The response of crustacean zooplankton production to variations in food quantity, quality, and primary production in coastal marine ecosystems

by

Karyn Dawn Suchy
B.Sc., University of Manitoba, 2003
M.Sc., University of Manitoba, 2006

A Dissertation Submitted in Partial Fulfillment
of the Requirements for the Degree of

DOCTOR OF PHILOSOPHY

in the Department of Biology

Supervisory Committee

The response of crustacean zooplankton production to variations in food quantity, quality, and primary production in coastal marine ecosystems

by

Karyn Dawn Suchy
B.Sc., University of Manitoba, 2003
M.Sc., University of Manitoba, 2006

Supervisory Committee

Dr. John F. Dower, Department of Biology
Supervisor

Dr. Steve J. Perlman, Department of Biology
Departmental Member

Dr. Diana E. Varela, Department of Biology
Departmental Member

Dr. Debby Ianson, School of Earth and Ocean Sciences
Outside Member

Abstract

Supervisory Committee
Dr. John F. Dower, Department of Biology
Supervisor

Dr. Steve J. Perlman, Department of Biology
Co-Supervisor or Departmental Member

Dr. Diana E. Varela, Department of Biology
Departmental Member

Dr. Debby Ianson, School of Earth and Ocean Sciences
Outside Member

Crustaceans, the most abundant group of organisms that make up zooplankton, form a critical link in the food web between primary-producing phytoplankton and planktivorous fish. Examining this link is essential in order to effectively estimate the amount of energy available to higher trophic levels. The most appropriate currency for tracking energy flow through these food webs is to measure production, or the amount of new biomass generated over a given period of time. Although measurements of primary productivity are routinely made in oceanographic studies, estimates of secondary productivity are rare due to their historical reliance on time-consuming methods. The overall objective of this thesis was to determine the factors influencing temporal variations in community-level crustacean productivity. A simplified lab experiment was used to establish a relationship between diet and chitobiase-based estimates of copepod productivity in response to single versus mixed species phytoplankton diets. In addition, the relationships between primary productivity and chitobiase-based productivity for the entire crustacean zooplankton community were examined over two years in Saanich Inlet, British Columbia, Canada. Lastly, this work determined the abiotic and biotic factors most strongly influencing crustacean productivity in the tropical Guanabara Bay, Rio de Janeiro, Brazil, dominated by the microbial loop. Results from this work show that: (i) copepod populations fed a poor food item take longer to develop through early stages, have lower daily growth rates, and exhibit lower productivity than those fed a good quality food item; (ii) important variations in crustacean productivity are missed when biomass estimates, alone, are used to represent food available to higher trophic levels; (iii) relationships between primary productivity and crustacean productivity can vary interannually and are not necessarily controlled by bottom-up processes; (iv) substantial interannual variations in trophic transfer efficiency (TTE) occur even if average TTE is the same across years; and (v) community-level crustacean productivity in tropical regions dominated by the microbial food loop can be as high as, if not higher than, productivity measured in temperate regions. Ultimately, this work provides insight into how accurate productivity estimates can improve our understanding of zooplankton dynamics in both laboratory and field settings in marine ecosystems worldwide.

Table of Contents

List of Tables

List of Figures

Acknowledgements

I am grateful to my supervisor, John Dower, for his continuous support, encouragement, patience, and for providing me with numerous opportunities for both professional and personal growth throughout my duration in his lab. I also thank my lab mates, past and present, especially Lu Guan and Dan Bevan for useful discussions, camaraderie, and assistance with both field and lab work.

I thank my supervisory committee, Diana Varela, Steve Perlman, and Debby Ianson for their support of this project and for the helpful suggestions and guidance along the way. Special thanks to Diana for the countless hours of advice and for making me an honorary member of her group. Also thanks to Karina Giesbrecht and Marcos Lagunas for putting up with my endless questions about phytoplankton.

Many thanks to Captain Ken Brown and the crew of the *MSV John Strickland*. Field work would not have been possible without the efforts of: Danielle Willmon, Molly Neil, Aidan Neill, Shea Wyatt, Joel White, Arielle Kobryn, and many other graduate and undergraduate volunteers. Thanks to Jonathan Rose for being ever so helpful and to Ian Beveridge for assistance with cruise prep. Also thank you to Dave Mackas, Doug Yelland, and Marie Robert for allowing me to take part in the La Perouse research cruises on the *CCGS JP Tully*.

I am especially grateful to Akash Sastri for the invaluable help with the chitobiase component of this project, and for the inspiring and thought-provoking discussions over the years. Thank you to Moira Galbraith for taking the time to teach me about marine zooplankton identification. Thanks to Rana El-Sabaawi for sharing her knowledge on fatty acids and to Sergei Verenich for analyzing my samples.

I also thank my colleagues in Brazil (Gisela Figueiredo, Jean Valentin, Tatiana Avila, Raquel Neves, and Vanessa Baptista) for welcoming me into their lab for three months, putting up with my broken Portuguese, and for looking out for me while living in a foreign country.

Last, but not least, I am deeply indebted to my parents Dawna and Dave Suchy, my twin sister Corinne Suchy, Robert Munro, Marie Noel, and many other friends and family members for their unwavering love and support throughout this journey. Thank you for seeing me through the tough times. This dream began many moons ago and I could not have reached this point without you.

Dedication

This thesis is dedicated to my high school biology teacher, Cheryl Bailey, for inspiring me to pursue a career in biology and for leading me down a path that would help me to find my place in the world.

Chapter 1: Introduction

1.1 General introduction

Crustaceans, the most abundant group of organisms that make up zooplankton, form a critical link in the food web between primary-producing phytoplankton and planktivorous fish. Examining this link is essential in order to effectively estimate the amount of energy available to higher trophic levels. Although the classical view of the food chain suggests that energy flows from large diatoms through copepods and euphausiids to fishes and whales (Pomeroy 1974), it is now generally understood that energy and carbon may also be channelled via bacteria through the microbial loop and subsequently transferred to protozoa, larger zooplankton, and eventually fish and higher trophic levels (Pomeroy 1974, Azam et al. 1983, Pomeroy et al. 2007). While the classical food chain is assumed to be characteristic of temperate regions wherein large diatoms dominate the phytoplankton community during the spring bloom, the dominant phytoplankton species in tropical regions are often too small to ingest (Ryther 1969, Sommer et al. 2002) and thus copepods feed primarily on the 'microbial loop' throughout the year. The most appropriate currency for tracking energy flow through food webs is to measure production, or the amount of new biomass generated over a given period of time (Rigler and Downing 1984). Although measurements of primary productivity have been routinely made in oceanographic studies since the 1950s (Steeman-Nielsen 1952), estimates of secondary productivity are rare due to their historical reliance on time-consuming methods. Moreover, these estimates have typically been limited to production rates for only a single (or a few) copepod species.

The overall objective of this thesis is to determine the factors influencing temporal variations in community-level crustacean productivity. Specifically, this thesis examines the effects of variations in phytoplankton diet on crustacean productivity in a controlled lab setting as well as in natural field settings in regions representing either a classical or a microbial-dominated food web. Ultimately, this study represents the first research to quantitatively link routine estimates of community-level crustacean productivity to changes in food availability, food quality, and primary productivity. Throughout the thesis, the term crustacean productivity is used instead of secondary productivity because some of the individuals captured in our estimates do not necessarily occupy the second trophic level.

1.2 Methods for estimating crustacean production

1.2.1 Traditional methods

Traditional methods for estimating copepod growth rates and thus production have often relied on time-consuming incubations of copepods (e.g. Landry 1978, Uye 1982, Berggreen et al.1988, McKinnon and Duggan 2003). One of the most common methods is the artificial cohort method (Kimmerer and McKinnon 1987), which involves size-fractionating zooplankton samples with a variety of mesh sizes in order to create artificial cohorts. These cohorts are then incubated for a specific period of time (up to 50 h) at which point individuals are enumerated to stage. Direct weight estimates of the animals before and after the incubations are then used to estimate growth rates. Another, albeit less common, incubation method is the "moult rate method" (Peterson et al. 1991, Hirst et al. 2005). This method involves incubating a specific stage or size class of animals over a defined period of time, counting the number of moulted individuals, and

then measuring the change in weight from one stage of development to the next. Aside from being time consuming, incubation methods involve the repeated handling of animals, which may damage individuals or increase the likelihood of mortality. In addition, these methods require the arduous task of sorting and identifying individuals to specific life stages. As a result, these techniques are not very practical, particularly in a field setting.

The egg production method (Kiørboe and Johansen 1986, Poulet et al. 1995), on the other hand, has been widely accepted due to its feasibility in the field. Since adult copepods do not moult, all of their production is expressed as egg production as opposed to somatic growth. The egg production method involves sorting adult females from zooplankton tows and incubating them for ~24 h in bottles containing *in situ* water (Kiørboe and Johansen 1986). At the end of the incubation period, the number of eggs is counted and converted to production estimates. One key limitation to the egg production method is that it assumes the estimated egg production during incubation reflects *in situ* conditions, which does not always hold true (Saiz et al. 1997). In addition, the fact that egg production is temperature dependent may lead to biased results at higher temperatures (Saiz et al. 1997). More importantly, production estimates using this method cannot be applied to the entire community unless measurements are made showing that juvenile growth rates and female egg production rates are equivalent (Calbet et al. 2000).

1.2.2 Global predictive models

As an alternative to the more traditional methods of estimating production, copepod growth rates and copepod production have been predicted using global

mathematical models. Derived from the literature of previously published copepod growth rates, these models are based on temperature, body size, food quantity, or a combination of these factors (e.g. Huntley and Lopez 1992, Hirst and Lampitt 1998, Hirst and Bunker 2003). The use of models has grown in popularity because production can be estimated for the entire community as opposed to for a single or a few species (Huntley and Lopez 1992). That said, these simplistic models have been met with some criticism. For example, Kleppel et al. (1996) criticize the temperature-dependent model arguing that although the regression between temperature and growth rate is predictive, it does not imply cause and effect. Furthermore, while these models may provide realistic estimates of copepod growth rates, zooplankton biomass estimates from net tows are still required in order to calculate production.

1.2.3 Instantaneous methods

Efforts have been made to develop more instantaneous methods for estimating crustacean production. For instance, Roff et al. (1994) developed a radiochemical method for measuring crustacean growth rates based on the incorporation of ^{14}C into the exoskeleton of *Daphnia*. Although this method is comparable to the widely used methods for measuring primary production (Steeman-Nielsen 1952), radiochemical labeling in crustaceans still requires long incubation periods (>35 h) and thus this technique has not been used routinely in a field setting. In terms of nucleic acid methods, RNA content has been used to directly estimate somatic growth in *Calanus finmarchicus* (Wagner et al. 1998). However, the main drawback to this method is that it provides a useful index of physiological condition for a single species or population as opposed to the entire

community. In addition, aminoacyl-tRNA synthetases (AARS) are a group of enzymes that have been used as an index for estimating growth in zooplankton (Yebra and Hernández-León 2004, Yebra et al. 2005), yet only weak relationships between daily growth rates and AARS activity have been found. Moreover, as is the case with measuring RNA content, the AARS method is not applicable across an entire population or community.

1.2.4 Chitobiase method

Within the last twenty years, a promising method for obtaining routine estimates of community-level secondary productivity involving measurements of the crustacean moulting enzyme chitobiase has been developed (e.g. Oosterhuis et al. 2000, Sastri and Roff 2000, Knotz et al. 2006). This method was originally studied in freshwater systems (Espie and Roff 1995a, Espie and Roff 1995b); however, initial studies severely overestimated growth rates due to the inability to differentiate between the digestive and moulting isomers of the chitobiase enzyme. Recently, this method has been validated in terms of making *in situ* measurements at sea in both temperate (Sastri and Dower 2006, Sastri and Dower 2009) and tropical (Avila et al. 2012) regions. Upon moulting, crustaceans release chitobiase into the surrounding water column, thereby allowing direct estimates of productivity to be made by measuring the decay rate of chitobiase activity. Significant relationships between crustacean moulting rate and mean chitobiase activity (Oosterhuis et al. 2000) and between body length (and weight) and chitobiase activity (Sastri and Dower 2006) have been established.

The main advantage of the chitobiase method is that productivity estimates can be made directly and rapidly in a field or laboratory setting without the need for biomass estimates or measurements of growth rates. Furthermore, productivity estimates are representative of the entire community as opposed to just for a single or a few dominant species. Compared to many of the traditional methods that rely on repeated handling, measuring, and identification of individuals, the chitobiase method provides production rates for all stages and size classes of moulting crustaceans (e.g. from nauplii through copepodites). Even though this method applies only to crustaceans, it likely represents most of the zooplankton production in a given region due to the fact that zooplankton communities are comprised mainly of copepods and other crustaceans (Chisholm and Roff 1990b).

1.3 Factors influencing crustacean production

1.3.1 Temperature

Early studies investigating the factors that influence copepod growth and development focused mainly on the effects of temperature. In fact, Huntley and Lopez (1992) suggest habitat temperature, alone, explains more than 90% of the variance in copepod growth rates. Simple Q_{10} effects, the measure of the rate at which a physiological or biochemical reaction increases or decreases when subjected to a 10°C change in temperature, provide the most obvious explanation for differential growth and developmental rates observed both within and among copepods species. Copepod growth rates have been shown to increase with increasing temperature (Campbell et al. 2001), whereas development time tends to decrease with increasing temperature (Landry 1975,

Lonsdale and Levinton 1985, Campbell et al. 2001). As a result, high growth rates coupled with fast development times result in copepods generally exhibiting a shorter lifespan at higher temperatures (Uye 1988).

In addition, because weight-specific respiration increases with environmental temperature, tropical copepods have a higher rate of respiration than boreal copepods of the same size (Huntley and Boyd 1984). Due to Q_{10} effects, the metabolic and nutritional requirements of copepods increase rapidly with temperature and are more likely to influence the growth rate at high rather than low temperatures (Kleppel et al. 1996). McLaren and Corkett (1981) determined that copepods reared in the laboratory tended to be slightly larger at lower temperatures. Although temperature certainly plays a key role in copepod growth and development, the aforementioned studies have predominantly been conducted in the laboratory under food-saturated conditions. As such, the strong effect of temperature observed for copepod growth and development could be a result of the fact that food quantities were never limited in these studies.

1.3.2 Body size/weight

Under the food-saturated conditions mentioned above, early studies generally found no evidence of weight-dependence when analyzing maximum growth rates (e.g. Huntley and Boyd 1984). In contrast, Banse and Mosher (1980) suggested that a species' body mass at maturity could be used to estimate the mass-specific production rates of invertebrates in the absence of growth rate measurements. Subsequent studies have shown that body size, in terms of both weight and length, may be a good correlate of growth rate for calanoid copepods (Hopcroft et al. 1998b). For instance, juvenile

copepods tend to have a constant growth rate over a wide range of body sizes (from 0.3 to 5 µg dry weight), while the growth rate of individuals >5 µg declines with body size (Peterson et al. 1991). In addition, the mean moulting rate for small copepods is approximately 50% per day, equivalent to a stage duration of 2 days, whereas larger species moult at a lower rate with a longer stage duration (Peterson et al. 1991). In general, copepod growth is higher during early (naupliar) stages and decreases during the copepodite stages as individuals reach their maximum body weight (Campbell et al. 2001, Liu and Hopcroft 2007). Furthermore, adult female weight-specific growth decreases with increasing body size in both broadcast-spawners and sac-spawners under food-saturated conditions, while growth of juveniles is independent of weight (Hirst and Lampitt 1998). These studies propose that larger species and stages may be able to sustain a lower growth rate because of lower metabolic costs and higher lipid reserves (Hopcroft et al. 1998b). Caution should be taken when interpreting body size versus growth rate results, however, as higher growth rates of smaller species and stages may be more apparent than real because of differential mortality and survivorship of slower and fast growing individuals, respectively (Hopcroft et al. 1998b).

1.3.3 Food quantity

Food quantity, most commonly measured as total chlorophyll *a* concentration, has been extensively studied, both on its own and in conjunction with other variables (e.g. temperature, body size), as one of the factors influencing growth and development of copepods. A critical food concentration must be met in order for copepods to grow and develop at maximum rates (Huntley and Boyd 1984). Conversely, if the food

concentration falls below this critical concentration, growth and development become food-limited. Copepods in different oceanic regimes may experience different levels of food-limitation. For example, copepods in open ocean regions are more likely to be food-limited than zooplankton in coastal regions; however, open ocean species may be subjected to occasionally high food concentrations (Huntley and Boyd 1984). Strongly coastal environments, on the other hand, may experience periods of the year (e.g. winter) when food availability falls below the critical concentration and thus growth becomes food-limited (Huntley and Boyd 1984). In general, the growth and development of larger copepods is highly dependent on food concentration because the critical food concentration increases with increasing body size (Vidal 1980a, 1980b). High food concentrations have been shown to increase the body size and weight of copepodite stages (Davis and Alatalo 1992, Hopcroft et al. 1998b). Therefore, growth in smaller species and stages is less affected by low food concentrations, while large species and stages are more severely compromised in their growth at low food concentrations (Hopcroft et al. 1998b).

