Analysis of zooplankton species composition and depth-based habitat preference in Nootka Sound, B.C., Canada

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31 May 2015

Abstract

The composition and abundances of zooplankton were studied in Nootka Sound, an estuarine fjord in British Columbia, Canada in winter 2014. Net tows were conducted at ten stations with varying water column depth and analyzed for composition, diversity and organism size. Diversity did not vary with depth, however, organism size showed a distinct and significant pattern with depth. A higher proportion of small copepods were found at stations with shallower depth. Depth is a factor in habitat preference, and is most likely associated with visibility to predators.

Introduction

Nootka Sound is an estuarine fjord located on the west coast of Vancouver Island, Canada. Little is known about the biological distributions or abundances of planktonic organisms in Nootka Sound. The biological processes of lower trophic level organisms such as phytoplankton and zooplankton are essential in energy transfers up to higher levels, which include fish and crustacean species. Several local Northwest Pacific fish species including Chinook salmon, coho salmon, chum salmon and pink salmon incorporate zooplankton species such as Calanoid copepods into their diets (Gardner 1982 and Bollens et al. 2010). Lower trophic organisms are also greatly important in the movement of carbon into the ocean and sediments, often referred to as the "Biological Pump" (Longhurst and Harrison 1989) through deposition of marine snow and fecal pellets (Turner 2002). Understanding what types of zooplankton are present in different environments is valuable information in terms of food web ecology and the efficiency of the biological pump marine and aquatic environments.

It is well documented that zooplankton species vertically migrate during different times of day or light conditions (Hays 2003), particularly to avoid predation (Cushing 1951, Hutchinson 1967, Lambert 1989). Different patterns of migration have been observed, including twilight migration (Cohen and Forward 2005, Valle-Levinson et al. In press) or reverse migration (Ohman et al. 1983, Cohen and Forward 2009). Vertical migrations permit zooplankton to feed during different conditions or avoid predation in surface waters when they are visible in daylight conditions (Lampert 1989). Some species undergo a more dramatic vertical migration than others (Hutchinson 1967), meaning they

will travel deeper in the water column to avoid predation or to follow prey, which also undergo vertical movement throughout the day.

Habitat preference can vary significantly based on species size, as swimming ability is a function of size. Larger zooplankton are more efficient in their movements and can inhabit waters with heavier currents. Water near the seafloor is often turbulent and can prevent certain species with limited swimming ability from aggregating there (Rothschild and Osborn 1988), or could play a factor in where zooplankton are advected. However, a large factor in habitat preference may be attributed to predator visibility, also a function of organism size. Larger zooplankters will be more visible than smaller organisms in shallow environments and thus more prone to predation, as "fish disproportionately consume larger individuals across their range sizes of prey" (Wellborn et al. 1996). Thus, deeper water could increase survivability by reducing risk of predation as a function of decreased predator visibility.

Biological analyses, specifically zooplankton identification in Nootka Sound, are extremely limited. However, identification has been conducted in Puget Sound, a fjord with a series of narrow interconnecting channels. Physical features in Puget Sound seem to have a strong effect on the geographic distribution of zooplankton (Allan 1976), and similar influences are expected in Nootka Sound based on bathymetric features like sills and river inputs (Pickard 1963). Water properties, such as salinity and temperature also play a key role in the habitat preference of zooplankton, as physiological factors such as osmoregulation are affected by these properties (Roddie et al. 1984). Both Muchalat Inlet and Tahsis Inlet of Nootka Sound experience the highest annual freshwater input from the Gold and Tahsis Rivers respectively, in addition to several small streams spread

throughout each Inlet (Pickard 1963), giving Nootka Sound a variety of different sub-regions with various surface salinities.

Abundances of zooplankton species will have seasonal variability (Colebrook 1982) due to effects of reproductive timing (Giles and Cordell 1998), changing seasonal water properties or light conditions (Herman 1998). Evolutionary strategies, such as diapause in copepods can also affect abundances, as those organisms undergoing a dormant period will inhabit deep or buoyantly balanced water depths (Hairston and Munns 1993).

