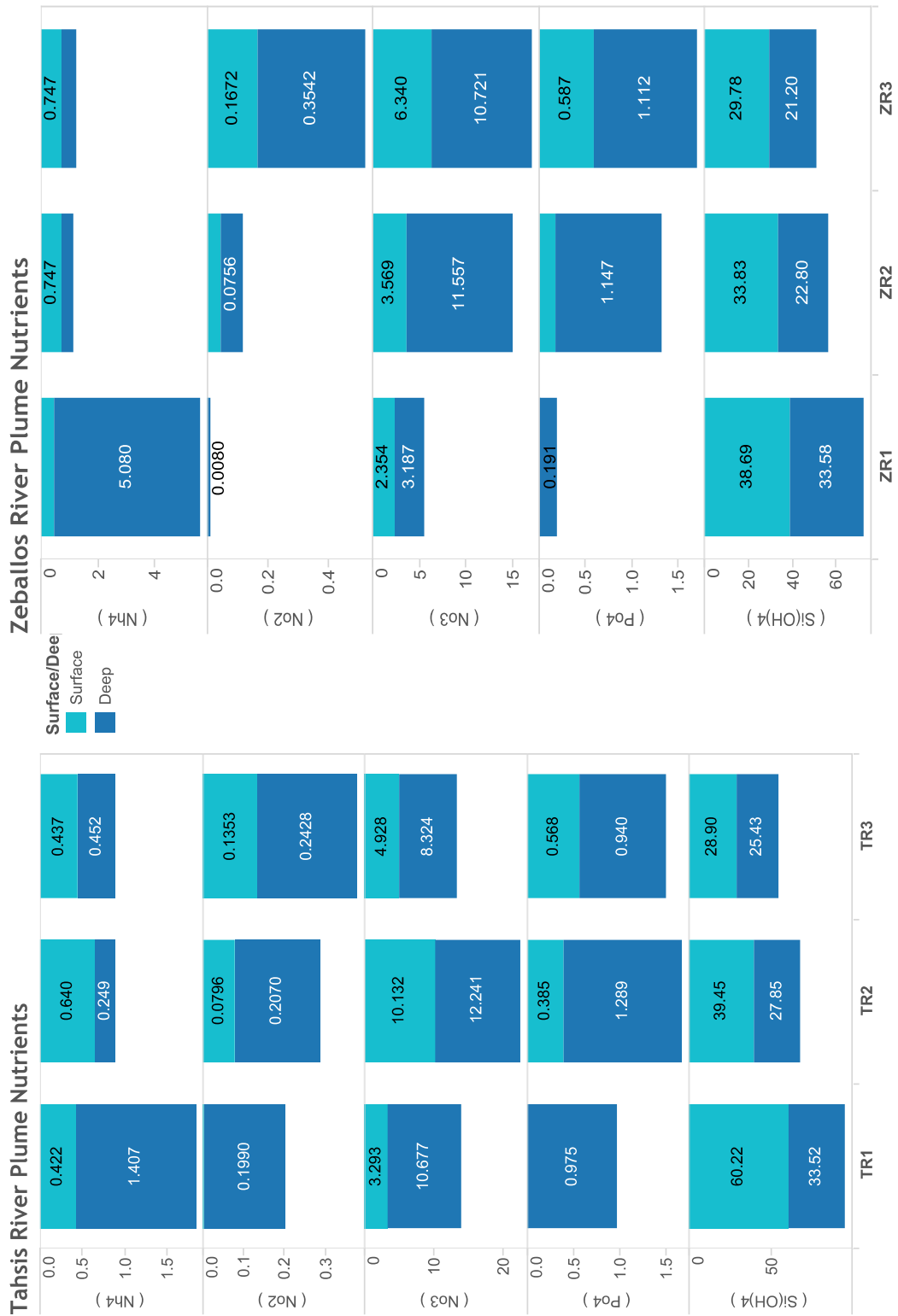


Figure 2. Nutrients concentration in uM in Tahsis and Zeballos River plumes taken from two separate depths.



Figure 3. Tahsis River phytoplankton composition in cells/Liter.