Not surprisingly, temperature also confounds the influence of food quantity on development and growth. For example, Lonsdale and Levinton (1985) found that algal densities required to attain faster copepod growth rates and higher survival increased with increasing temperature, thereby suggesting the influence of food concentration was dependent on temperature (Lonsdale and Levinton 1985). While the degree of food limitation in nature increases with increasing temperature for adults, it does not have the same effect on juveniles as their growth is close to food saturation at all temperatures (Hirst and Bunker 2003). Many laboratory and incubation studies examining the effects

of varying food concentrations on the growth and development of copepods have been conducted at constant temperatures to avoid the confounding effects of varying temperatures (e.g. Davis and Alatalo 1992, Hopcroft et al. 1998b, Campbell et al. 2001). Liu and Hopcroft (2006) found that after removing the effects of temperature, growth rates for CIV and CV stages of *Metridia pacifica* are more dependent on food concentration than are earlier stages CI to CIII. In contrast, growth of a smaller calanoid copepod species, *Pseudocalanus* sp., tends to be more dependent on temperature than on food (Liu and Hopcroft 2008). Furthermore, the development rates of *Pseudocalanus* sp. were similar to other calanoid copepods; however, growth rates of this species were considerably lower, likely an adaptation to keep this species smaller and reduce potential visual predation (Liu and Hopcroft 2008). In addition to body size, the development, growth rate, and even survival of copepods has been shown to decrease from high to low food concentrations (e.g. Lonsdale and Levinton 1985, Davis and Alatalo 1992, Campbell et al. 2001).

Hirst and Bunker (2003) suggest that food-limitation impacts juveniles and adults differently with respect to survival. For example, if sufficient weight is not added between moults during juvenile stages, slower growing individuals will not survive; however, adults can survive for long periods of time without having sufficient food to produce eggs (Hirst and Bunker 2003, Koski and Breteler 2003). To date, there have only been a few studies that have addressed specifically the effects of food quantity on the survival of copepods and no clear relationship between the two variables has been found (e.g. Davis and Alatalo 1992, Crain and Miller 2001). For instance, being exposed to pulses of food versus a constant food supply had no significant effect on the survival of

copepods (Davis and Alatalo 1992). Similarly, Crain and Miller (2001) found that although food conditions may fall below the level required to sustain maximal development, mortality caused directly by food-limitation is probably low. Thus, it seems more likely that individuals whose development slows due to food limitation may suffer increased mortality through a higher probability of predation rather than their survival being affected directly by food concentration (Hopcroft et al.1998b). The idea that slower growth leads to an increased risk of predation has already been formalized in the larval fish literature as the "stage-duration" (Leggett and Deblois 1994) and "growth-mortality" hypotheses (Ware 1975).

1.3.4 Food quality

Recent studies on the factors influencing growth and development of copepods have moved beyond estimates of food quantity and have focused on addressing the issue of food quality with the use of fatty acid "bioindicators". Fatty acid analyses have provided information on the diet and feeding strategies of copepods (e.g. Graeve et al. 1994a, Graeve et al. 1994b, Stevens et al. 2004, Arendt et al. 2005, El-Sabaawi et al. 2009a). Therefore, in contrast to the use of chlorophyll *a* as a snapshot representation of what copepods are eating, fatty acids provide information on the nutritional quality of a copepod diet over a longer time period. As copepods are unable to readily synthesize fatty acids, these compounds must be obtained from their phytoplankton diet for growth and development (Bell et al. 2007). Certain essential fatty acids are characteristic of diatoms and dinoflagellates and can thus be used to detect the phytoplankton groups ingested by zooplankton (Graeve et al. 1994a). More specifically, because dinoflagellates

are rich in docosahexaenoic acid (DHA) while diatoms are rich in eicosapentaenoic acid (EPA), the ratio of DHA to EPA can be used to determine the proportion of dinoflagellates to diatoms in the copepod diet (Dalsgaard et al. 2003, Viso and Marty 1993).

While the majority of studies involving food quality for copepods have focused on the effects of various diets on egg production and hatching success (e.g. Jónasdóttir et al. 2005, Koski et al. 2006, Vargas et al. 2006), a few key studies have examined the impact of food quality on development and growth of juveniles and adult stages. Peterson et al. (1991) showed that different nutritional requirements might result in variations in female and juvenile growth rates. Furthermore, in a study examining the effects of food quality on size, weight, and development time of *Centropages typicus*, Bonnet and Carlotti (2001) found that there was a significant impact of diet on copepod weight, in addition to the duration of all stages (except for CIV and CVI). In yet another study, Klein Breteler et al. (2005) determined the effects of nutrient-limited algae on the growth and development of copepods and found that nitrogen- and phosphorus-limited algae reduced the growth rate of copepods while completely halting development altogether. Specifically, a change in the content of polyunsaturated fatty acids (PUFAs) and/or sterols in the algae resulted in a decrease in the essential building blocks required for copepod development (Klein Breteler et al. 2005). Mixed diets of dinoflagellates and diatoms, and thus favourable ratios of these essential fatty acids, have been shown to result in higher growth rates and higher reproductive success in copepods (e.g. Klein Breteler et al. 1990). In contrast, low ratios of DHA:EPA corresponded to the collapse of the local population of the copepod, *Neocalanus plumchrus*, in the Strait of Georgia,

British Columbia (El-Sabaawi et al., 2009), coinciding with a failure of this species to moult past the CII copepodite stage (Sastri and Dower 2009). Copepod survival, on the other hand, appears to be only affected by food quality at high food concentrations, whereas population dynamics are not limited by food quality at food concentrations too low to support growth (Koski and Breteler 2003). Ultimately, unfavourable ratios of essential fatty acids in the algal diet of copepods can significantly impact higher trophic levels by varying the growth rates of larval fish (St. John et al. 2001).

1.4 Current limitations

1.4.1 Using biomass to estimate zooplankton production

Due to the lack of consensus as to how zooplankton production should be estimated, total zooplankton biomass is often used to represent food available to higher trophic levels. One of the major shortcomings of using this crude estimate is that it incorrectly assumes that everything collected in the sample represents food available to the consumer. Moreover, zooplankton biomass estimates may be biased depending on the mesh size of the net used in collecting the sample. For example, larger mesh sizes (over 200 μm mesh) have been shown to undersample a large proportion of the zooplankton community (Hopcroft et al. 2001), whereas smaller mesh sizes (less than 100 μm) may result in a higher abundance and biomass of small-bodied zooplankton compared to larger individuals (see Turner 2004 for review). In addition, undersampling may occur if large zooplankton exhibit avoidance behaviour in response to smaller nets with smaller mouth diameters (Hovekamp 1989). Furthermore, smaller nets may clog more easily in productive coastal waters, which could also result in undersampling the zooplankton biomass.

Despite these shortcomings, estimates of biomass or standing stock are often extrapolated to estimates of zooplankton production, which becomes problematic in oceanic systems wherein advection, patchiness, and vertical migration come into play (Oosterhuis et al. 2000). Additionally, inaccuracies may result when net-based biomass values are used to calculate production from global predictive models. Therefore, a consistent method of routinely estimating crustacean productivity would allow oceanographers to more accurately interpret results across studies. Also, accurate estimates of crustacean productivity allow for the direct testing of major assumptions in terms of energy flow in food webs, i.e. that changes in primary productivity result in corresponding changes to crustacean zooplankton productivity. Ultimately, routine productivity estimates provide critical information as to how efficiently energy is transferred throughout the food web, and how much of this energy is potentially available to consumers.

1.4.2 Chlorophyll *a* as a proxy for phytoplankton biomass

Despite its wide use as a proxy for phytoplankton biomass, many studies have failed to show a strong correlation between chlorophyll *a* concentrations and growth in copepods (e.g. Kimmerer and McKinnon 1987, Hopcroft et al. 1998a, Shreeve et al. 2002). Hopcroft et al. (1998a) found that including chlorophyll *a* data did not improve the regression between body size and growth rate. Similarly, when Kimmerer and McKinnon (1987) measured the *in situ* growth rate of *Acartia tranteri*, they found that temperature and chlorophyll together explained only 50% of the variance in the growth rate of this species. Along the same lines, increased growth rates of copepods in the southern

Benguela system were not related to changes in chlorophyll *a* concentration (Hutchings et al. 1995). In addition, in the waters around South Georgia, neither stage duration nor growth rates were related to temperature or chlorophyll *a* (Shreeve et al. 2002). Lastly, although temperature, chlorophyll *a*, body size and development stages combined accounted for more than 60% of the variability in growth rate for *Calanus marshallae*, the same study could not determine a similar relationship for *C. pacificus* (Liu and Hopcroft 2007).

Not surprisingly, weak or conflicting relationships between chlorophyll concentration and copepod growth may occur because chlorophyll concentration does not necessarily represent food available to copepods. For instance, although size-fractionated chlorophyll concentrations provide a rough idea of the range of food particles available to copepods, different species and/or developmental stages likely feed on specific size ranges of particles as opposed to feeding on the entire food spectrum (Paffenhöfer 1984). Moreover, chlorophyll concentration does not provide any information on the quality of the phytoplankton diet, which varies with the composition of essential fatty acids (Klein Breteler et al. 2005) and the age of the phytoplankton being consumed (Jónasdóttir, 1994; Jónasdóttir and Kiørboe, 1996). In addition, many copepods consume non-photosynthetic prey items (e.g. ciliates and heterotrophic nanoflagellates) (see Turner 2004), which are not represented by measurements of chlorophyll. As a result, relationships between chlorophyll *a* and crustacean growth are less likely to be observed in regions wherein individuals are feeding primarily on the microbial loop. Lastly, strong correlations between chlorophyll and copepod growth rates may be lacking due to the fact that copepod growth is more likely a function of the feeding environment experienced by the

copepod in the days/weeks prior to a given sampling date. Accurate estimates of productivity provide an opportunity to assess the relative importance of food availability, in addition to the usefulness of chlorophyll *a* as a proxy, on the somatic production of crustacean communities in different oceanic regions.

1.4.3 Extrapolating lab results to a natural field setting

Despite the evidence for the effects of low food concentrations on copepods, the majority of this research has been conducted in the laboratory setting (e.g. Vidal 1980a, 1980b, Davis and Alatalo 1992) or using incubation techniques at sea (e.g. Kimmerer and McKinnon 1987, Peterson 1991, Hopcroft et al. 1998a, Liu and Hopcroft 2006, 2007, 2008). Although these techniques are necessary because food-limited individuals are usually not available for the assessment of growth and production in nature (Kleppel et al. 1996), they may not necessarily represent conditions that a copepod experiences in the field. For example, although copepods likely experience a highly variable food environment on time scales of minutes and length scales of centimeters, they may be able to effectively locate non-limiting food patches (Huntley and Lopez 1992). The copepod *Centropages typicus* has been shown to integrate temporal fluctuations in food supply over 0.5-1.0 days indicating that, at least to some degree, this species has the capacity to buffer against changing food conditions (Davis and Alatalo 1992). Therefore, regardless of the oceanic regime within which they are found, copepods may always be able to find sufficient food in order to grow at maximal rates. Huntley and Lopez (1992) suggest that appearances to the contrary may be due to sampling at the wrong scales, and that

copepods may always be able to find food on the micro scales relevant to their feeding and movement.

A major problem with using fatty acids as bioindicators of food quality stems from the inconsistent and/or contradictory results between lab and field studies. The majority of lab studies have used the fatty acid composition of phytoplankton to investigate reproductive success (i.e. egg production, hatching success) and copepod development (e.g. Jónasdóttir et al. 2005, Koski et al. 2006, Vargas et al. 2006), yet conflicting results even between lab studies have left these data difficult to interpret. Although lab studies have clearly demonstrated the potential for variations in food quality to affect the growth of copepods (e.g. Jónasdóttir et al. 2009) and that this variability can be transmitted to larval fish in terms of variable growth rates (St. John et al. 2001), these studies often focus on a single species of copepod feeding on one (or a few) species of phytoplankton. Therefore, the observation that simple lab manipulations of food quality can affect copepod growth is no guarantee that such effects actually occur in nature, where consumers are exposed to a much wider prey field and, presumably, a range of foods of differing nutritional quality. To date, poor food quality has been inferred to contribute to low crustacean productivity as a result of the collapse of a biomass-dominant copepod species (El-Sabaawi et al. 2009, Sastri and Dower 2009). However, explicit testing of the relationship between food quality and crustacean productivity in a field setting is lacking. Given that energy (carbon) is converted into zooplankton biomass at a lower rate when food quality limits growth (Vargas et al. 2010), accurate estimates of crustacean productivity provide a crucial link between food quality of phytoplankton and the survival, growth, and recruitment of the dominant fish species in a given region.

1.5 Thesis objectives and structure

The three primary objectives of this thesis are to:

1) **Use a simplified lab experiment to establish a relationship between diet and chitobiase-based estimates of copepod productivity in response to single versus mixed species phytoplankton diets (Chapter 2).**

It has been demonstrated that variations in diet, and associated food quality, produce changes in egg production rates of copepods. However, a direct relationship between diet and somatic production has yet to be established. This chapter tested the sensitivity of the chitobiase method as a tool for measuring the productivity response of a single copepod species to different diets by rearing the harpacticoid splash-pool copepod, *Tigriopus californicus*, on different phytoplankton diets while keeping temperature and food quantity constant. Although formatted for this thesis, this chapter has already been published (Suchy et al. 2013) in the Journal of Plankton Research. Our use of the chitobiase method in a lab setting demonstrated the potential utility and sensitivity of this approach for field studies examining the impact and significance of short-term shifts in food quality on entire crustacean zooplankton communities.

2) **Examine the relationships between primary productivity and chitobiase-based productivity for the entire crustacean zooplankton community over two years in a highly productive, temperate fjord (Chapter 3).**

The goal of this chapter was to determine how the dominant crustacean zooplankton in Saanich Inlet, BC, Canada, are influenced by seasonal and interannual variations in a phytoplankton community representative of a temperate coastal marine food web. The abiotic and biotic factors that best explained the variation in both primary productivity and crustacean productivity were determined and the ways in which these factors influenced the relationship between phytoplankton and zooplankton production rates were examined. This study was a collaborative effort between the Dower and Varela labs at the University of Victoria. The Varela lab measured nutrients, chlorophyll concentrations, biogenic silica, phytoplankton abundance, and primary productivity. All other analyses were performed by the Dower lab. This chapter will be submitted to Marine Ecology Progress Series. Results from this study provide insight into how energy is transferred to higher trophic levels based on interannual variations in the structure of both the phytoplankton and zooplankton communities.

3) **Determine the abiotic and biotic factors that most strongly influence crustacean productivity in a tropical coastal bay dominated by the microbial loop (Chapter 4).**

Estimates of copepod productivity in tropical regions are even sparser than comparable estimates in temperate regions. The overall aim of this study was to use the chitobiase method to obtain routine estimates of community-level crustacean productivity for the highly eutrophic Guanabara Bay, Rio de Janeiro, Brazil. Our main objective was

to determine the abiotic and biotic factors most strongly influencing crustacean productivity in Guanabara Bay, wherein copepods feed primarily on the microbial food web. This work was a collaborative effort with colleagues from the Federal University of Rio Grande and the Federal University of Rio de Janeiro, Brazil. This chapter will be submitted for publication to Marine Biology. Ultimately, this study reveals that small, fast growing copepods can contribute just as much, if not more, energy to higher trophic levels in tropical regions compared to temperate regions.

The thesis concludes with a synthesis of the major findings (Chapter 5), addressing the importance of routine estimates of crustacean productivity and the implications that the results from Chapters 2 to 4 have on future research in the field of zooplankton production and energy transfer in marine food webs.

Chapter 2: Influence of diet on chitobiase-based production rates for the harpacticoid copepod *Tigriopus californicus*

2.1 Introduction

In marine ecosystems, copepods represent the major link between phytoplankton and higher trophic levels. As such, an understanding of whether different phytoplankton diets influence the productivity of copepod communities is necessary in order to examine energy transfer within marine food webs. The effects of variations in diet on copepod egg production and hatching success have been extensively studied in both laboratory (Koski et al. 2006, Vargas et al. 2006, Koski et al. 2010, Daase et al. 2011) and field settings (Jónasdóttir et al. 2005); however, inconsistent and/or conflicting results between lab and field studies still persist in the literature. These inconsistencies may arise from species-specific differences in the growth and development responses of copepods to a given algal species grown in the lab (Koski and Klein Breteler 2003). Furthermore, the prevalence of food-saturated conditions in the lab versus the food limiting conditions often present in a natural setting may further complicate the interpretation of results.

Mixed phytoplankton diets have often been suggested as being better for copepods than a single algal species because a diverse diet is more likely to ensure that all of the nutrients required by copepods can be obtained (Kleppel 1993, Kleppel and Burkart 1995, Arendt et al. 2005, Koski et al. 2006). A mixed diet increases the likelihood that copepods will obtain digestible food items in order to ensure survival, while an array of food items with different biochemical compositions enhances growth and reproduction (Koski and Klein Breteler 2003). Favourable ratios of the essential fatty acids found in diets consisting of both dinoflagellates rich in docosahexaenoic acid

(22:6n-3, DHA) and diatoms rich in eicosapentaenoic acid (20:5n-3, EPA), have been demonstrated to result in higher growth rates and higher reproductive success in copepods (e.g. Klein Breteler et al. 1990). A poor diet lacking in any of the essential nutritional requirements for a given stage may result in slow growth or even developmental arrest (Klein Breteler et al. 2005), thereby affecting overall production (Kleppel and Burkart 1995). Low ratios of DHA:EPA were shown to correspond with the collapse of an entire copepod species, *Neocalanus plumchrus*, in the Strait of Georgia, British Columbia (El-Sabaawi et al. 2009), coinciding with a failure of this species to moult past the CII copepodite stage (Sastri and Dower 2009). The collapse of an entire biomass-dominant species would undoubtedly impact the overall production of the zooplankton community and thus the energy available as food for higher trophic levels. For instance, a decline in growth and survival of the major juvenile fish species in the Strait of Georgia has recently been linked to low food production (Beamish et al. 2012, Thomson et al. 2012). Curiously, despite the compelling evidence that mixed diets are necessary for optimal copepod growth and development, single species diets of the diatom *Thalassiosira weissflogii* have also been shown to support high levels of both egg production (Jónasdóttir et al. 2009) and copepod development (Klein Breteler et al. 2005). As a result, the relationships between diet and copepod growth parameters still require further clarification.