Abundant throughout coastal waters near Washington in Puget Sound, the Straight of Juan de Fuca and the Straight of Georgia, are various copepods, including *Paracalanus, Pseudocalanus, Microcalanus, Aetideus, Acartia, Euchaetae, Metridia, Oithona, Corycaeus and Calanus* (B.W. Frost, unpubl.) Nootka Sound is a geographically and climatically similar estuarine region and similar species are expected to inhabit both Puget Sound and Nootka Sound. Diversity and richness are often used to describe the ecology of an environment. The Shannon-Wiener index is a commonly used tool to quantify both diversity and richness and will be used in the biological analysis of zooplankton in Nootka Sound. I aim to identify what species are present in different regions of the sound, as well as examine population diversity and organism size. Different depths are hypothesized to exhibit varying diversity and organism size composition.

Methods

Field Methods

Zooplankton samples were collected aboard the R/V *Thomas G. Thompson* on cruise TN314 from 11-21 December 2014, in Nootka Sound, British Columbia, Canada. Sample stations (Figure 1) were chosen to incorporate a wide range of water column depths and water properties such as surface salinity and turbidity. Four stations selected in Muchalat Inlet ranged from 74-380 m depth. Five stations selected in Tahsis had depth ranges from 35-181m. One station with high oceanic input was selected and had a depth of 151 m. At all stations, a half-meter diameter net vertical tow from five meters from the seafloor to the surface was used to collect zooplankton while the vessel was stationary. The net apparatus was equipped with a 211-micron mesh net, flow meter and Sensus depth probe. Winch speed was kept at 30 meters per minute at all stations for both descent and ascent of the net. Station BB01 was sampled from the R/V *Weelander* and the net was manually pulled through the water column.

Samples were collected in 700 mL polypropylene jars and filled with 5% buffered formalin for preservation. Two surface water samples were taken at each station and analyzed for chlorophyll, as a proxy for primary productivity and phytoplankton following the procedure outlined in Yentsch and Menzel (1963). An average value of the two samples at each station were used.

Laboratory Methods

Zooplankton samples were filtered and diluted to 100 mL, then subsampled with a 1 mL syringe. Zooplankton were counted using a dissecting scope and identified to genus level for most organisms or taxonomic group (i.e. amphipod or krill). Large, rare organisms that did not appear in subsamples were removed from station sample jar,

identified, counted and measured. Organism density (individuals m^{-3}) was calculated using:

$$\frac{V * x}{0.031 * f * s}$$

where *V* is the diluted volume, *x* is the number of individuals in the subsample, 0.031 is the flow meter calibration factor, *f* is the flow meter reading and *s* is the total subsample volume.

Shannon-Wiener Index values were calculated for each station using the standard Shannon-Wiener methodology

$$H' = -\sum_{i=1}^{n} P_i ln P_i$$

where H' is the diversity at a given station, *n* is the number of species and P_i is the proportion belonging to the *i*th species.

Biomass estimates were calculated for each species based on organism length (Table 2). The largest and smallest organism in each taxonomic group was measured and a midpoint length was used as the average in biomass calculations.

For size based analysis, small organisms were defined by genus or group as those equal to or less than 2 mm in length. These groups included *Oithona*, *Microcalanus*, *Paracalanus*, *Corycaeus* and *Acartia.* Though they fit the sizing criteria to be included in the category, gastropod larvae, ostracods, pteropods, isopods and bivalves were not included in small organism analysis.

Results

The two stations with the highest zooplankton densities were both located in Tahsis Inlet with >1000 organisms m^{-3}; the two stations with the lowest abundances were both located in Muchalat Inlet, with < 330 organisms m^{-1} (Table 2). Deeper stations (>150 m) contained more organisms than shallow ones. Organism densities decreased with station depth (Fig. 2A), while total water column abundance increased as station depth increased (Fig. 2B).

The copepods *Acartia, Calanus, Metridia, Paracalanus* and *Oithona* were present at every sampling station, though abundance at each station varied considerably. The most abundant organism at every station was *Oithona,* a small copepod ranging between 0.5-1 mm, while several organisms were only present at one station. These instances include a single crab megalops (*Cancer*) found at station M13, an individual squid found at station M06, one organism from the order *Isopoda* caught at station T01, and one nectophore, bivalve and pteropod at station BB01. Station M10 was the only station to have polychaete worms present. Other rare organisms only present at three or fewer stations were jellies and ostracods.

N08 had the highest biomass per cubic meter, mostly from a single large krill (Fig. 3, Table 3). Station M01 had the lowest biomass per cubic meter, and also the second lowest abundance of the stations. Biomass did not correlate with depth (data not shown).