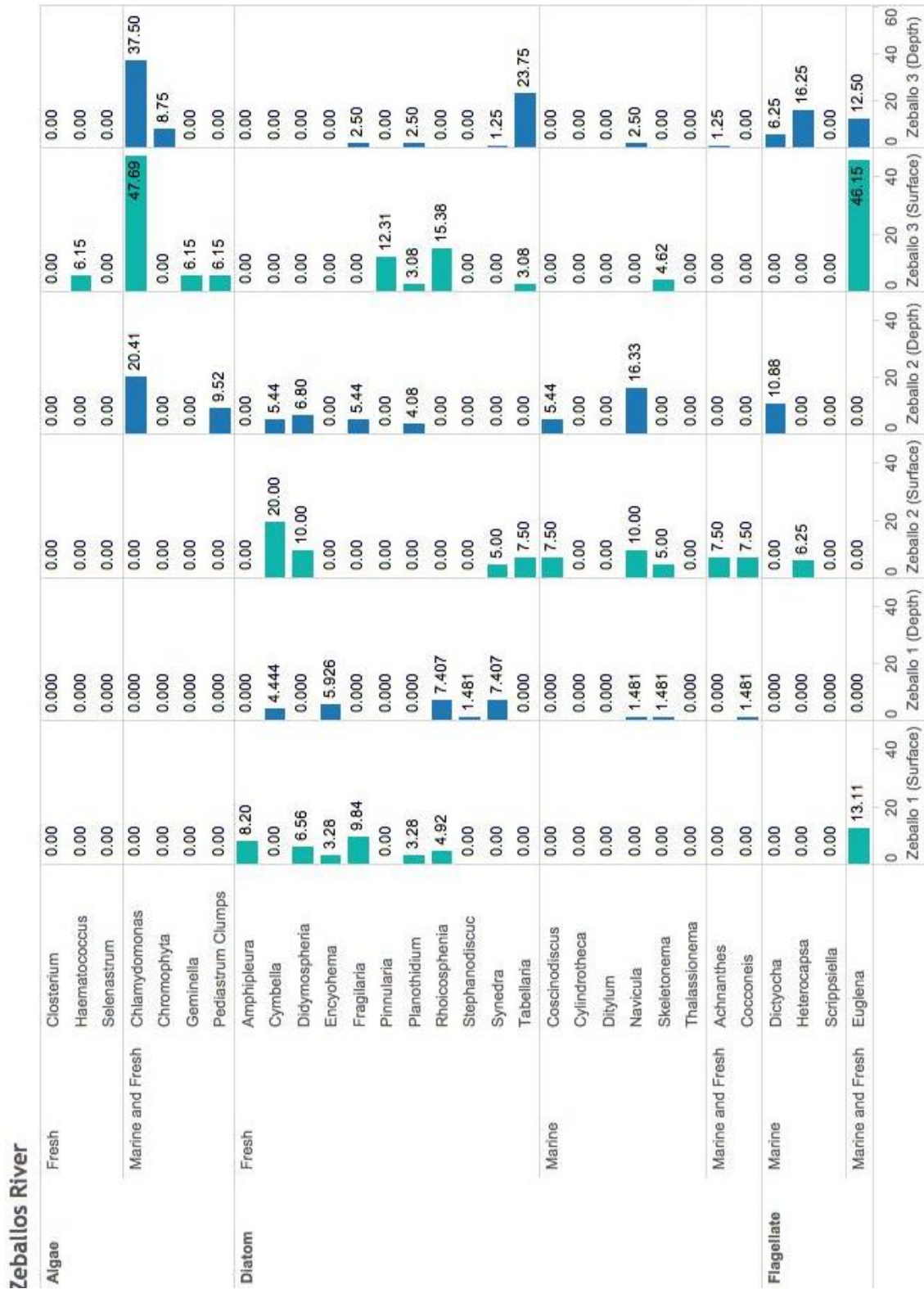


Figure 4. Zeballos River phytoplankton composition in cells/Liter.