Specifically, it has been demonstrated that variations in diet, and associated food quality, do produce changes in egg production rates of copepods, however, these estimates are limited to the effects of diet on adult production. A direct relationship between diet and non-reproductive production, i.e. the production associated with

somatic growth, has yet to be established. Historically, estimates of zooplankton production have been rare due to the time-consuming nature involving incubations of a specific size class of copepods (e.g. Peterson et al. 1991) or the development of artificial cohorts (e.g. Kimmerer and McKinnon 1987). More recently, measurements of the crustacean moulting enzyme chitobiase have been used to obtain rapid estimates of production rates for zooplankton communities without the need for repeated handling of organisms (Sastri and Dower 2006, Sastri and Dower 2009, Avila et al. 2012). Upon moulting, crustaceans release chitobiase into the surrounding water column, thereby allowing direct estimates of biomass production rates to be made by measuring the decay rate of this enzyme in the ambient water. Significant relationships between body length (and weight) and the rate of production of chitobiase activity (Sastri and Dower 2006) have already been established for marine copepods. In addition to providing zooplankton biomass production rates, chitobiase activity and decay rate measurements may also be used as proxies for the number of actively moulting individuals in the water column, the mean individual stage duration, and the growth rate of copepods (Sastri and Dower 2006).

Here, we tested the sensitivity of the chitobiase method as a tool for rapidly capturing the response of copepods to variations in their diet. Temperature and food quantity were kept constant while newly hatched nauplii of the harpacticoid copepod *Tigriopus californicus* were reared on different phytoplankton diets until at least one generation of development was complete. Previous studies have shown that *T. californicus* can be reared on a wide range of dietary items including algae, shrimp powder, bacteria, and even rat chow (see Powlik et al. 1997), thereby making it an ideal

species to use in laboratory studies. The phytoplankton species used in this experiment were chosen on the basis of similarity of size in order to minimize the confounding influences of particle size on digestibility and food handling time (see Kleppel 1993). The overall goal of this study was to use a simplified lab experiment to establish a relationship between diet and chitobiase-based estimates of copepod production rates in response to single versus mixed species phytoplankton diets. Given that the influence of a variable diet on somatic copepod production still remains largely unknown, results from this study will provide the foundation for investigating more complex interactions in a natural field setting. Ultimately, the baseline relationships established in this study may provide insight for future studies examining the combined effects of diet, food availability, food quality, and their overall impact on community-level zooplankton production in the field.

2.2 Methods

2.2.1 Copepod cultures

The experiment was conducted using the harpacticoid copepod *Tigriopus californicus*, commonly found in splash pools (above the high tide mark and replenished with seawater due to splashing waves). This species is known for its tolerance to extreme environments exhibiting wide ranges of temperature and salinity (Lear et al. 1962, Powlik et al. 1997, Lewis et al. 1998). Individual *T. californicus* were collected during the summer (June) in 1 L Nalgene bottles from splash pools located near 10 Mile Point, Victoria, British Columbia, Canada (48° 27′ N, 123° 16′ W). Copepods were transferred to 2 L glass jars in the laboratory and maintained in 1.5 L of natural splash pool water at

21°C with a 16:8 h light: dark cycle for at least one month in order to allow for acclimation to laboratory conditions. Copepod culture water was replenished every few days with freshly collected 0.2 μm-filtered splash pool water.

2.2.2 Phytoplankton cultures

The phytoplankton species used in this experiment was the diatom *Thalassiosira weissflogii* and the dinoflagellate *Amphidinium carterae* (average cell sizes of 11.3 x 3.8 μm and 14.1 x 8.6 μm, respectively). Although the present study focused on the effects of variations in diet rather than on the issue of food quality per se, *T. weissflogii* and *A. carterae* were chosen to represent the essential fatty acid compositions characteristic of diatoms and dinoflagellates. Specifically, *T. weissflogii* is known to have a low DHA:EPA ratio, whereas *A. carterae* has a high ratio of DHA:EPA (see Table 1 and references therein). It is important to note, however, that in addition to these characteristics, the potential toxicity of *A. carterae* has been reported in previous studies (Graeve et al. 1994a, Murray and Marcus 2002, Koski and Klein Breteler 2003, Koski et al. 2006). Phytoplankton cultures were grown at 18°C in 1 L batch cultures in f/2 medium with sodium silicate added to the diatom cultures. Phytoplankton cultures were exposed to 75 μmol photons m^{-2} s^{-1} irradiance in a 16:8 h light: dark cycle. Cultures were allowed to acclimate to these laboratory conditions for at least 10 generations before they were used in the feeding experiment. Cell densities were estimated daily using a Beckman Coulter Z2 Coulter Particle Count and Size Analyzer and *in vivo* chlorophyll fluorescence. Phytoplankton were kept in early exponential growth phase by discarding a fraction of the culture stock and replacing it with new f/2 medium every third day as

Table 1. Contribution of the essential fatty acids 22:6n-3 (DHA) and 20:5n-3 (EPA) reported as percent of total fatty acids in the literature for the phytoplankton species used in the present study.

	% Total Fatty Acids		DHA:EPA	Reference
	22:6n-3	20:5n-3		
Amphidinium carterae	17.4	8.0	2.2	Viso and Marty 1993
	24.9	9.9	2.5	Graeve et al. 1994a
Thalassiosira weissflogii	1.9	8.4	0.2	Arendt et al. 2005
	6.4	23.0	0.3	Jónasdóttir 1994

previous studies have shown that the quality of phytoplankton as a food source for copepods may deteriorate with age (Jónasdóttir 1994, Jónasdóttir and Kiørboe 1996).

2.2.3 Feeding experiment

Newly hatched nauplii were transferred to 500 mL glass jars (20 nauplii per jar) corresponding to one of four diet treatments. Each treatment had three replicates. Hatching was initiated by removing egg sacs from female copepods in order to release a maternal-inhibited hatching mechanism previously described for *T. japonicas* (Kahan et al. 1988). Egg sacs were transferred to individual well plates containing filtered seawater and placed under a 60 Watt incandescent desk lamp until hatching occurred (approximately one hour). Newly hatched nauplii were promptly removed from the well plates, transferred through a series of 1 mL filtered seawater sterile baths, and subsequently placed into one of four treatments corresponding to different phytoplankton diets: dinoflagellates (*A. carterae*), diatoms (*T. weissflogii*), mixed (*A. carterae*/*T. weissflogii*), and a natural food assemblage consisting of water from the splash pool which had been filtered with a 40 μm sieve to remove crustaceans and other zooplankton. Subsequent analysis of water from the splash pool determined that dinoflagellates and nanoflagellates comprised the majority of the natural food assemblage, with diatoms representing only a small fraction of the phytoplankton available in this treatment.

Water in the treatment jars was maintained at 400 mL throughout the duration of the experiment. All treatment jars were monitored with daily cell counts and kept at a constant concentration of at least 250 μg C L^{-1} (approximately 7000 cells mL^{-1}) to ensure food-saturating conditions (Jónasdóttir et al. 2009). Treatment jars were replenished with

150 mL of filtered seawater every second day in order to account for *in situ* phytoplankton growth. Gentle aeration was used to keep the food items suspended. For the mixed diet, approximately 3500 cells mL^{-1} of each of *T. weissflogii* and *A. carterae* were mixed immediately before addition to the treatment. Upon completion of the experiment, the number of surviving individuals in each treatment was counted and prosome lengths (mm) of the surviving copepods were measured using a dissecting microscope.

2.2.4 Chitobiase enzyme assays

Measurements of chitobiase activity (CBA) followed Sastri and Dower (2006) (see Appendix A for terminology and calculations). Enzyme assays were initiated by adding the substrate 4-methylumbelliferyl-β-D-glucosaminide (0.1 mmol MBF-NAG; Sigma) to seawater samples. Assays were conducted at 25ºC and terminated after 60 minutes with the addition of a 2 M NaOH and 0.4 M EDTA solution. Previous work by Sastri and Dower (2006) determined that the substrate remained saturated (there was a linear increase in MBF fluorescence) after 60 minutes when individual *T. californicus* were incubated in much smaller volumes per individual (2 mL) than those used in the current study. The reaction was buffered to pH 6.0 (optimal for copepods) using a 0.15 M citrate-phosphate buffer. Chitobiase activity (nmol MBF liberated L^{-1} h^{-1}) was estimated by measuring the fluorescence of the liberated MBF using a Turner BioSystems Modulus Fluorometer with a UV-absorbance (365 nm excitation and 450 nm emission). Raw fluorescence was then converted to nmol MBF using a standard curve of known 4-methylumbelliferone concentrations against fluorescence.

2.2.5 Chitobiase decay rate estimates

CBA decay rates were estimated from 150 mL aliquots of seawater collected from the treatment jars every four days while food suspensions were being replenished and until the experiment was terminated, i.e. once a second generation of newly hatched nauplii were present in the treatments. Seawater samples were screened with a 40 μm mesh in order to remove any copepods. Individuals retained on the mesh were gently rinsed with 0.2 μm-filtered splash pool water and returned to the treatment jar. On each sampling date, copepods were quickly inspected for a coarse determination of developmental stage. Approximately 15 mL of the seawater sample from each treatment was immediately filtered (0.2 μm) in order to remove any bacteria and subsequently used to estimate the native *in situ* chitobiase activity (CBA_{nat}). A crude homogenate of 20-30 *T. californicus* (freshly ground in 3 mL of seawater) was filtered (0.2 μm) and then used to "spike" the original samples from each treatment to differentiate the decay of CBA from background fluorescence (see Sastri and Dower 2006). Seawater samples were sampled at t=0 (just after homogenate was added), t=1 (6 hours) and t=2 (12 hours), 0.2 μm filtered, and stored at 4°C in disposable glass tubes until assayed. Over the 12-hour incubation period, samples were maintained at 21°C.

Estimates of CBA decay rate (h^{-1}) were calculated as the slope (k) of the natural logarithm of CBA versus time (Sastri and Dower 2006). The reciprocal of the negative slope ($1/-k$) was used to represent the average stage duration, or the time (T_{CBA}) taken for moulting individuals to produce CBA equivalent to the chitobiase activity (CBA_{nat}) in the treatment jar. One of the assumptions of the chitobiase method is that the rate of production of the enzyme by moulting copepods is balanced by its rate of decay in the

water column due to bacterial degradation of the enzyme (Sastri and Dower 2006). However, this assumption is only valid in asynchronously developing populations (and communities). Given that *T. californicus* in our experiment developed synchronously from nauplii through to the adult, we employed the calculation described by Oosterhuis et al. (2000) wherein day-specific change in CBA_{nat} and the rate of decay of CBA were combined to estimate the total amount of chitobiase released per day (Table 2). In addition, dilution of the chitobiase enzyme resulting from the addition of fresh media to the treatments every second day was also taken into consideration when estimating CBA_{nat}.

In order to calculate the absolute amount of biomass produced (ΔB), we applied a known relationship between CBA and the growth increment of marine copepods ($\log(g_{inc}) = 0.864 \log (CBA_i) - 1.78$; Sastri and Dower 2006) to the average CBA_{nat} in each treatment. Daily copepod production rates ($\mu g\ C\ L^{-1}\ d^{-1}$) were then calculated as the biomass production divided by stage duration, or $\Delta B/T_{CBA}$. In this controlled experiment, a stage duration of two days was used for all production rate calculations as this was the time over which the estimated biomass was produced. Chitobiase-based daily growth rates (g, d^{-1}), representing the average weight-specific growth rate of the mean-sized moulting individual, were calculated as the ratio of the daily copepod production rate to the developing biomass as estimated from our corrected values of CBA_{nat} (equivalent to daily P:B ratios determined for crustacean communities; Sastri et al. 2012). As comparable estimates of growth rates are lacking in the literature for *Tigriopus spp.*, g (d^{-1}) was calculated by converting summer body length measurements for all developmental

Table 2. Example of calculation applied to synchronously developing populations of *Tigriopus californicus* used in this study. CBA_{nat} (nmol L^{-1} h^{-1}) and the rate of decay of CBA (d^{-1}) were combined in order to calculate the total amount of chitobiase released per day (after Oosterhuis et al., 2000).

		(*x*)		(*y*)	(*x* + *y*)
	CBA_{nat}	ΔCBAnat	Decay of CBA	Mean	Corrected CBA_{nat}
Day 2	5.2		37.6		
		-0.5		24.7	24.2
Day 6	4.7		11.8		
		-0.7		9.1	8.4
Day 10	4.0		6.3		
		4.6		5.1	9.7
Day 14	8.6		3.8		

stages of *T. californicus* (Powlik et al. 1997) to body weight using the length-weight regression for harpacticoid copepods reported in Hopcroft et al. (1998a) (Table 3). Values of g (d^{-1}) were determined based on a general exponential growth equation and were used as a comparison to the chitobiase-based growth rates obtained over the duration of the experiment.

2.2.6 Statistical analysis

Student's t test was used to test for differences between prosome lengths of male and female survivors within the treatments. One-way analysis of variance (ANOVA) and Kruskal-Wallis one way ANOVA on ranks was used to test for differences between mean stage duration, mean copepod production rates, and daily growth rates amongst the treatments on each sampling date, in addition to differences in mean adult prosome length of surviving individuals. All statistical tests were performed using SigmaPlot version 12.3 (Systat Software Inc., San Jose, CA, USA).

2.3 Results

Upon completion of the 24-day experiment, only one individual survived on the *Amphidinium carterae* diet (Fig. 1). The highest numbers of survivors occurred in the *Thalassiosira weissflogii* and the natural food assemblage treatments (Fig. 1). Although some individuals did survive in the mixed (*A. carterae*/*T. weissflogii*) treatment, the number of individuals was much lower than in the *T. weissflogii* or the natural food assemblage treatments (Fig. 1). Nauplii that fed upon *T. weissflogii* and the natural food assemblage reached the adult stage sooner than all other treatments (Day 18), while

Table 3. Developmental stage, development time from egg (days), and body length (µm) for *Tigriopus californicus* as reported in Powlik et al. (1997). Weight (µg) was calculated using the length-weight relationship for harpacticoid copepods (log W = 2.74 ln L – 16.41) presented in Hopcroft et al. (1998a). These values were then used to calculate g (d^{-1}) using the general exponential growth equation $g = \ln (W_{t+1}/W_t)/T$, where W_{t+1} and W_1 are the weights of each successive time and T represents the stage duration reported by Powlik et al. (1997).

Developmental Stage	Development time from egg (days)	Body Length (µm)	Weight (µg)	g (d^{-1})
NVI	10	250	0.28	-
CI	13	445	1.35	0.25
CII	15	535	2.23	0.32
CIII	18	675	4.22	0.14
CIV	20	750	5.64	0.52
CV male	21	1100	16.09	0.66
CV female	21	1000	12.39	0.26

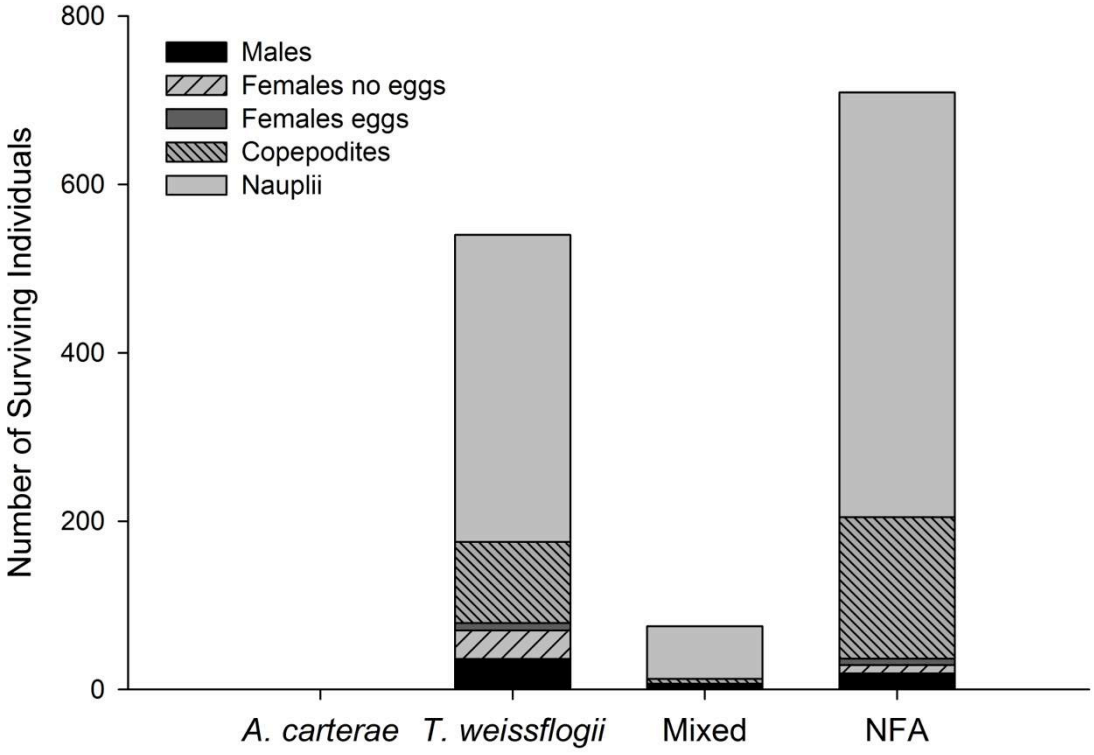


Figure 1. Number of surviving individuals of *T. californicus* for each development stage at the end of the experiment for each treatment: *Amphidinium carterae*, *Thalassiosira weissflogii*, mixed *A. carterae*/*T. weissflogii*, and natural food assemblage (NFA).

copepods that fed upon the mixed diet reached adult stage by the following sampling date on Day 22.