Diversity did not vary significantly (r = 0.260, p = 0.469) with depth (Fig. 4). However, organism size played a significant (r =0.814, p = 0.004) role in habitat preference based on depth (Fig. 5).

Surface chlorophyll values were very low, between 0.06-1.78 μg L^{-1}, and did not correlate with density, with the exception of station T18 which had the highest density and highest surface chlorophyll (Table 4). All stations in Muchalat had values less than or equal to those in Tahsis Inlet.

Discussion

Perhaps the most important finding and distinction made in this study is the difference between diversity and size difference among different species. The Shannon-Wiener diversity index used in this study takes into account two quantifiable measures, richness and evenness. A value of 0 indicates only one group present, while a higher value indicates that many groups are present and are evenly distributed. Across the ten stations, richness ranged from 10-14 different organisms, while evenness was greatly variable as some stations included identifications with only one organism from that group, or several groups with significantly larger abundance than others. The downfall of the Shannon-Wiener index in this study is that speciation has a greater effect on the quantifiable result than grouping similar organisms based on function or behavior, such as large copepods that are avoiding shallow waters. The results of the size analysis have shown that there are different populations in habitats with varying depths. There were more small copepods inhabiting shallow water and more large copepods found in areas with greater water depth (Fig. 5), possibly due to avoidance of predators. Habitat preference can be influenced by predation pressure (Reynolds et al. 2002). Small copepods like *Oithona*, *Acartia*, *Microcalanus* and *Paracalanus* have higher survivability

than larger copepods such as *Calanus, Metridia* and *Paraeuchaeta* in shallower water, and therefore have higher population proportions in these environments (Fig. 5).

It has been previously suggested that *Oithona* is the most important and most abundant copepod in all of the oceans (Bigelow 1926). *Oithona* was the most abundant organism sampled at all ten sampling stations. Perhaps one reason is that cyclopoid copepods such as *Oithona* are known to graze on heterotrophic protists, ciliates and heterotrophic dinoflagellates, more so than larger calanoid copepod species do (Turner 2004). It is unknown from this study the abundance of protists, but it can be assumed that more prey is available to cyclopoids and could be a reason for high *Oithona* abundance. Low abundance of larger copepods could be due to decreased solar radiation in the wintertime, which can limit phytoplankton growth. Low surface chlorophyll (8 stations $<0.15\ \mu g\ L^{-1}$) (Table 4) indicate low prey availability for most larger copepod species that graze on larger autotrophic phytoplankton.

All Muchalat Inlet stations had extremely low surface chlorophyll (Table 4). Visual observations aboard the cruise described high sediment output from the Gold River, which would effectively reduce light penetration in the water column and limit phytoplankton growth. In addition, *Oithona* are documented as euryhaline and eurythermal (Turner 2004) and seem to tolerate a large range of environments.

Though *Oithona* was the most abundant organism, actual abundances may be higher than reported values from this study. A 211-micron mesh net was used to sample at all locations. However, many studies have found nets with mesh sizes 200-333-micron greatly underestimate small copepod (*Oithona, Microcalanus, Paracalanus, Acartia*) compared to studies that utilize a 100-micron net (Gallienne and Robins 2001, Turner

2004). Given this understanding, biomass estimates of small copepod species may have been underestimated. Large zooplankton may also have been underestimated. Larger zooplankton with superior swimming capabilities will avoid the net and fewer will be caught than is representative of the true composition (Clutter and Anruka 1968).

Stations were selected to cover a broad range of depths, between 35-380 m. Temperatures did not vary from station to station (data not shown), and salinity ranges were similar between stations (Table 4). This is surprising, particularly at stations that seemed to have influence from freshwater input from the Gold River in Muchalat Inlet (Stations M01, M06), the Tahsis River (Stations T01 and T02) and the Tsowwin River in Tahsis Inlet (Station T18), based on proximity and the highest annual freshwater input from these sources during the sampling period (Pickard 1963). However, salinity values were measured using the CTD and surface salinities (depth < 3 meters) were unavailable. It is expected that the freshwater layer is thinner than 3 meters and could potentially be preferential to organisms who select for less saline waters. Surface salinity was only available at station BB01, where three identification groups were found exclusively. Surface salinities were unknown at the other stations, but perhaps these organisms (nectophore, bivalve, pteropod) have selected for fresher water, though further speciation is required to analyze salinity preference.