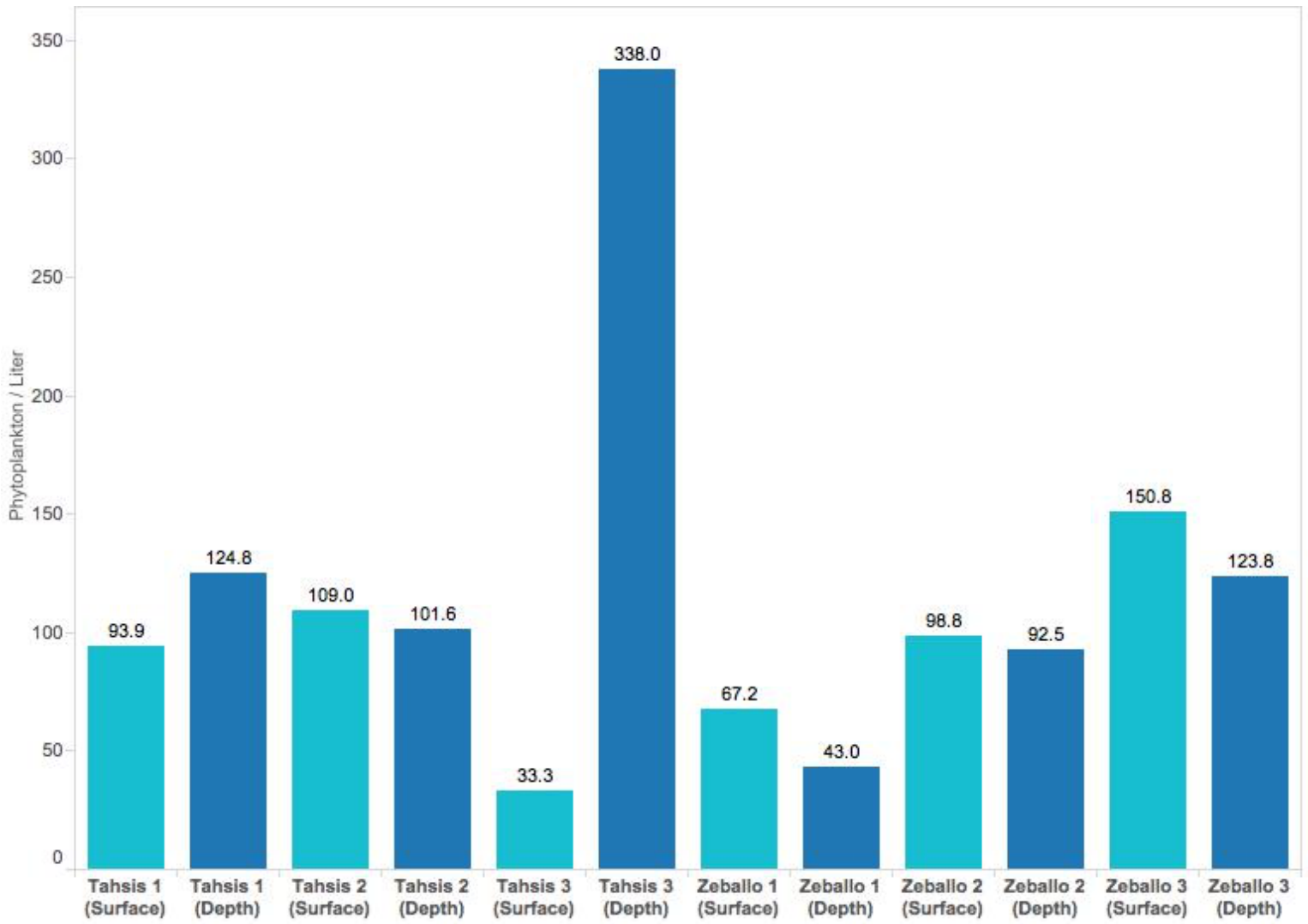


Figure 5. Abundance of phytoplankton per depth at each station.