The average prosome length (mm) of surviving adult males was significantly larger than that of surviving females in all treatments (excluding *A. carterae*) ($p < 0.05$, unpaired two-tailed Student's t test, Table 4). A Kruskal-Wallis one-way ANOVA on ranks revealed no difference in the average prosome length of adult males between food treatments ($p = 0.445$). The only statistically significant size-based difference was a smaller average female prosome length for animals in the *T. weissflogii* treatment relative to the natural food assemblage treatment ($p = 0.009$, Kruskal-Wallis one-way ANOVA followed by Dunn's Method).

Average CBA_{nat} values for *T. californicus* fed *A. carterae* remained below 25 nmol MBF L^{-1} h^{-1} throughout the experiment, with the exception of one spike in CBA_{nat} activity on Day 12, after which time the population crashed (Fig. 2a). In contrast, the average CBA_{nat} in the *T. weissflogii* treatment remained below 40 nmol MBF L^{-1} h^{-1} throughout the experiment (Fig. 2b). Similar to the *A. carterae* treatment, the mixed *A. carterae*/*T. weissflogii* diet showed a peak on Day 12; however, this peak was much lower than the peak observed in the *A. carterae* treatment (Fig. 2c). Average CBA_{nat} values for the natural food assemblage remained below 30 nmol MBF L^{-1} h^{-1} throughout the experiment with no discernable pattern (Fig. 2d). In order to explore the impacts of the *T. californicus* crash on CBA_{nat} values, average CBA_{nat} within each treatment were pooled before and after the population crashed on the *A. carterae* diet. A significant difference in average CBA_{nat} was revealed for both the *A. carterae* and mixed treatments,

Table 4. Average prosome length (mm) (± SD, 1 standard deviation) of surviving adult *T. californicus* for each food treatment. No results were available for *A. carterae* as there were no surviving adults in this treatment. N = number of individuals measured.

	A. carterae			*T. weissflogii*			Mixed			Natural Food Assemblage		
	N	Length (mm)	± SD	N	Length (mm)	± SD	N	Length (mm)	± SD	N	Length (mm)	± SD
Males	-	-	-	15	1.137	0.266	10	1.088	0.081	57	1.142	0.147
Females	-	-	-	8	0.882	0.066	11	0.979	0.091	54	1.075	0.189

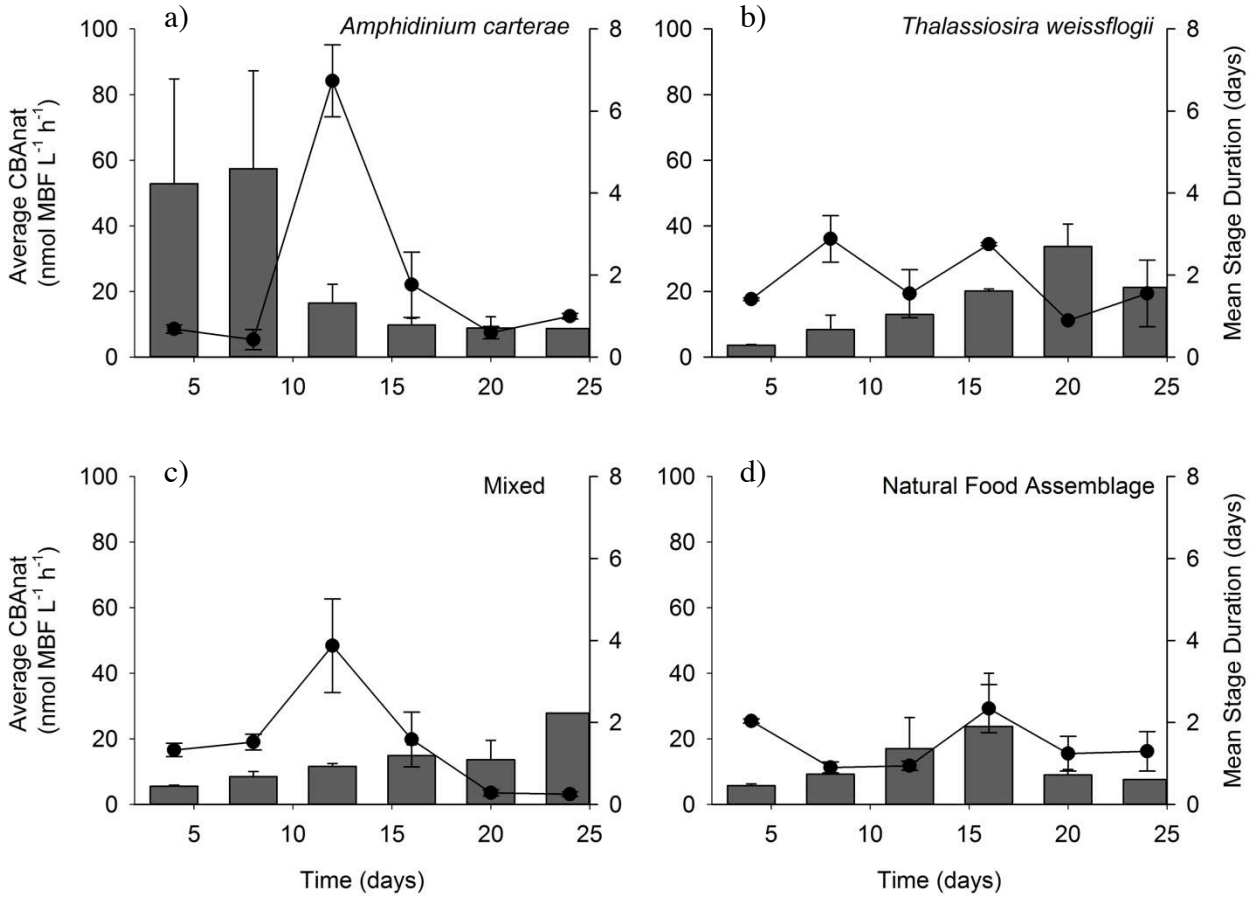


Figure 2. Average CBAnat (left y-axis, lines) and mean stage duration (right y-axis, bars) for *T. californicus* throughout the duration of the experiment for each food treatment. (a) *A. carterae*, (b) *T. weissflogii*, (c) mixed *A. carterae* and *T. weissflogii* and (d) natural food assemblage. Error bars are mean standard error for all replicates.

suggesting that the population crash did influence the average CBA_{nat} within the treatments ($p < 0.05$, Paired t-test).

Average stage duration for the first 10 days of the experiment for copepods fed *A. carterae* was much longer (>4 days) compared to the other three treatments (Fig. 2a). Populations of *T. californicus* fed both the *T. weissflogii* and the mixed *A. carterae*/*T. weissflogii* diet spent a shorter time (<0.5 days) in early developmental stages followed by an increase in stage duration as the experiment progressed (Fig. 2b, c). Average stage duration for copepods fed the natural food assemblage remained below 2 days throughout the experiment (Fig. 2d). Kruskal-Wallis one-way ANOVA on ranks followed by a post hoc Tukey test revealed a significant difference between the longest and shortest mean stage durations in the treatments on Days 4 ($p = 0.025$) and 20 ($p = 0.021$). On a few occasions, no discernable decay could be measured when the individuals in a treatment reached the adult stage as no moulting occurs once the final developmental stage is reached (Knotz et al. 2006) and no stage duration could be calculated.

Copepod production rates were highest (greater than 50 µg C L^{-1} d^{-1}) on the first sampling day of the experiment (Day 4) for the *T. weissflogii* and the natural food assemblage treatments, while copepod production rates of the *A. carterae* diet on this date were substantially lower (Fig. 3). Production rates for copepods fed *A. carterae* diet remained below 40 µg C L^{-1} d^{-1} until Day 12 when estimates peaked to just above 200 µg C L^{-1} d^{-1}. A similar peak (over 100 µg C L^{-1} d^{-1}) was also observed on this date for the mixed treatment. One-way ANOVA revealed statistically significant differences in copepod production rates between the treatments on Day 4 ($p < 0.001$), Day 8 ($p = 0.003$), and Day 12 ($p = 0.003$). Over the duration of the experiment, both total and average

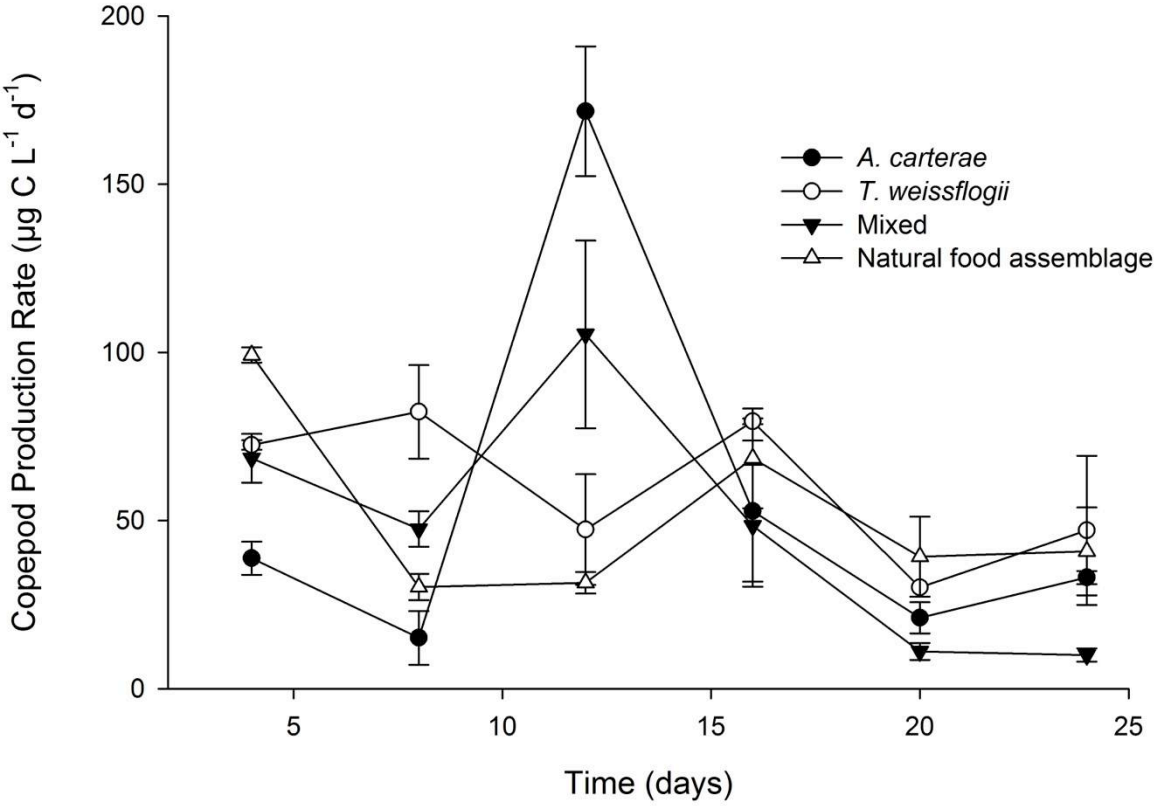


Figure 3. Average copepod production rates (µg C L^{-1} day^{-1}) based on chitobiase decay rates for *T. californicus* throughout the duration of the experiment for each food treatment. Error bars are mean standard error for all replicates.

copepod production rates were highest in the *T. weissflogii* treatment at 358.58 µg C L^{-1} and 59.76 µg C L^{-1} d^{-1}, respectively (Table 5). In contrast, the *A. carterae* treatment had the lowest total (194.72 µg C L^{-1}) and average (32.45 µg C L^{-1} d^{-1}) production compared to the other treatments (Table 5).

In general, our chitobiase-based growth rates were in good agreement with the range of calculated literature-based estimates of weight-specific growth rates (Table 6). Mean *g* (d^{-1}) was higher in the *T. weissflogii* and natural food assemblage treatments than in the *A. carterae* or *A. carterae*/*T. weissflogii* treatments. A comparison of the growth rates between treatments using a one-way ANOVA revealed a statistically significant difference between mean *g* (d^{-1}) in the *T. weissflogii* and *A. carterae* ($p = 0.018$) treatments, in addition to between the *T. weissflogii* and mixed *A. carterae*/*T. weissflogii* treatments ($p = 0.026$).

2.4 Discussion

Using chitobiase-based techniques, this study confirms the findings that a single species algal diet of *T. weissflogii* is a good food item for copepods (Klein Breteler et al. 2005, Ismar et al. 2008, Jónasdóttir et al. 2009), even though this treatment yielded lower overall prosome lengths for surviving adult females. With only one individual surviving the *A. carterae* treatment, our results are in agreement with previous studies, which suggest that the potential toxicity of *A. carterae* results in decreased survival of *Calanus* spp. (Graeve et al. 1994a), *Acartia tonsa* (Kleppel and Burkart 1995), *Centropages hamatus* (Murray and Marcus 2002), *Temora longicornis*, and *Pseudocalanus elongatus* (Koski and Klein Breteler 2003, Koski et al. 2006). A toxic food item, such as

Table 5. Chitobiase-based mean dry weight per individual (µg C), total (µg C L^{-1}) and average (µg C L^{-1} d^{-1}) copepod production rates for *T. californicus* fed with *Amphidinium carterae*, *Thalassiosira weissflogii*, mixed *A. carterae*/*T. weissflogii*, and natural food assemblage (NFA) over the duration of the experiment. Note: values from Day 12 were removed from calculations of total and average copepod production rates for both the *A. carterae* and the mixed treatments.

		Copepod Production Rates	
Treatment	Dry Weight (µg C)	Total (µg C L^{-1})	Average (µg C L^{-1} d^{-1})
A. carterae	50.9	194.72	32.45
T. weissflogii	98.9	358.58	59.76
Mixed	77.2	233.40	38.90
NFA	72.6	309.36	51.56

Table 6. Maximum, minimum, and mean ± SD chitobiase-based daily growth rates (g, d^{-1}) for *Amphidinium carterae*, *Thalassiosira weissflogii*, mixed *A. carterae*/*T. weissflogii*, and natural food assemblage (NFA) over the duration of the experiment. Literature-based estimates of g (d^{-1}) as determined from Table III are given as a comparison. Note: g (d^{-1}) values calculated for Day 12 were removed from all subsequent analyses for both the *A. carterae* and the mixed treatments.

	Chitobiase-based values of g (d^{-1})				Literature values of g (d^{-1})
	A. carterae	*T. weissflogii*	Mixed	NFA	
Maximum	0.69	0.72	0.67	0.71	0.66
Minimum	0.27	0.44	0.35	0.46	0.14
Mean ± SD	0.51 ± 0.09	0.62 ± 0.08	0.51 ± 0.12	0.58 ± 0.08	0.36 ± 0.14

A. carterae, could hinder the ability of nauplii to ingest enough food to grow and thus moult to the next stage, particularly if offered as a single species diet (Klein Breteler et al. 1990). While the mixed diet of *A. carterae*/*T. weissflogii* resulted in higher survival than did the *A. carterae* diet, the number of survivors was substantially lower than in the *T. weissflogii* or natural food assemblage treatments. Therefore, even though ingestion rates were not measured in this study, it is likely that *T. californicus* preferentially ingested *T. weissflogii* over *A. carterae* in the mixed diet (Kleppel and Burkart 1995).

Differences between single versus mixed algal diets were also evident in our estimates of native chitobiase activity (CBA_{nat}). CBA_{nat}, which represents the average sum of all individual growth, has been shown to increase with the number of moulting individuals (Espie and Roff 1995a, Espie ad Roff 1995b, Oosterhuis et al. 2000, Sastri and Dower 2009). In the *T. weissflogii* and natural food assemblage treatments, CBA_{nat} remained relatively constant throughout the experiment. This apparent stability in CBA_{nat} was previously observed in cultures of *Temora longicornis* despite a continuous increase in total biomass and is thought to reflect a balance between chitobiase production and its bacterial degradation (Oosterhuis et al. 2000). The lowest overall CBA_{nat} values were found in the *A. carterae* and mixed treatments, which both showed peaks on Day 12. However, as only one individual was observed in any of the *A. carterae* replicates after Day 12, we speculate that these peaks correspond to a mass mortality of *T. californicus* and likely result from the release of chitobiase from both the apolytic space and the digestive tracts (Peters et al. 1999) of decomposing individuals.

Chitobiase decay rates also provide a correlate of stage duration, or T_{CBA}, of the average-sized individual in the population (Sastri and Dower 2009). With the exception

of the *A. carterae* treatment, all of our treatments fell within the 1-3 days stage duration for *T. californicus* NI to CVI at temperatures of 18-20°C (Powlik et al. 1997). Copepods from the *T. weissflogii* and natural food assemblage treatments reached the adult stage by Day 18, while those in the mixed diet reached the adult stage by Day 22. With the exception of the *A. carterae* treatment, a trend towards increased stage duration throughout the development of *T. californicus* was evident over the course of the experiment (Powlik et al. 1997). In addition, our T_{CBA} estimates showed that copepods fed *A. carterae* spent a significantly longer period of time in the early developmental stages compared to the other three treatments. Similarly, Koski et al. (2006) found that development of *T. longicornis* and *P. elongatus* was slower during the early naupliar stages and stopped altogether at the early copepodite stage when fed *A. carterae*. Given their lower critical food concentration required for growth and thus moulting, the nauplii and copepodites present earlier on during our experiment were likely more sensitive to poor food items than adults (Koski and Klein Breteler 2003; Koski et al. 2006).