Light transmission showed no correlation to abundance (data not shown). Station M06, which had the lowest abundance, showed a high light transmission throughout the water column, between 91-96%. Significant light penetration would provide exposure to predators. However, a significant pattern was not observed across all stations. Station

T02 showed a similar light transmission range of 90-95% throughout the water column, but high abundance was measured there.

Conclusions

The results of this study can be used to predict where higher trophic level organisms, such as salmon, will find food, which is a large factor in their habitat preferences. Ultimately, depth encompasses many factors in habitat preference, including salinity, temperature, predator and prey abundance, light transmission and current advection, as many of these properties show significant vertical patterns in the water column. Further study of Nootka Sound should take place during different times of the year, to analyze how temporal patterns affect composition and diversity at different habitats.

Acknowledgements

I would like to thank my mentor, Dr. Julie Keister for her help in idea development, laboratory usage and zooplankton identification training. I would also like to thank BethElee Herman and Amanda Winans fro the Keister Lab for further help in zooplankton identification, and Arthur Nowell for draft revisions. Lastly, thank you to fellow students aboard cruise TN316 who helped collect samples.

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Figures and Tables

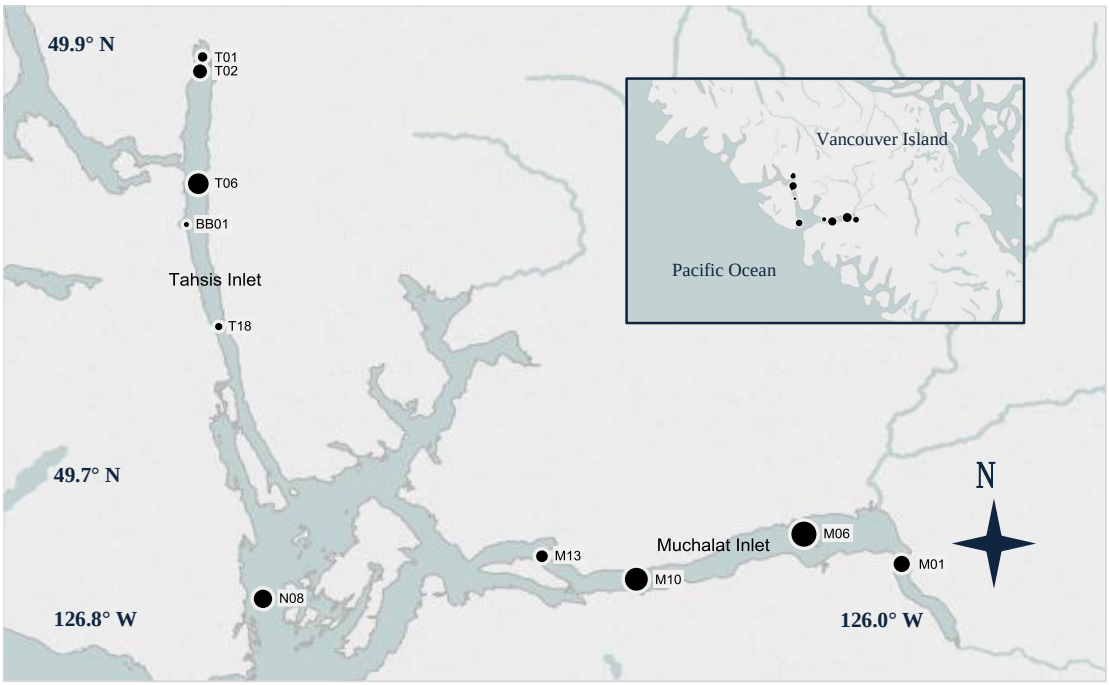

Fig. 1. Station map of sampling sites in Nootka Sound, B.C., Canada. Size of station mark is representative of water column depth at that station.

Table 1. Sampling station names, depths and locations from cruise TN316 aboard the R/V *Thomas G. Thompson* December 11-21, 2014.

Name	Depth (m)	Location
M01	123	49.65° N 126.09° W
M06	380	49.67° N 126.17° W
M10	265	49.64° N 126. 31° W
M13	74	49.66° N 126.38° W
T01	59	49.91° N 126.66° W
T02	93	49.91° N 126.66° W

T06	181	49.85° N 126.66° W
T18	49	49.78° N 126.65° W
N08	151	49.63° N 126.61° W
BB01	35	49.83° N 126.67° W

Table 2. Organism carbon or dry weight calculations equations based on organism length (L).