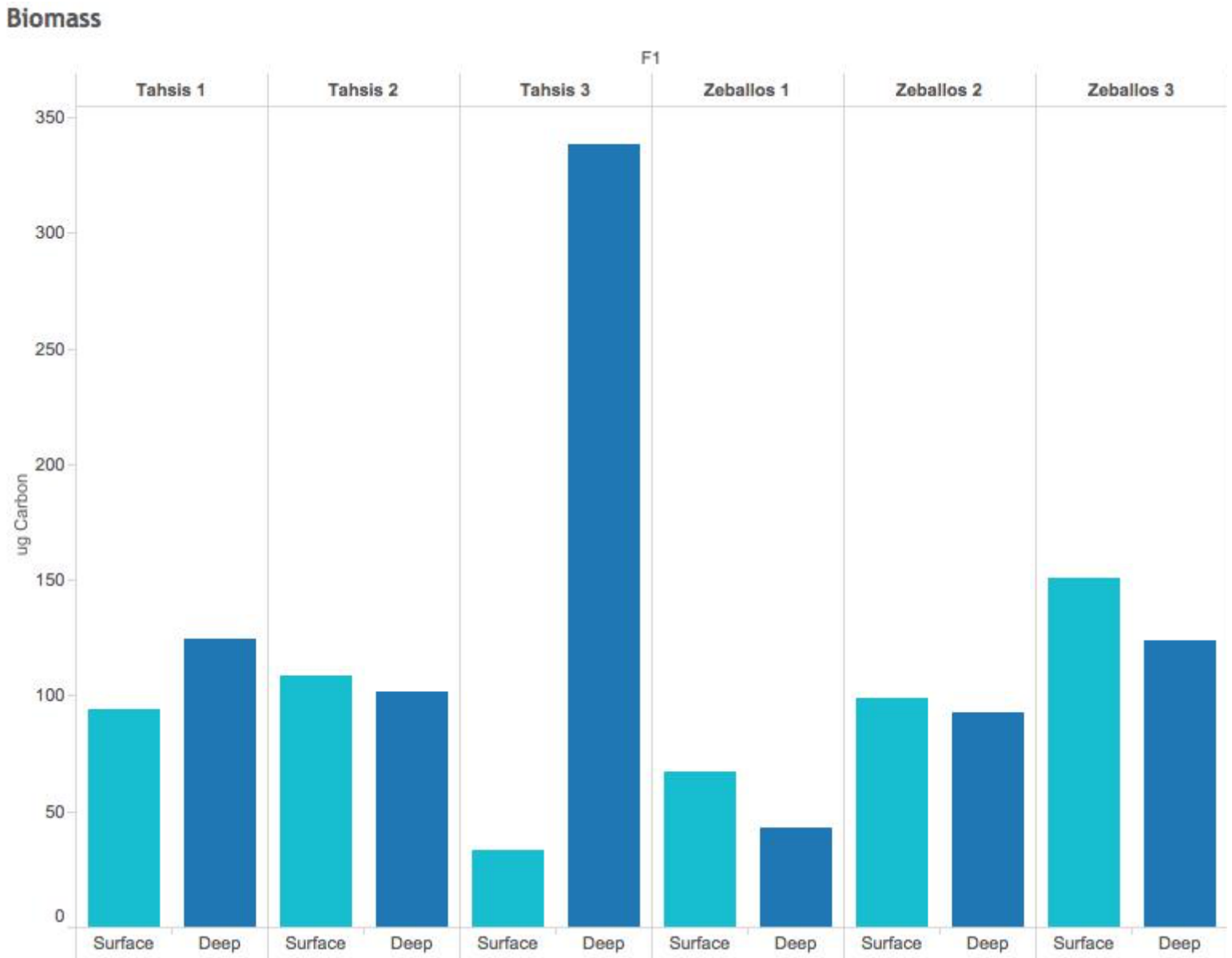


Figure 6. Biomass of phytoplankton at each station.

Table 1.

Station	Lat.	Long.	Depth (m)	Salinity
Tahsis 1	49° 55' 25.3"	126° 39' 30.9"	0	0.33
			2.5	26.77
Tahsis 2	49° 46' 10.3"	126° 39' 28.8"	0	8.1
			3.5	32.8
Tahsis 3	49° 46' 13.0"	126° 38' 32.7'	0	20.01
			8.5	26.9
Zeballos 1	49° 59' 04.1"	126° 50' 59.8"	0	0.18
			1	5.75
Zeballos 2	49° 58' 18.8"	126° 51' 06.0"	0	15.3
			3	33.2
Zeballos 3	49° 54' 03.0"	126° 44' 40.0"	0	18.96
			4	32.1

Table of river plume site details.