Variations in the number of moulting individuals, in addition to the length of time the average individual spends in each stage of development in response to different diets, undoubtedly have an effect on copepod production. Vargas et al. (2010) suggest that carbon is converted into zooplankton biomass at a lower rate when growth is limited by food quality. Our results support this notion given that measurements of copepod production rates in the *A. carterae* treatment were significantly lower than all other treatments, particularly at the beginning of the experiment. Jónasdóttir et al. (2009) found a similar effect of food quality on egg production in *Temora longicornis*, which was highest when fed a 100% *T. weissflogii* diet and lowest on a diet of *A. carterae*. As

mentioned, although the peak in production rates for *T. californicus* fed with *A. carterae* appeared to be delayed until Day 12 in contrast to the other treatments, this peak was likely an artifact of chitobiase being released into the surrounding water by decomposing copepods. Therefore, removing dead individuals during the course of future experiments would prevent the confounding contribution of this source of chitobiase in the seawater samples. In addition, as these findings are novel to this study, future studies should investigate the relationship between mortality and the release of chitobiase into the surrounding water. Indeed, both total and average production for *T. californicus* over the course of the experiment were much higher on the *T. weissflogii* and natural food assemblage diets than they were when copepods were fed *A. carterae* or the mixed diet, thereby illustrating the fact that a poor food item can translate into lower production rates even when the copepods are offered a mixed diet.

Although direct measurements of *T. californicus* growth rate were not made during this study, our indirect estimates of g (d^{-1}) provide a reasonable approximation of the daily weigh-specific growth rate for *T. californicus* over the course of our experiment, thereby demonstrating that the chitobiase enzyme can be effectively used to estimate growth without repeated handling of copepods. Mean g (d^{-1}) for all treatments (ranging between 0.51-0.62 d^{-1}) fell within the high end of the range of g (0.14-0.66 d^{-1}) calculated for *T. californicus* from the literature. Our results indicate that diet does indeed influence growth, as mean g (d^{-1}) was higher when *T. californicus* was fed with *T. weissflogii* and the natural food assemblage compared to the treatments containing *A. carterae*. Specific growth rates for *Pseudocalanus elongatus* fed with *T. weissflogii* have also been shown to be higher than those fed a diet of *A. carterae* (Koski et al. 1998). Future studies would

benefit from a direct measure of growth on individual copepods, coupled with chitobiase estimates, in order to clarify how variations in diet affect copepod growth rates throughout their development from nauplii to adults.

A notable result from our experiment occurred on a few occasions in the *T. weissflogii* (Day 22) and mixed *A. carterae*/*T. weissflogii* (Days 22 and 24) treatments, wherein no detectable decay of the enzyme was measured. On these dates, we observed that adult females had produced egg sacs, however, hatching was delayed and only non-moulting individuals were present in the treatment jars on these sampling dates. In contrast, we did not observe this delay in hatching in the natural food assemblage treatment, wherein moulting individuals were always present in the treatment jars. We speculate that delayed hatching in these instances in the treatments containing *T. weissflogii* may have been influenced by the presence of either inhibitory compounds produced by diatoms (Ban et al. 1997, Vargas et al. 2006) or a lack of essential polyunsaturated fatty acids in the diets containing diatoms (specifically, low levels of DHA) (Evjemo et al. 2008, Jónasdóttir et al. 2009). This notion is further supported by the fact that decay rates were always measureable for the natural food assemblage, which consisted mainly of dino- and nano-flagellates, while diatoms made up only a small fraction of the available food items.

2.5 Conclusions

This study is the first of its kind to use the chitobiase method to follow production rates of developing copepod populations in response to a manipulation of diet. We have shown that this method is sensitive enough to detect variations in the number of moulting

individuals being reared on different diets. Furthermore, our chitobiase-based estimates showed that copepods fed a diet consisting of a (likely) toxic species (*A. carterae*) took longer to develop during the early life stages, had lower productivity, and lower daily growth rates than did the other treatments, which corresponded to our observation that individuals in this treatment failed to survive through the copepodite stages to adulthood. The chitobiase method has promising implications for examining the effects of food quality in a natural field setting without the need for repeated handling of copepods. While we have only included one copepod species, *in situ* measurements of chitobiase production capture the productivity of all members of the crustacean zooplankton community. Both food quality (El-Sabaawi et al. 2009) and chitobiase estimates of zooplankton production rates (Sastri and Dower 2009) have been shown to vary concurrently with the seasonal cycle of the spring phytoplankton bloom and zooplankton biomass, respectively, in the Strait of Georgia, British Columbia. Given that the effects of this variability on field populations remain largely unknown, the next step is to use fatty acid analyses to determine the magnitude of short-term shifts in food quality for copepods and whether such shifts have a significant impact on the production of an entire copepod community.

Chapter 3: Interannual variability in the relationship between in situ estimates of primary productivity and crustacean productivity in Saanich Inlet, British Columbia, Canada

3.1 Introduction

Crustacean zooplankton provide a key link in marine ecosystems between primary producing phytoplankton and higher trophic levels. Knowledge of the efficiency of energy transfer between trophic levels is critical to our understanding of marine food web dynamics. A general assumption in oceanographic studies is that trophic transfer efficiency (TTE) is typically 10% (Lindeman 1942), with a range of 2-24% being observed across most marine ecosystems (Pauly and Christensen 1995). However, TTE has typically been calculated on the basis of biomass estimates alone, which do not necessarily represent the amount of energy available to higher trophic levels. For example, regions with low overall biomass dominated by small crustaceans with fast growth rates may contribute substantially to overall production (Hopcroft and Roff 1998b). Moreover, temporal mismatches may occur when instantaneous estimates of phytoplankton biomass are used to represent the amount of energy available to zooplankton given that zooplankton growth at any point in time is generally the result of the food consumed and environmental conditions encountered days to weeks prior to the time of capture. Therefore, a more accurate way to examine how energy is transferred between phytoplankton and zooplankton requires reliable estimates of both primary and secondary productivity. Although primary productivity in marine ecosystems has been routinely measured since the 1950s (Steeman-Nielsen 1952) estimates of secondary productivity are far less common.

Historically, measurements of secondary productivity have been time-consuming, usually involving incubations of a specific size class of copepods (e.g. Peterson et al. 1991) or the development of artificial cohorts (e.g. Kimmerer and McKinnon 1987). More importantly, these methods have been limited to estimating production for only a single (at most a few) copepod species (e.g. Poulet et al. 1995). Over the last twenty years, a method for obtaining routine estimates of community-level secondary productivity involving measurements of the crustacean moulting enzyme chitobiase has been developed (e.g. Oosterhuis et al. 2000, Sastri and Roff 2000, Knotz et al. 2006). Chitobiase is one of two chitinolytic enzymes responsible for the degradation and reutilization of the exoskeleton during moulting in arthropods (Muzzarelli 1977). Upon moulting, crustaceans release chitobiase into the surrounding water column thereby allowing direct estimates of crustacean production rates to be made by measuring the decay rate of chitobiase activity. Significant relationships between body length (and weight) and the rate of production of chitobiase activity have already been established (Sastri and Dower 2006) and this method has recently been validated in terms of making *in situ* measurements at sea (Sastri and Dower 2006, Sastri and Dower 2009).

While primary producing phytoplankton are mainly controlled by light and nutrient availability, growth and productivity of crustacean zooplankton may be controlled by a combination of factors such as: temperature, body size, food quantity, and food quality. Given that crustacean productivity is a function of both biomass and growth rate (Kimmerer 1987, Huntley and Lopez 1992), changes in the composition of the zooplankton community and resulting biomass will undoubtedly impact production rates. Interannual variations in temperature have been shown to influence the composition of

copepod communities (Greve et al. 2004, Mackas et al. 2012). In addition, increasing water temperatures may lead to a mismatch between phytoplankton and zooplankton by accelerating the peak timing of nauplii compared to the timing of the spring bloom (Sommer et al. 2007), which may result in lower zooplankton biomass and/or variations in the dominant species (Tommasi et al. 2013b). Therefore, the combined effects of temperature variations and a mismatch between spring phytoplankton and zooplankton peaks could potentially decrease the amount of energy transferred from primary producers to zooplankton grazers and, ultimately, throughout the food web (Cushing 1990, Aberle et al. 2012).

Here, we routinely coupled measurements of primary productivity with chitobiase-based production rates for the entire crustacean zooplankton community over two years in a highly productive fjord. Our goal was to determine how productivity of the dominant crustacean zooplankton in the region is influenced by seasonal and interannual variations in the phytoplankton community. In addition, we determined the abiotic and biotic factors best explaining the variation in both primary productivity and crustacean productivity, and examined how these factors influenced the relationship between production rates. Furthermore, fatty acid analyses of *Calanus marshallae* were used to determine the magnitude of short-term shifts in food quality for copepods and whether such shifts have a significant impact on community-level crustacean productivity. Finally, productivity estimates were used to calculate TTE in order to quantify how temporal variations in phytoplankton and zooplankton production rates influence the transfer of energy between these trophic levels.

Results from this study will provide insight into how energy is transferred to higher trophic levels based on the structure of both the phytoplankton and zooplankton communities. Given that the composition and biomass of zooplankton communities are known to exhibit substantial interannual variations (Mackas et al. 2011), knowledge of how variations in phytoplankton dynamics influence the crustacean zooplankton community is crucial to our understanding of how efficiently energy is transferred between lower trophic levels. For instance, the collapse of a biomass-dominant copepod species, *Neocalanus plumchrus* feeding on a diet dominated by diatoms (El-Sabaawi et al. 2009) has been linked to low crustacean productivity in the nearby Strait of Georgia, British Columbia, Canada (Sastri and Dower 2009). High versus low productivity at critical times in a given year (i.e. during the spring) may have significant implications throughout the food web. For example, a decline in growth and survival of the major juvenile fish species has recently been linked to low food production during years of unfavourable oceanic conditions in the Strait of Georgia. (Beamish et al. 2012, Thomson et al. 2012). Therefore, accurate estimates of crustacean productivity are becoming increasingly important in order to determine how temporal and spatial variations in productivity influence fish production, especially in light of the potential impacts of a changing climate on marine ecosystems.

3.2 Methods

3.2.1 Study site

Sampling took place approximately every two weeks from March to August 2010 and 2011 at a single station (48°35′N, 123°30′W) in Saanich Inlet, British Columbia, Canada (Fig. 4). Saanich Inlet is a 24-km-long highly productive fjord with a maximum

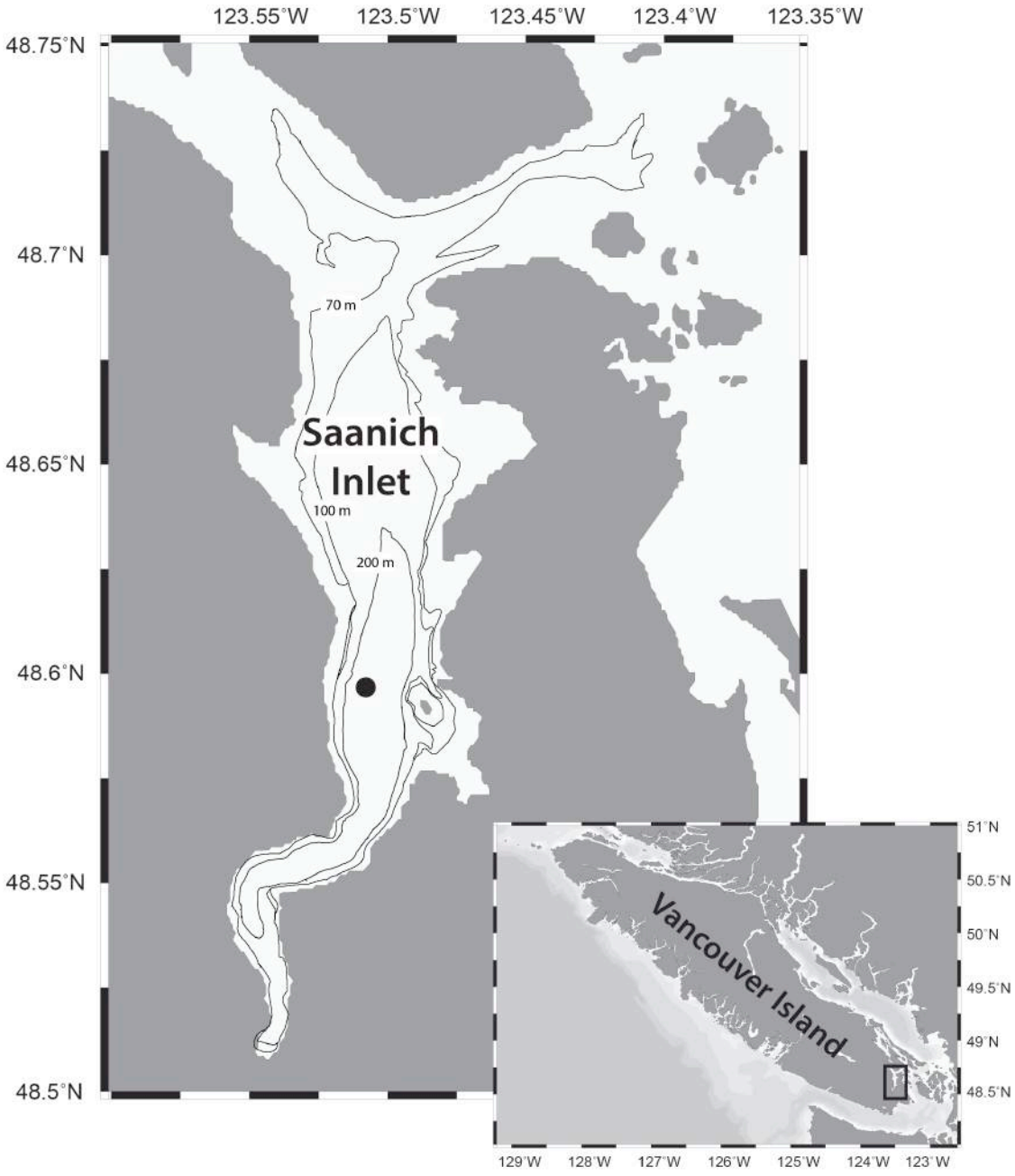


Figure 4. Location of sampling site (48°35′N, 123°30′W) in Saanich Inlet, BC.

depth of approximately 230 m and a shallow (75-80 m deep) sill located at the mouth, which prevents deep water from routinely moving in and out of the inlet (Timothy and Soon 2001, Gargett et al. 2003). Due to the small inputs of the freshwater sources at the head of Saanich Inlet (Goldstream River and Shawnigan Creek), the dominant freshwater sources come from outside of the inlet (Cowichan River during winter and spring; Fraser River during summer) (Takahashi et al. 1977). Saanich Inlet is strongly influenced by tidal flows varying with the fortnightly cycle with the strongest flows occurring during the spring tide and the weakest during neap tides (Gargett et al. 2003). All sampling dates occurred within the first four days of the start of the spring tide in order to capture a seasonal range of plankton dynamics without the confounding influences of sampling during spring versus neap tides. Seasons were defined as follows: late winter (March), spring (April and May), and summer (June to August).

3.2.2 Physical and chemical measurements

Water column temperature, salinity, chlorophyll fluorescence, and dissolved oxygen were recorded on each sampling date from the surface to approximately 10 m above the bottom using a SeaBird Electronics SBE 19+ conductivity, temperature, and depth (CTD) recorder equipped with a SeaBird Electronics SBE 43 dissolved oxygen sensor and a Wet Labs WetStar fluorometer. A stratification parameter, $\Delta \sigma_t$ (kg m^{-3}), was calculated as the difference in density between the surface (average for the top 10 m) and 50 m (Drinkwater and Jones 1987). A depth of 50 m was chosen to represent the "functional bottom depth" of the surface layer, however, similar results were obtained using other depths (i.e. 20 and 100 m) to calculate $\Delta \sigma_t$ as most of the variability occurs

in the surface layers. Furthermore, the surface mixed layer was almost always included in the upper 10 m average. Values of $\Delta \sigma_t$ greater than 1 kg m^{-3} are indicative of a more stratified water column. Average air temperature, wind speed, and precipitation data were provided by Environment Canada's National Climate Data and Information Archive (http://www.climate.weather.gc.ca/) from the meteorological station at the Victoria International Airport, which was the station nearest to our sampling site.

Seawater samples were taken at four different depths within the euphotic zone determined as a percentage of the surface irradiance (i.e., 100% (surface), 60%, 15% and 1%). Sampling depths were determined by calculating extinction coefficients using vertical profiles of photosynthetically active radiation (PAR) collected with a Biospherical QSP-200L on the CTD. The euphotic zone depth was defined as the depth at which PAR decreased to 1% of the surface level. Samples for dissolved inorganic nutrients (i.e., phosphate, nitrate and silicic acid) were filtered through 0.7 μm pore size glass fiber filters and collected in previously acid washed 30 mL polypropylene bottles. Samples were frozen at -20°C for later analysis. The determination of nutrient concentrations was performed spectrophotometrically using an Astoria II nutrient autoanalyzer (Barwell-Clarke and Whitney 1996).

A variable volume of seawater (0.75-1.5 L) was collected for biogenic silica ($bSiO_2$) measurement as a proxy for the biomass of siliceous plankton (mainly diatoms). Water was filtered onto a 0.6 μm pore size polycarbonate filter, dried at 60°C for at least 48 h, and stored in a vacuum desiccator at room temperature until further analysis. An alkaline digestion of the material collected in the filters was performed to convert the $bSiO_2$ into silicic acid ($Si(OH)_4$) (Brzezinski and Nelson 1989). The concentration of the

$Si(OH)_4$ produced in the solution was measured spectrophotometrically using a Beckman DU 530 UV/Vis spectrophotometer as described in Brzezinski and Nelson (1986).