Identification	Biomass or Dry Weight Equation (μg or μg C)	Source
Copepoda		
Acartia spp.	Log C = 3.03 * Log L - 8.52	(Uye 1982)
Calanus spp.	Log C = 2.64 * Log L - 7.00	(Uye 1982)
Metridia spp.	$DW = 4.13 * L^{3.28}$	(Coyle)
Corycaeus spp.	Ln DW = 1.7 * Ln L - 9.92	(Clarke & Roff 1990)
Paraeuchaeta spp.	Log C = 2.45 * Log L - 6.25	(Uye 1982)
Oithona spp.	Log C = 1.45 * Log L -4.25	(Uye 1982)
Paracalanus spp.	Ln DW = 2.78 * Ln L - 16.52	(Webber and Roff 1995)
Microcalanus spp.	Ln DW = 2.78 * Ln L - 16.52	(Webber and Roff 1995)
Other		
Pteropod/ Gastropod Larvae	Ln DW= 0.969 + 0.529 Ln L	(Collins 1992)
Ostracod	Log C = 4.15 L -11.15	(Uye 1982)
Polychaete worm	1500 μg	Keister, personal ref.

Krill	Log C = 3.174 * Log L - 0.473	(Ross 1982)
Amphipod	Ln DW = 2.4614 * Ln L -5.051	(Williams and Robins 1979)
Chaetognath	Log C = 3.16 * Log L -1.29	(Uye 1982)
Larvacean	Log DW = 1.7812 * Log L - 3.6034	(Gorsky 1987)
Isopoda spp.	$DW = 0.011 * L^{2.68}$	(Strong 1979)
Nectophore	$C (\mu g) = 20.47 * L^{0.834}$	Keister, personal ref.
Cancer (megalops)	DW = 1300	(Sulkin and McKeen 1994)
Hydromedusae/ Squid	$C (\mu g) = 1.8885 * L^{2.619}$	(Takahashi and Ikeda 2006)

Table 3. Abundances of zooplankton and other taxa caught in nets at the ten sample stations, given in number of organisms per cubic meter. Total abundances at each station are also given.

	Station									
Identification	**M01**	**M06**	**M10**	**M13**	**T01**	**T02**	**T06**	**T18**	**BB01**	**N08**
Copepoda										
Acartia spp.	19	17	4	41	44	90	64	40	18	78
Calanus spp.	14	58	96	58	10	130	121	24	95	101
Corycaeus spp.	-	2	8	8	20	41	7	64	18	23
Metridia spp.	6	34	139	58	10	16	20	8	7	69
Microcalanus spp.	8	14	28	347	171	187	10	160	-	-
Oithona spp.	226	92	142	356	210	342	158	664	133	485
Paracalanus spp.	14	2	50	58	103	155	60	88	95	18
Paraeuchaeta spp.	1	22	-	-	-	8	3	-	-	5
Other										
Amphipod	-	-	1	8	-	-	10	8	7	14
Chaetognath	1	14	8	18	5	1	8	16	-	-
Gastropod Larvae	30	-	-	-	88	-	3	-	14	-
Krill	1	2	5	-	-	1	17	-	-	1
Larvacean	6	-	-	8	20	57	-	8	4	-
Nectaphore	-	-	-	-	-	-	-	-	1	-
Pteropoda spp.	-	-	-	-	-	-	-	-	1	-
Crab megalops	-	-	-	1	-	-	-	-	-	-
Polychaete	-	-	4	-	-	-	-	-	-	-
Ostracoda spp.	1	-	4	-	-	-	-	-	-	-
Squid	-	1	-	-	-	-	-	-	-	-
Isopoda spp.	-	-	-	-	1	-	-	-	-	-
Bivalve	-	-	-	-	-	-	-	-	1	-
Jelly	1	-	-	-	-	1	-	-	-	1
Total	**328**	**258**	**489**	**961**	**682**	**1028**	**481**	**1080**	**394**	**795**

Table 4. Biomass (μg C/m^3 or dry weight/m^3) of zooplankton at the ten sample stations. Total biomass for each station is also given.

	Station									
Identification	**M01**	**M06**	**M10**	**M13**	**T01**	**T02**	**T06**	**T18**	**BB01**	**N08**
Copepoda										
Acartia spp.	187.7	156.7	37.2	397.7	433.6	834.4	777.0	391.5	165.3	733.3
Calanus spp.	2895.9	6760.5	17255.6	13152.5	1611.8	27401.2	27422.4	4305.8	18411.6	22867.1
Corycaeus	-	98.9	301.9	330.8	738.2	1612.1	267.0	2510.7	707.4	906.3
Metridia spp.	1384.3	9316.6	23405.3	14560.1	1482.6	3374.8	3771.0	1659.6	1763.5	17259.7
Microcalanus spp.	822.7	1486.2	-	34611.6	16404.6	17950.5	1046.1	15950.0	-	-
Oithona	18272	6981.7	11531.1	28795.0	15385.3	28440.5	13317.3	48635.5	10992.4	40355.4
Paracalanus spp.	1596.2	281.5	5558.6	6431.3	11400.9	17176.1	7036.5	9628.1	10798.3	1965.9
Paraeuchaeta spp.	399.3	8932.7	-	-	-	3447.5	1775.1	-	-	2053.3
Other										
Amphipod	-	-	73.3	561.5	-	-	515.3	945.5	2003.6	3021.8
Chaetognath	24.2	1715.3	5774.4	3057.3	179.0	601.8	4003.5	1603.9	-	-
Gastropod Larvae	84.8	-	-	-	611.1	-	9.4	-	39.3	-
Jelly	71.3	-	-	-	-	102.8	-	-	-	1.2
Krill	3964.6	29884	41412.1	-	-	90368.6	67619.0	-	-	131790
Larvacean	1282.0	-	-	1858.4	4471.7	13836.3	-	1883.6	825.1	-
Nectaphore	-	-	-	-	-	-	-	-	195.9	-
Pteropoda spp.	-	-	-	-	-	-	-	-	6.3	-
Crab megalops	-	-	-	1300.0	-	-	-	-	-	-
Polychaete	-	-	6000.0	-	-	-	-	-	-	-
Ostracoda spp.	0.3	-	1.2	-	-	-	-	-	-	-
Squid	-	114.9	-	-	-	-	-	-	-	-
Isopoda spp.	-	-	-	-	19.0	-	-	-	-	-
Bivalve	-	-	-	-	-	-	-	-	25.2	-
Total	**30985.8**	**65729**	**111350.7**	**105056.2**	**52737.8**	**205146.6**	**127559.6**	**87514.2**	**45933.9**	**220954**

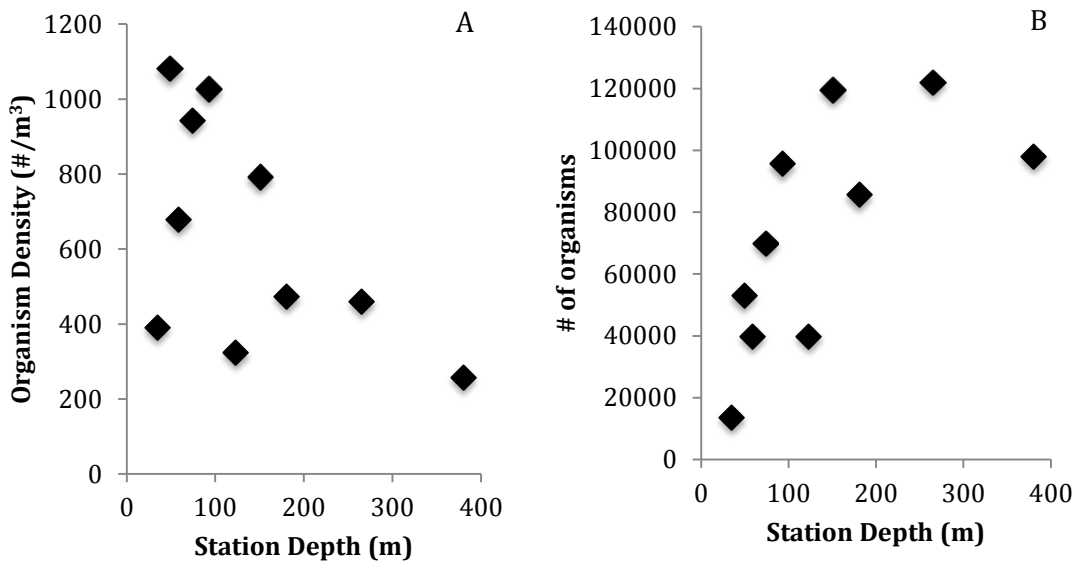

Fig. 2. A) Organism density (individuals m^{-3}) versus station depth, and B) total abundance versus station depth.