3.2.3 Phytoplankton

Total and size fractionated chlorophyll a

The concentration of chlorophyll *a* was used as a proxy for phytoplankton biomass. To estimate the contribution of different size fractions of phytoplankton, two samples were taken: 1) a volume of seawater (250-500 ml) was filtered onto a 0.7 µm pore size glass fiber filter (GFF) for the calculation of the chlorophyll *a* concentration of the total phytoplankton assemblage, 2) between 250-500 ml of seawater was filtered through a series of polycarbonate filters of 20 µm, 5 µm and 2 µm pore size for the calculation of the chlorophyll *a* concentration of three size fractions (i.e., >20 µm; 20 µm > x > 5 µm; and 5 µm > x > 2 µm). Filters were kept at -20°C until further analysis. Pigments were extracted in 90% acetone over a 24 h period. The fluorescence of the extract was measured using a Turner Designs 10AU field fluorometer as described in Parsons et al. (1984). A correction for phaeopigment interference was done by measuring the fluorescence after acidification of the extract with HCl (1.2 N).

Phytoplankton community composition

Samples were collected in 250 mL amber glass bottles, fixed with Acidified Lugol's solution, and stored at room temperature in the dark until analysis. After gentle homogenization, a 50 mL aliquot was settled in a Utermöhl settling chamber and left to

settle for 24 hours. Abundance of the dominant phytoplankton taxa was determined using an inverted microscope (Utermöhl 1958).

Primary productivity

Samples were transferred from the Niskin bottles used for their collection into acid washed 1 L polycarbonate bottles. They were immediately spiked with ^{15}N-labelled nitrate (Cambridge Isotope Laboratories, +98 atom % ^{15}N) and ^{13}C-labelled bicarbonate (Cambridge Isotope Laboratories, 99 atom % ^{13}C) with a final enrichment target of 10%. Samples were incubated on deck at their corresponding irradiance levels for 4 h in a polycarbonate incubator. *In situ* irradiance was achieved by covering the bottles with different layers of neutral density screening. Surface water was pumped continuously through a tank to maintain a constant temperature. The incubation was terminated by filtering the samples onto pre-combusted 0.7 µm pore size glass fiber filters. Samples were then dried at 60°C for at least 48 h and stored in a desiccator until further processing.

The isotopic ratios of the particulate matter collected on the filters (i.e. ^{14}N:^{15}N and ^{12}C:^{13}C) were measured using a PDZ Europa ANCA-GSL elemental analyzer and a PDZ Europa 20-20 isotope mass spectrometer at the Stable Isotope Facility at the University of California Davis. Carbon uptake rates were calculated following Hama et al. (1983) and nitrate uptake rates were calculated according to Dugdale and Wilkerson (1986, equation (3)). Depth-integrated uptake rates were trapezoidally integrated from the ocean surface to the depth of the euphotic zone. New productivity was estimating by multiplying nitrate uptake rates by the C:N ratio of the particulate matter.

3.2.4 Zooplankton

Community composition

Zooplankton were collected using a SCOR net with a 57 cm diameter mouth and a 236 µm mesh. The net was equipped with a TSK flow meter and hauled vertically from 100 m to the surface at 0.5 m s^{-1}. Contents of the cod end were preserved in 10% borate-buffered formalin. In the lab, samples were split (up to 6 times) using a Folsom splitter. More than 400 copepods were enumerated in each sample. Zooplankton were identified according to taxonomic descriptions provided by the Institute of Ocean Sciences (IOS), Sidney, BC and Fulton (1968). Copepod data are presented at the genus level because we were unable to identify some genera to species level. All other zooplankton were identified to major taxonomic grouping. Abundances (individuals m^{-3}) were converted to biomass (mg m^{-3}) using species-specific length-weight relationships from a database provided by IOS, and a carbon conversation factor of 0.45 (Paffenhöfer and Harris 1976).

Fatty acid analysis

Variations in the dietary fatty acid composition of *Calanus marshallae* was examined on each sampling date. For each sample, 10 stage CV *C. marshallae* were immediately sorted and stored in Cryovials® and kept on dry ice until they were transferred to the lab to be stored at -80°C until analysis. In the lab, sampling followed the protocol outlined in El-Sabaawi et al. (2009). Briefly, copepods were freeze-dried at -40°C for 48 h and placed in 2 mL HPLC-grade chloroform. Samples were flushed with nitrogen gas (N_2), sealed with Teflon®-lined caps, wrapped in Teflon® tape to prevent leakage, and stored at -23°C until extraction. Fatty acids extractions were performed according to Parrish (1999) and Kainz et al. (2004). Samples were sonicated and vortexed

three times in a 4:2:1 chloroform:methanol:water mixture. The organic layers were removed, pooled, and capped off with N_2 in order to prevent degradation of the samples. Fatty acids were analyzed as methyl esters (FAME) that formed using hexane and BF_3-CH_3OH at 85°C for 1 h (Kainz et al. 2004). Esterified fatty acids were analyzed using a gas chromatograph (GC; Varian CP-3800, Varian Inc., Palo Alto, CA, USA) equipped with a flame ionization detector. A Supelco 2560 capillary column (100 m, 0.25 mm inner diameter and 0.2 μm film thickness) was used to compare retention times with known standards. Fatty acid data were converted to % total fatty acids. Given that dinoflagellates are rich in docosahexaenoic acid (22:6n-3, DHA) while diatoms are rich in eicosapentaenoic acid (20:5n-3,EPA), the ratio of DHA:EPA was used as an indicator of the proportion of dinoflagellates to diatoms in the diet (Dalsgaard et al. 2003, Viso and Marty 1993).

Crustacean productivity

Water samples for chitobiase incubations were collected from Niskin bottles at six depths (5, 15, 25, 50, 75, 100 m). Chitobiase decay rates were estimated from 500 mL seawater samples screened with a 40 μm mesh in order to remove any copepods. Approximately 15 mL of the seawater sample from each treatment was immediately filtered (0.2 μm) in order to remove any bacteria and subsequently used to estimate the native *in situ* chitobiase activity (CBA_{nat}). A crude homogenate of 20-30 medium-sized copepods (freshly ground in 3 mL of seawater) was filtered (0.2 μm) and then used to "spike" the original samples from each treatment to differentiate the decay of chitobiase activity (CBA) from background fluorescence (see Sastri and Dower 2006). Seawater

samples were sampled just after homogenate was added (t=0), then every three to six hours thereafter over a 24-hour period. Samples were 0.2 μm filtered, and stored at 4°C in glass tubes until assayed. Samples were maintained at ambient seawater temperature over the 24-hour incubation period.

Measurements of chitobiase activity (CBA) followed Sastri and Dower (2006). Enzyme assays were initiated by adding the substrate 4-methylumbelliferyl-β-D-glucosaminide (0.1 mmol MBF-NAG; Sigma) to seawater samples. Assays were conducted at 25°C and terminated after 60 minutes with the addition of a 2 M NaOH and 0.4 M EDTA solution. Substrate saturation tests were carried out in order to confirm that there was a linear increase in MBF fluorescence over the length of our assay incubations (Sastri and Dower 2006). The reaction was buffered to pH 6.0 (optimal for copepods) using a 0.15 M citrate-phosphate buffer. Chitobiase activity (nmol MBF liberated L^{-1} h^{-1}) was estimated by measuring the fluorescence of the liberated MBF using a Turner BioSystems Modulus Fluorometer with a UV-absorbance (365 nm excitation and 450 nm emission). Raw fluorescence was converted to nmol MBF using a standard curve of known 4-methylumbelliferone concentrations against fluorescence.

Estimates of CBA decay rate (h^{-1}) were calculated as the slope (k) of the natural logarithm of CBA versus time (Sastri and Dower 2006). A Q_{10} correction was applied to the slope in order to account for differences between *in situ* temperature and incubation temperature (Sastri and Dower 2006). The reciprocal of the negative slope ($1/-k$) was used to represent the average stage duration, or the time (T_{CBA}) taken for moulting individuals to produce CBA equivalent to the chitobiase activity (CBA_{nat}). In order to calculate the absolute amount of biomass produced (ΔB), we applied a known

relationship between CBA and the growth increment of marine copepods ($\log(g_{inc}) = 0.864 \log (CBA_i) – 1.78$; Sastri and Dower 2006) to the average CBA_{nat}, the equivalent to the average sum of all individual growth, in each treatment. Depth-integrated daily crustacean production rates (mg C m^{-2} d^{-1}) were then calculated as the biomass production divided by stage duration, or $\Delta B/T_{CBA}$. The ratio of daily crustacean production to the developing biomass was estimated from our corrected values of CBA_{nat} (P:B), which is equivalent to daily growth rate (g, d^{-1}) (Sastri et al. 2012).

3.2.5 Trophic transfer efficiency (TTE)

Although the transfer of energy between phytoplankton and zooplankton is not instantaneous, the time lag between changes in primary and resultant changes in crustacean production rates remains poorly understood (Gladyshev et al. 2011). Therefore, we calculated a moving average of productivity values over three sampling dates (approximately 6 weeks) in order to integrate the variability in our productivity estimates. Time-averaging over three sample dates reduced the occurrence of sampling artifacts (i.e. unreasonably high values of TTE) that result from comparing productivity estimates incorporated over different timescales (Gladyshev et al. 2011). TTE was then calculated as the percent of the time-averaged crustacean production rate divided by time-averaged primary production rate.

3.2.6 Statistical analysis

Best subsets regression was used to select the best-fitting model to explain our time-averaged estimates of primary productivity and crustacean productivity based on a

specified set of explanatory variables (Miller 1990). All explanatory variables were transformed when appropriate using logarithmic (abundance, biomass, and productivity), arcsin-square root (percentage data), or reciprocal (temperature) transformations. Only statistically significant models without multicollinearity were selected based on the highest adjusted-R^2 and lowest mean squared error values. Multiple linear regression was then performed on the explanatory variables represented in the best model. In addition, Pearson's product moment correlation was used to measure the correlation between chitobiase- and net-based values, and between our productivity estimates and TTE. All analyses were performed using Sigmaplot® version 12.3 and R version 3.0.2 (R Development Core Team, 2013).

3.3 Results

3.3.1 Seasonal variation at sampling station

Our sampling program spanned the transition from El Niño (warmer, drier) conditions in early 2010 to La Niña (colder, wetter) conditions beginning in July 2010 and continuing throughout 2011 (Appendix B). Differences in environmental conditions between the two sampling years resulted in slight differences in water column characteristics. Overall, average water column temperatures (to 100 m) were higher in 2010 than in 2011, although both years showed a steady increase in temperature throughout the season with a maximum occurring at the beginning of August (Fig. 5a). Water column salinity was very similar in spring 2011 (ranging from 29.6 to 29.7) and in spring 2010 (ranging from 29.7 to 30.0) (Fig. 5b). However, the water column was slightly more stratified in March and April 2011 as indicated by higher values of the $\Delta\sigma_t$

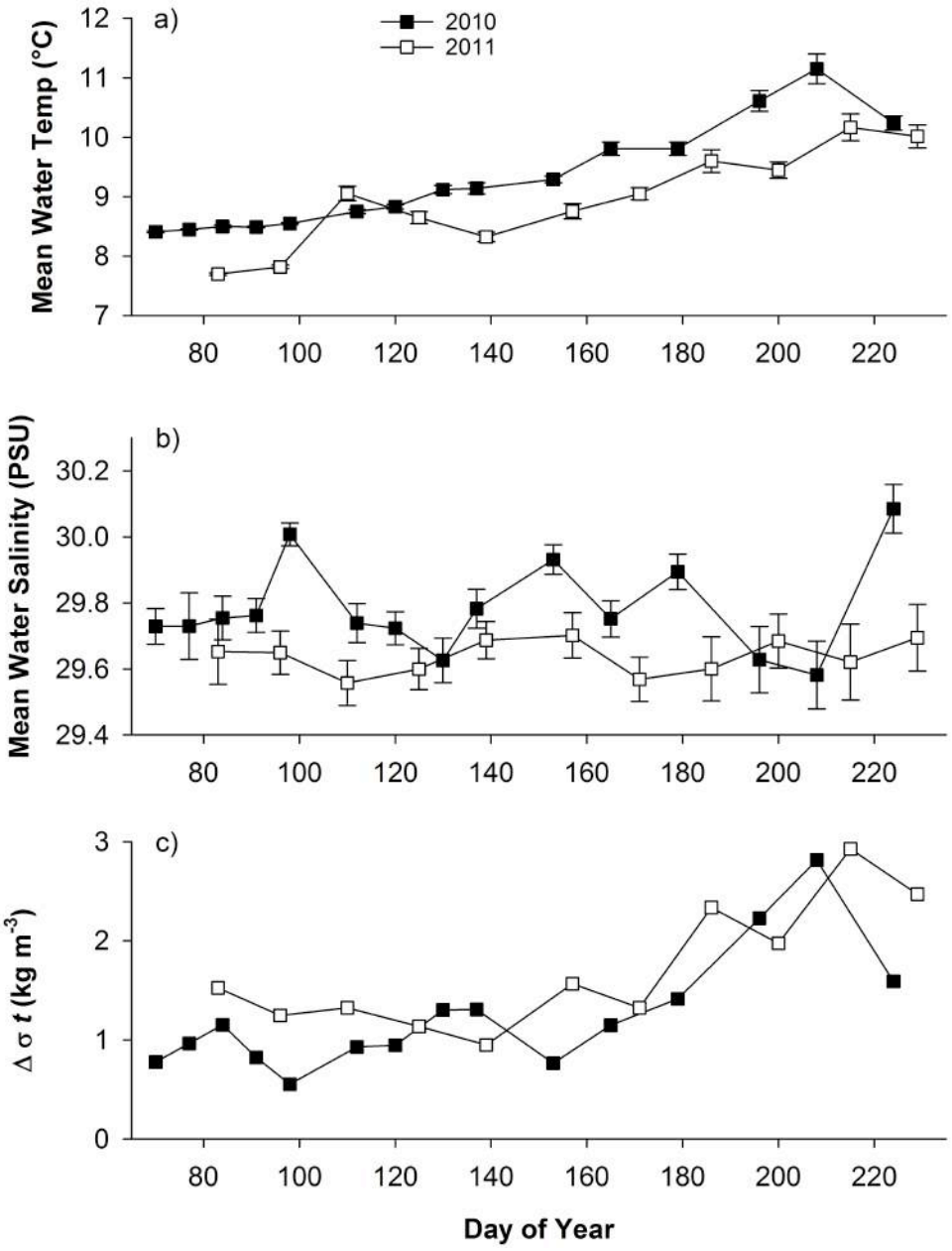


Figure 5. Average water column properties (to 100 m) from March to August for 2010 and 2011. Sigma-t (density) change (*Δ σ t) represents the difference between surface (average from top 10 m) and 50 m. Error bars are ± SE.

stratification parameter (values of 0.6 to 2.8 and 0.9 to 2.9 kg m^{-3} for 2010 and 2011, respectively) (Fig. 5c).

3.3.2 Nutrients

The depth of the euphotic zone (1% light level) varied between 10-20 m throughout the 2010 and 2011 sampling seasons. Overall, NO_3^- and PO_4^{3-} concentrations were lowest at the surface and increased with depth except during the late winter (early March) sampling dates; however, $Si(OH)_4$ was more variable throughout the euphotic zone. During both years, there was a drawdown of all nutrients after day 100 (early April) (Fig. 6). Integrated values of NO_3^- and $Si(OH)_4$ normalized to the depth of the euphotic zone were 24 µmol L^{-1} and 44 µmol L^{-1}, respectively, which were twice as high as values measured at the beginning of our sampling program in 2011 (Fig. 6a,b) In comparison, depth-integrated PO_4^{3-} was only slightly lower on our first sampling date in 2011 (Fig. 6c).

3.3.3. Phytoplankton

Community composition

Flagellates/coccoid cells (less than 5 µm) and dinoflagellates dominated the community in late winter and summer, whereas diatoms were the most abundant phytoplankton group during spring (Fig. 7a,e). In spring 2010, diatoms <20 µm were more abundant than diatoms >20 µm (with the exception of day 130, May 10) (Fig. 7a). In 2011, the highest abundance of large diatoms occurred a few weeks earlier on day 110 (April 20) (Fig. 7e). The most abundant diatom genera during our sampling period were

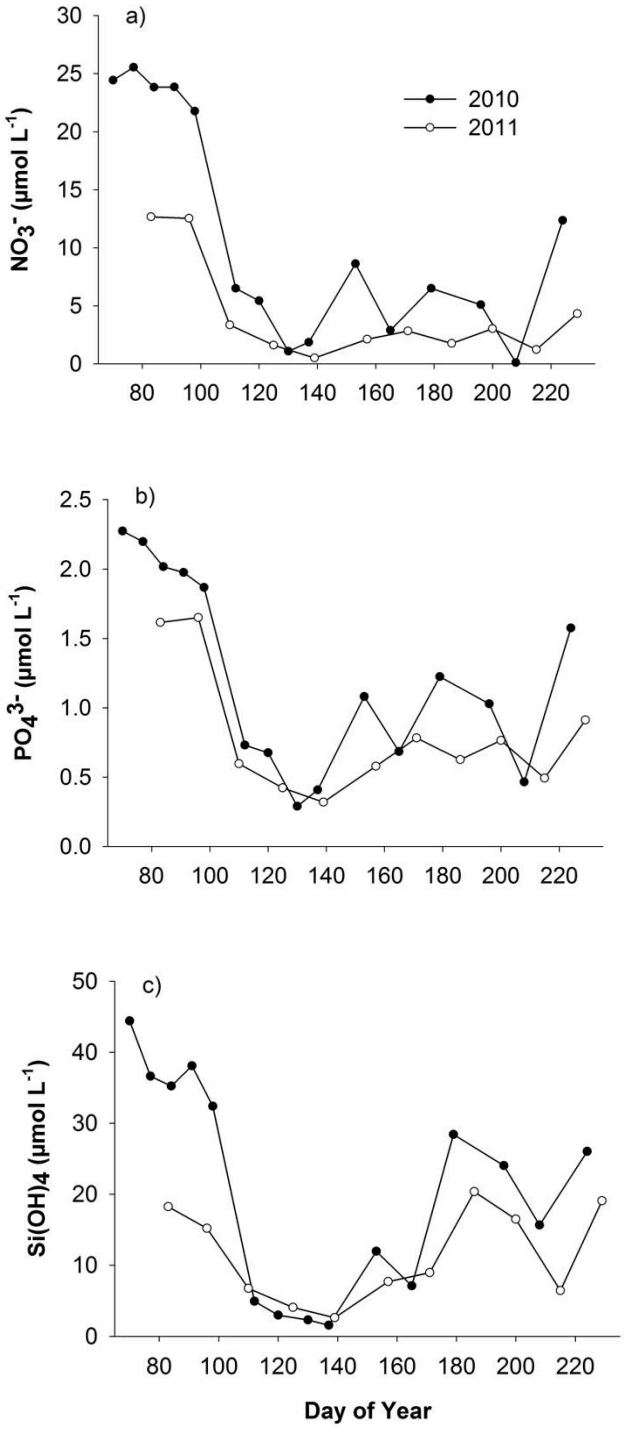


Figure 6. Integrated nutrients a) NO_3^-, b) PO_4^{3-}, and c) $Si(OH)_4$ normalized to euphotic zone depth from March to August 2010 and 2011.

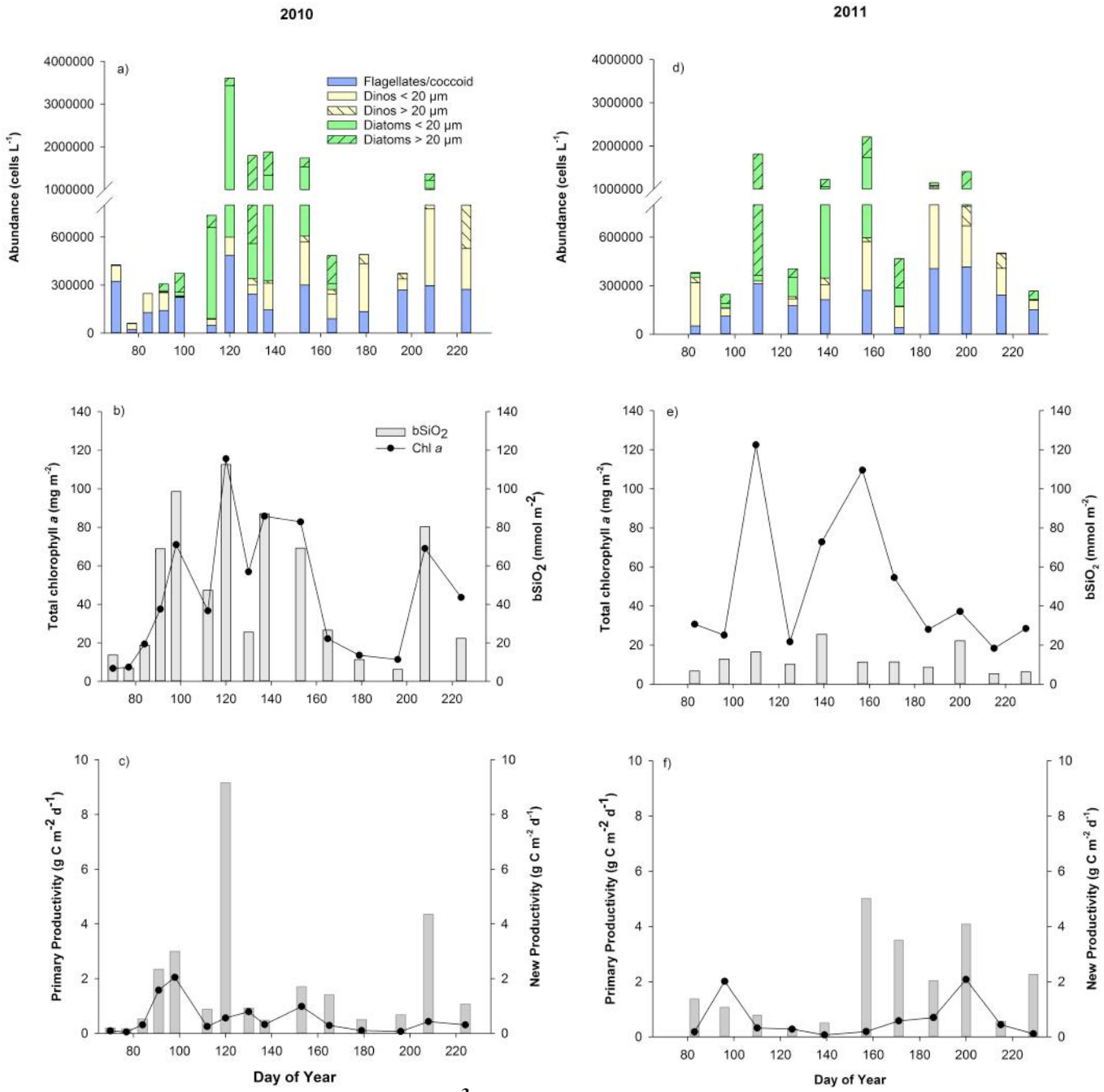


Figure 7. Abundance (individuals m^{-3}) of major taxonomic groups comprising the phytoplankton community (a,d); depth-integrated total chlorophyll *a* (mg m^{-2}; left axis, bars) and $bSiO_2$ (mmol m^{-2}; right axis, lines) (b,e); depth-integrated primary productivity (g C m^{-2} d^{-1}; left axis, bars) and new productivity (g C m^{-2} d^{-1}; right axis, lines) (c,f) throughout the euphotic zone from March to August 2010 and 2011.

Chaetoceros, *Thalassiosira*, *Cylindrotheca*, *Pseudonitzchia*, and *Skeletonema,* while the most common dinoflagellate genera were *Gymnodinium, Prorocentrum*, and *Protoperidinium*.

Chlorophyll a and Biogenic Silica

The depth of the chlorophyll max was typically between 5-10 m throughout the sampling period for both years. The first peak in total depth-integrated chlorophyll *a* occurred on day 98 (April 8) in 2010 and day 110 (April 20) in 2011 (Figs. 7b,e), corresponding to an increase in salinity and temperature in 2010 and 2011 (Fig. 5), respectively. During both years, this initial peak in phytoplankton biomass was followed by a substantial decrease in chlorophyll *a* on the next sampling date. Large phytoplankton (>20 µm) were the dominant size fraction in Saanich Inlet at all times with the >20 µm size fraction making up a larger proportion of the total chlorophyll in spring 2011 than in spring 2010 (Appendix C). Depth-integrated $bSiO_2$ was substantially higher in 2010 compared to 2011, with mean values of 46.36 ± 9.40 mmol m^{-2} in 2010 and 12.40 ± 1.97 mmol m^{-2} in 2011 (Figs. 7b,e).

Primary productivity

Primary productivity peaked on day 120 (9.17 g C m^{-2} d^{-1}) in 2010; however, a similar peak was not observed until day 160 (5.01 g C m^{-2} d^{-1}) in 2011 (Figs. 7c,f). Overall, primary productivity values were much lower during spring 2011 than they were in 2010, but were higher during the summer of 2011 than in 2010. Mean primary productivity over the sampling period was quite similar with values of 1.83 ± 0.60 g C m^{-}

^{2} d^{-1} and 1.95 ± 0.48 g C m^{-2} d^{-1} for 2010, and 2011, respectively. New productivity values followed the same pattern as primary productivity with the highest values observed during spring 2010 and summer 2011 (Figs 7c,f). New productivity ranged between approximately 0.05 and 2.0 g C m^{-2} d^{-1} throughout both sampling seasons. Note that on day 96 in 2011 new productivity values were higher than total productivity and we suspect this was due to sampling error.

3.3.4 Zooplankton

Abundance

Abundances of the copepods *Calanus* and *Corycaeus* were substantially higher in 2010 than in 2011, whereas the abundance of euphausiids was higher in 2011 (Fig. 8a,b). The abundances of other numerically dominant crustaceans (the copepods *Oithona* and *Metridia*) were similar for both years. The initial peak in copepod abundance occurred on approximately day 110 in 2010 compared to the first peak in 2011, which did not occur until after day 140 (Fig. 8a,b). *Calanus* and, to a lesser extent, *Metridia*, peaked multiple times in 2010 compared to a single peak in 2011. Copepods <2 mm length (e.g. *Oithona*) had a lower, albeit steady, abundance throughout our sampling program. In terms of seasonal variation, *Calanus*, *Metridia*, and euphausiids were more abundant in spring, whereas *Corycaeus* was the most abundant crustacean in the summer of both years. The abundance of cnidarians and other jellies (copepod predators) showed a single, high peak in summer 2010 compared to the two peaks observed in 2011 (late spring and summer) (Fig. 8c).

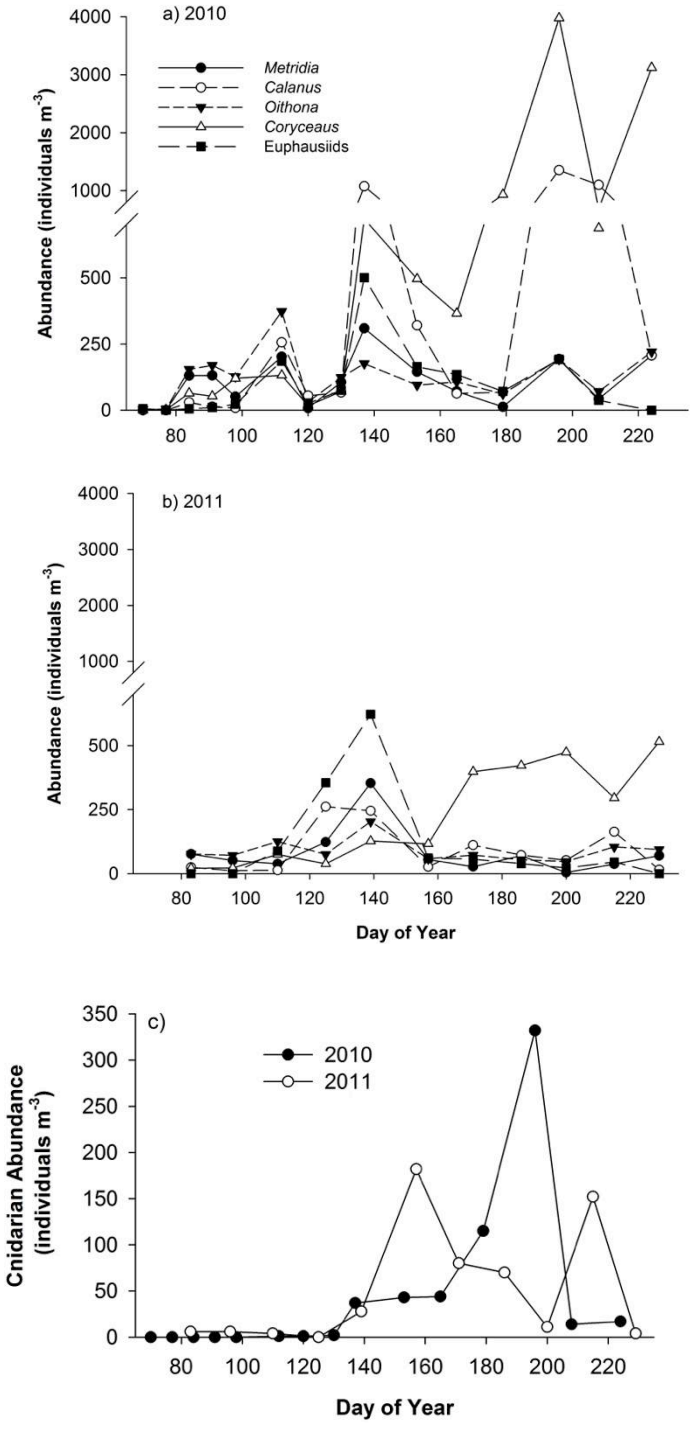


Figure 8. Abundance (individuals m^{-3}) of the dominant crustaceans in 2010 (a) and 2011 (b) and cnidarians (c) collected from 0-100 m vertical net hauls.

Biomass

Zooplankton biomass in Saanich Inlet was dominated by euphausiids, cnidarians (and other jellies), chaetognaths, larvaceans, and the copepods *Calanus*, and *Metridia* (Appendix D). Biomass of moulting crustaceans (nauplii and copepodites) followed similar patterns to crustacean abundance with multiple peaks in biomass occurring in 2010 compared to a single peak in 2011 (Fig. 9a). Although the initial peak in biomass of moulting crustaceans occurring slightly earlier in 2010, the highest adult crustacean biomass occurred at the same time during both years on Day 140 (mid-May) (Fig. 9b).

Fatty acid analysis

In general, the ratios of DHA:EPA in *C. marshallae* showed a similar pattern in both years with a higher ratio of DHA:EPA, reflecting a higher proportion of dinoflagellates in the diet, occurring in late winter/early spring and summer (Fig. 10, Appendix E). In contrast, the lowest DHA:EPA ratios, indicative of a diatom-dominated diet, were observed during the spring. A higher DHA:EPA ratio on Day 80 in 2010 compared to 2011 reveal that *C. marshallae* was feeding on a higher proportion of dinoflagellates in spring 2010 (Fig. 10). Combining data for both years, a significant negative correlation was found between DHA:EPA and *Calanus* abundance ($r = -0.40$, $p < 0.05$) and a near-significant negative correlation was found for DHA:EPA and *Calanus* biomass ($r = -0.36$, $p = 0.07$). However, it should be noted that the power of this correlation was quite low

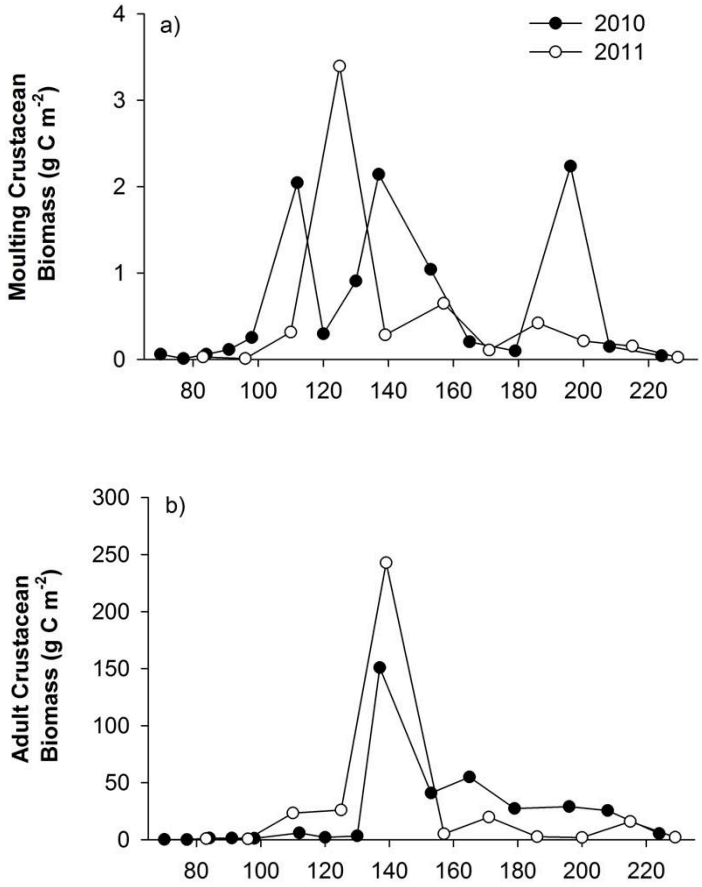


Figure 9. Depth-integrated biomass (g C m^{-2}) of moulting crustaceans (nauplii and copepodites) (a) and adult (i.e. non-moulting) crustaceans (b) from March to August in 2010 and 2011.

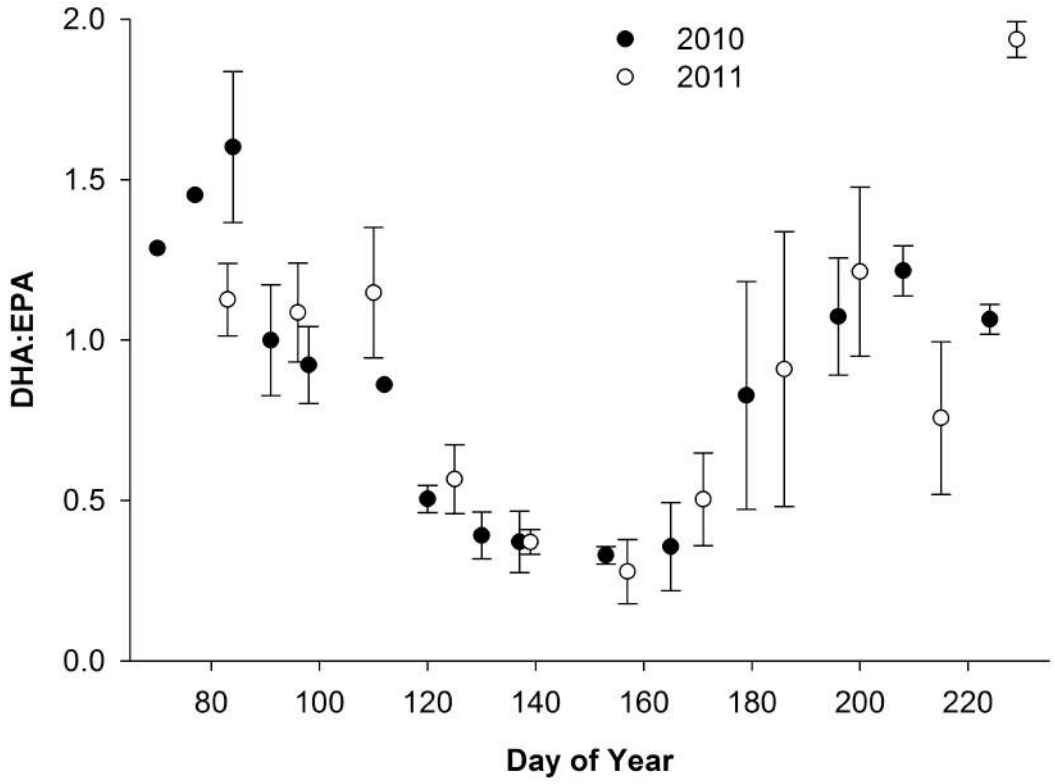


Figure 10. Seasonal variability in the DHA:EPA ratio of *Calanus marshallae* from March to August 2010 and 2011.