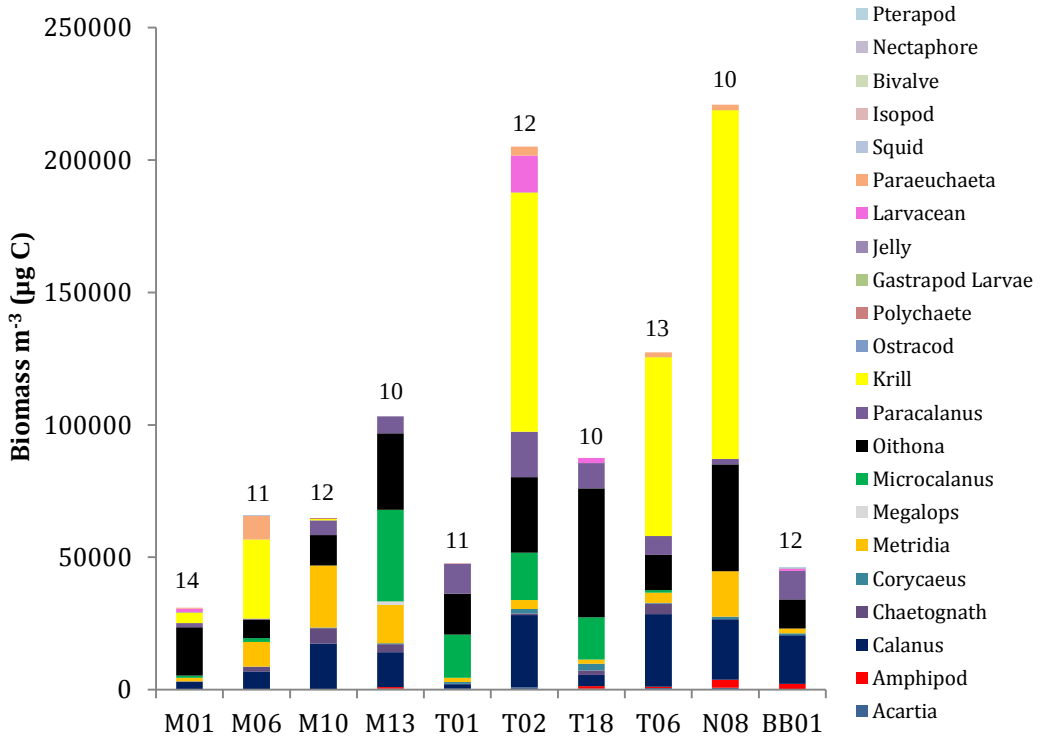

Fig. 3. Biomass distribution for each station (in µg C m^{-3}). Each organism genus or group is denoted by a different color indicated in the figure legend. Species richness is indicated above the bar for each station.

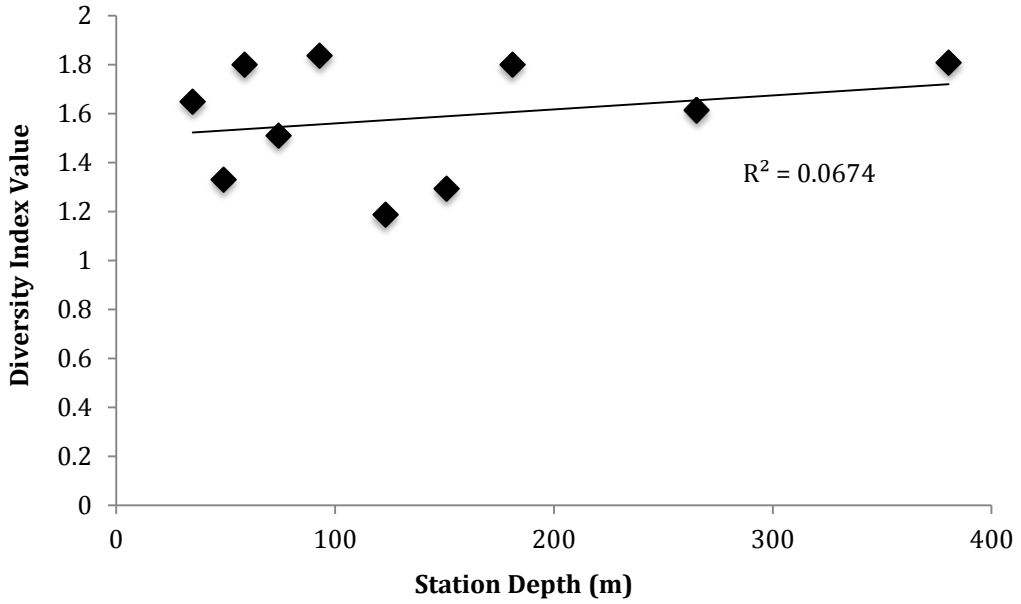

Fig. 4. Station depth plotted against Shannon-Wiener diversity index value at each station. A line of best fit with corresponding r value is given to show fit.

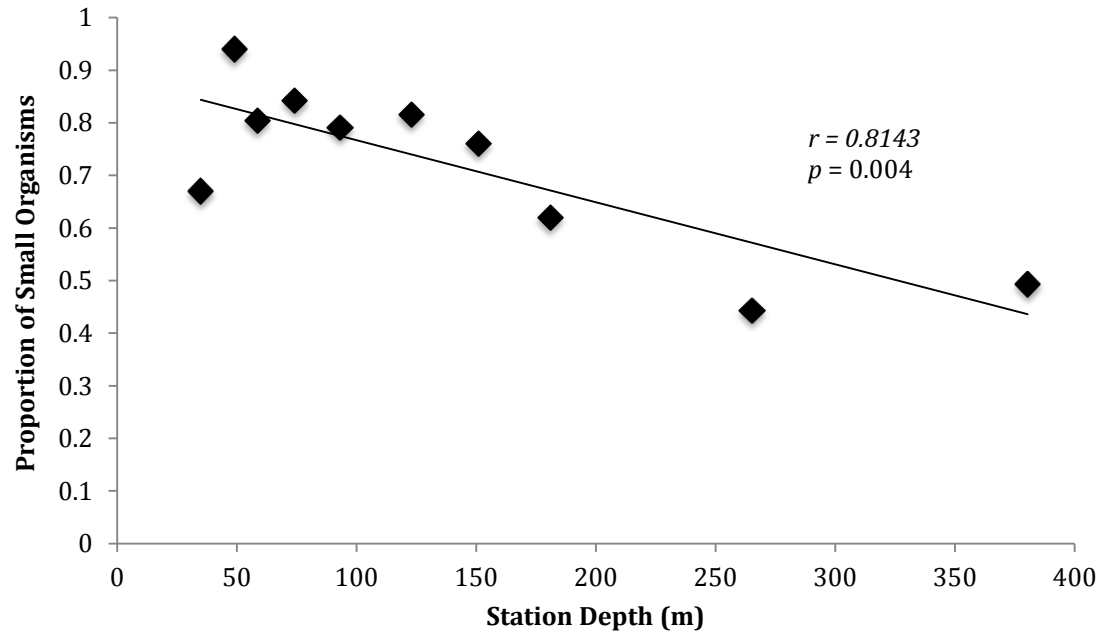

Fig. 5. Station depth plotted against the proportion of small organisms (genus or group less than 2 mm in length). A line of best fit is provided with the correlation coefficient (r).

Table 4. Average surface chlorophyll (μg L^{-1}) as a proxy for primary productivity. Surface chlorophyll was not measured at stations T01 and N08. Salinity range is given at maximum and minimum depth where data were available at each station.

Station	**Avg. Surface Chlorophyll (μg L^{-1})**	**Salinity range (PSU)**
M01	0.10	29.6 – 32.8
M06	0.06	29.6 – 32.6
M10	0.07	29.8 – 33.0
M13	0.07	29.0 – 32.6
T01	-	28.1 - 31.8
T02	0.10	28.3 - 32.1
T06	1.55	28.3 – 32.2
T18	1.78	-
BB01	0.16	10.9 - 34.9
N08	-	28.2 - 32.3