(0.44) and thus it is possible that we failed to detect a significant relationship between DHA:EPA and *Calanus* biomass when one actually existed.

Crustacean productivity

The highest CBA_{nat} values were observed in April and June of 2010, and in May of 2011; however, values for both years were similar in March and August (Table 7). The longest mean stage durations were observed in May and June 2010 and in March 2011. The shortest stage duration in 2010 occurred in July, whereas the shortest stage duration in 2011 occurred in May. Daily P:B was higher (faster growth rates) in spring 2010 (March and April) than in 2011. Ranges of daily P:B varied between 0.02 and 0.21 throughout both years.

In general, chitobiase-based estimates of crustacean productivity were higher in 2010 than in 2011 (Fig. 11). One notable exception is the extremely low productivity observed in May 2010 (Fig. 11a) compared to the high productivity values in May 2011 (Fig. 11b). The lowest production rates of 2011 did not occur until June. The lowest production rates in both years corresponded to the lowest DHA:EPA ratios, i.e. the highest proportion of diatoms in the diet of *C. marshallae* (Fig. 10).

3.3.5 Linking primary productivity and crustacean productivity

Best subset regression analysis was used to determine the model containing the explanatory variables which best explained variations in primary productivity and crustacean productivity (Table 8). In 2010, primary productivity was

Table 7. Chitobiase-based estimates of mean monthly CBA_{nat} (nmol L^{-1} h^{-1}), TCBA (stage duration; days), and daily P:B for 2010 and 2011.

	CBAnat (nmol L^{-1} h^{-1})		TCBA (days)		Daily P:B	
	2010	2011	2010	2011	2010	2011
March	3.52	3.40	27.5	49.8	0.10	0.04
April	6.02	4.75	20.0	18.8	0.18	0.07
May	3.62	6.82	55.9	5.6	0.02	0.18
June	6.14	4.22	51.0	34.0	0.21	0.04
July	4.07	5.23	9.0	17.4	0.05	0.06
August	4.92	4.43	19.3	14.3	0.13	0.10

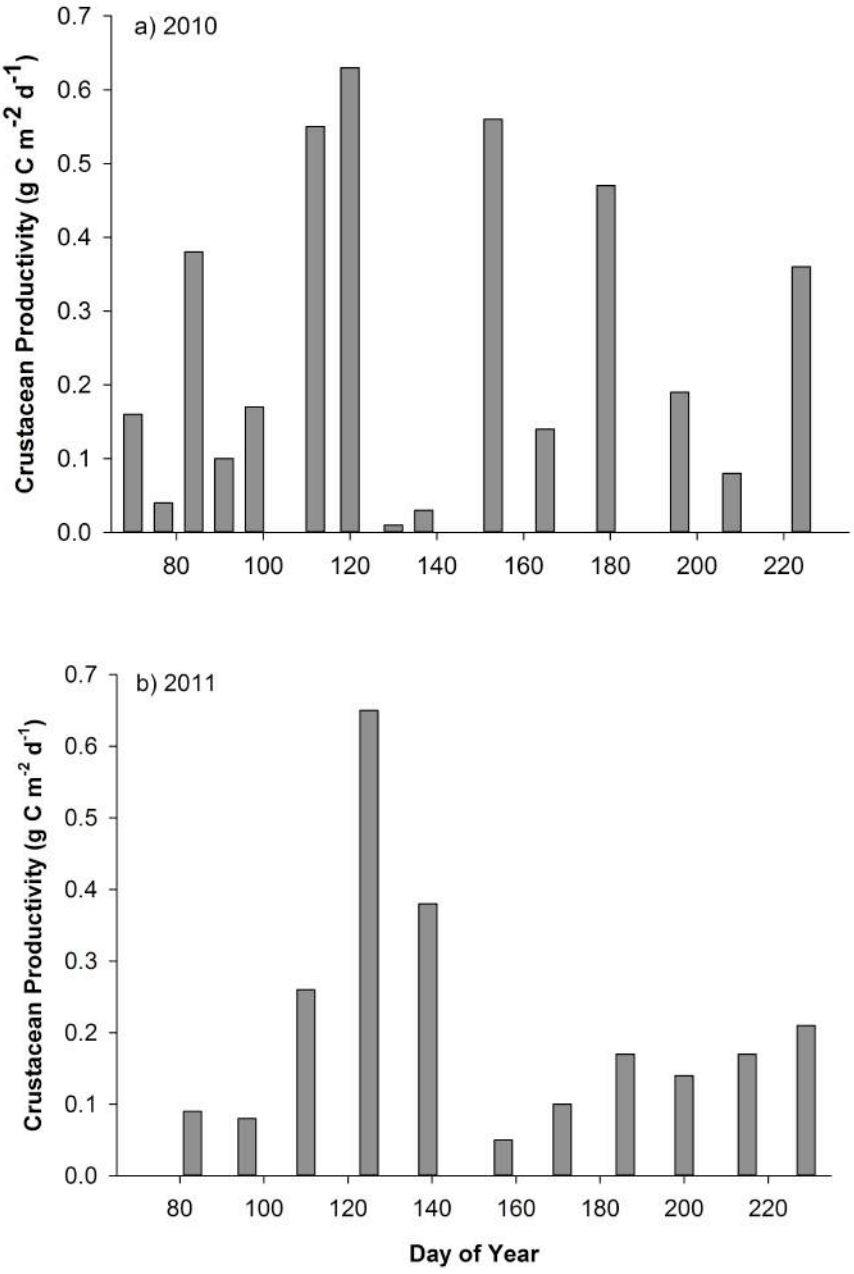


Figure 11. Depth-integrated crustacean productivity (g C m^{-2} d^{-1}) to 100 m for March to August 2010 (a) and 2011 (b).

Table 8. Results of multiple linear regressions and the significance of the models chosen by Best Subsets regression describing the explanatory variables influencing primary productivity and crustacean productivity in 2010 and 2011. Significant values are indicated in bold.

			Coefficient	SE	*P*-value
2010	Primary Productivity	Constant	30.575	22.115	
	N = 15	NO_3^-	0.002	0.001	0.197
	$R^2 = 0.711$	1/Temp	-20.041	11.705	0.118
	Adj $R^2 = 0.596$	Salinity	-0.977	0.730	0.195
		Log_{10}Chl *a*	1.216	0.277	**0.001**
	Crustacean Productivity	Constant	-57.954	26.252	
	N = 15	Salinity	1.964	0.887	**0.049**
	$R^2 = 0.418$	Log_{10}Chl *a*	-0.898	0.495	0.097
	Adj $R^2 = 0.259$	Log_{10}Primary productivity	0.849	0.412	0.064
2011	Primary Productivity	Constant	3.622	1.965	
	N = 11	NO_3^-	-0.005	0.002	0.051
	$R^2 = 0.789$	$Si(OH)_4$	0.002	0.001	0.081
	Adj $R^2 = 0.648$	Log_{10}Crustacean abundance	-1.572	0.584	**0.036**
		Log_{10}Chl *a*	0.874	0.291	**0.024**
	Crustacean Productivity	Constant	0.187	0.553	
	N = 11	Log_{10}Crustacean biomass	0.164	0.062	**0.038**
	$R^2 = 0.915$	Cnidarian abundance	-0.003	0.001	**0.009**
	Adj $R^2 = 0.858$	1/Temp	-12.709	3.959	**0.018**
		Log_{10}Primary productivity	-0.369	0.123	**0.024**

positively related to chlorophyll *a* (Adj $R^2 = 0.60$, $p = 0.001$), whereas in 2011, primary productivity was positively related to chlorophyll *a* ($p < 0.05$) and negatively related to crustacean abundance ($p < 0.05$) (Table 8). Crustacean productivity in 2010 was best predicted by a linear combination of salinity (significant, $p < 0.05$), chlorophyll *a* (non-significant, $p = 0.097$), and primary productivity (non-significant, $p = 0.064$) in 2010 (Adj $R^2 = 0.259$) (Table 8). In 2011, the variables best predicting crustacean productivity were crustacean biomass, cnidarian abundance, temperature, and primary productivity (Adj $R^2 = 0.858$).

Time-averaged values of primary and crustacean productivity were used to calculate TTE throughout the spring and summer of both years (Table 9). In 2010, the highest value of TTE occurred in early March (Day 77, 64%), whereas the highest TTE value for 2011 did not occur late April/early May (Days 110 and 125, 81%). Mean time-averaged TTE across the sampling season was approximately 20% for both years (Table 9).

3.4 Discussion

3.4.1 Timing of spring bloom

Due to the transition from El Niño to La Niña conditions over our sampling program, slightly warmer water temperatures occurred in 2010 compared to 2011 in Saanich Inlet and in the nearby Strait of Georgia (Irvine and Crawford 2012). La Niña conditions (colder, more precipitation) during the winter of 2010-2011 resulted in increased stratification in the early spring of 2011, which may have prevented high concentrations of nutrients from being replenished into the upper

Table 9. Calculations of trophic transfer efficiency based on time-averaged values for primary productivity and crustacean productivity. TTE = crustacean productivity/primary productivity (%) for each sampling date from March to August 2010 and 2011.

	Day of Year	Time-averaged Primary Productivity (g C m^{-2} d^{-1})	Time-averaged Crustacean Productivity (g C m^{-2} d^{-1})	Trophic Transfer Efficiency (%)
2010	77	0.30	0.19	64.24
	84	1.01	0.17	17.03
	91	1.96	0.22	11.02
	100	2.07	0.27	13.24
	110	4.35	0.45	10.37
	121	3.65	0.40	10.90
	129	3.52	0.22	6.33
	140	1.03	0.20	19.18
	152	1.20	0.24	20.21
	166	1.21	0.39	32.19
	180	0.86	0.27	30.81
	194	1.84	0.24	13.28
	209	2.03	0.21	10.24
			Average	**19.93**
2011	96	1.08	0.14	13.46
	110	0.72	0.33	46.15
	125	0.53	0.43	81.46
	140	1.94	0.36	18.56
	156	3.00	0.18	5.93
	171	3.51	0.11	3.09
	186	3.21	0.14	4.33
	200	2.23	0.16	7.16
	215	2.30	0.17	7.51
			Average	**20.85**

water column via vertical mixing as was observed during the previous year. However, despite the differences in nutrient concentrations at the beginning of our sampling seasons, nutrients were not limiting during late winter or early spring in either year and were comparable to those previously reported for Saanich Inlet (Grundle et al. 2009).

Although nutrient drawdown occurred at the same time during both years, the peak in chlorophyll *a* biomass was delayed by approximately two weeks in 2011. Previous studies have found that chlorophyll *a* biomass peaks in Saanich Inlet in both spring (mid-May; Takahashi et al. 1977) and summer (July and August; Grundle et al. 2009). The maximum chlorophyll *a* concentrations in the present study occurred in April and during the summer months (June and July) in both years. Model hindcasting in the Strait of Georgia determined that the spring bloom in 2011 was delayed and more intense than in 2010 due to strong wind events at the end of March/beginning of April in 2011 (Irvine and Crawford 2012). In contrast, our results showed that the magnitude of phytoplankton biomass was similar in both years. This discrepancy between results in nearby water bodies is likely due to the fact that wind forcing influences the timing of the spring bloom in the Strait of Georgia (Collins et al. 2009, Allen and Wolfe 2013), whereas winds play a relatively weak role in Saanich Inlet due to its north/south axis (Gargett et al. 2003).

3.4.2 Phytoplankton community and primary productivity

The seasonal succession of major phytoplankton taxa was similar for both years with the exception of diatoms, which were more abundant during the early spring of 2010 compared to 2011 (also shown by high concentrations of $bSiO_2$). The higher abundance

of diatoms was most likely a result of higher nutrient concentrations, particularly $Si(OH)_4$ (silicic acid), in the upper water column prior to the onset of the spring bloom in 2010. That said, the diatoms present in our samples from early spring 2010 were comprised mainly of the 5-20 µm size range in contrast to the greater proportion of >20 µm diatoms observed in 2011. Given that larger calanoid copepods (>2 mm) peaked earlier in 2010, we suspect that the dominance of diatoms in the 5-20 µm size range was a result of the grazing pressure exerted on the >20 µm diatom cells by herbivorous zooplankton. In terms of the proportion of phytoplankton cells available to grazers, the relative abundances of diatoms and dinoflagellates were the same in April of both years. Although the highest relative abundance of diatoms occurred in May (82%) in 2010, the highest relative abundance of diatoms was not observed until June 2011 (68%). These results were also reflected in our fatty acid analysis of *Calanus marshallae* given that the DHA:EPA ratio varies with the proportions of diatoms and dinoflagellates in the water column (El-Sabaawi et al. 2009). Specifically, *C. marshallae* was feeding on a higher proportion of diatoms (lowest DHA:EPA ratios) in May and June of 2010 and 2011, respectively.

The higher abundance of diatoms in spring 2010 likely contributed to the overall higher primary productivity we observed, which were comparable to those observed during previous El Niño conditions in Saanich Inlet (Timothy and Soon 2001). Our results showed that the >20 µm chlorophyll *a* size fraction was positively correlated to both primary productivity ($r = 0.63$, $p = 0.01$) and new productivity ($r = 0.59$, $p = 0.02$) in 2010, but no correlations between >20 µm chlorophyll *a* and productivity were observed in 2011. In contrast, 2011 primary productivity showed a more typical pattern in Saanich

Inlet in that the highest productivity values were observed during the summer (Grundle et al. 2009). Despite these differences, average primary productivity was the same for both years (1.8 and 2.0 g C m^{-2} d^{-1} in 2010 and 2011, respectively) and only slightly higher than the 1.6 g C m^{-2} d^{-1} average observed by Timothy and Soon (2001) for a station located at the mouth of Saanich Inlet.

3.4.3 Zooplankton community and crustacean productivity

The most notable difference in the zooplankton community between years was the earlier initial peak, and substantially higher abundance, of the larger calanoid copepods (e.g. *Calanus, Metridia*) in 2010. Zooplankton phenology is strongly correlated with early spring temperatures (Edwards and Richardson 2004, Greve et al. 2004, Mackas et al. 2007, Mackas et al. 2012). For example, the development of the dominant Northeast Pacific copepod *Neocalanus plumchrus* shifted by approximately one month with only a 1°C warming (Mackas et al. 2007). It is possible that the higher average water column temperatures in spring 2010 may have triggered earlier developmental cues for crustacean zooplankton. In addition, although our daily P:B (equivalent to daily growth rate) estimates for the planktonic crustacean community fit well with previous estimates for the region (Peterson et al. 2002, Sastri and Dower 2009), higher values of P:B were observed in March and April of 2010 compared to 2011. Although specific growth rates of copepods are known to increase as a function of temperature (Vidal 1980b, Uye 1988, Kimmerer and McKinnon 1987), it is unlikely that the slightly higher temperatures experienced by copepods contributed to the faster growth rates observed in early spring 2010. A more plausible explanation for the observed higher values of daily P:B in 2010

can be attributed to the higher abundance of nauplii and early-stage copepodites, which have faster growth rates than copepods in later developmental stages (Peterson et al. 1991).

The earlier peak in copepods may have contributed to the subsequent increase in overall copepod biomass during the summer of 2010. Delays in the seasonal cycle of zooplankton are known to result in a decrease in total abundance and biomass (Tommasi et al. 2013a). Looking at the copepod community alone, biomass in 2010 was much higher than previous estimates for other parts of the region (Sastri and Dower 2009, Tommasi et al. 2013a) due to the high abundance of larger-bodied *Calanus*. However, overall crustacean biomass was higher in 2011 due to the high numbers of euphausiids. Furthermore, despite the earlier onset of the spring bloom and the zooplankton seasonal cycle in 2010, adult crustacean biomass peaked at approximately the same time during both years, which is probably due to the fact that variations in seasonal biomass are closely linked to the life cycles of the dominant crustaceans (in this case: *Calanus* and euphausiids) (Mackas and Tsuda 1999).

The range of crustacean productivity obtained in this study is similar to the range of chitobiase-based estimates calculated for the Strait of Georgia (Sastri and Dower 2009). Nonetheless, our productivity estimates are much higher than more traditional estimates based on egg production rates (Nielsen and Andersen 2002) or incubations of specific size classes of copepods (Peterson et al. 2002). The discrepancy between these methods is likely a result of chitobiase-based productivity capturing the productivity of all crustaceans in the water column, including the nauplii and juveniles that are too small to be collected in standard zooplankton nets. The use of a 236 μm net may result in

under-sampling and even damaging small zooplankton, which can contribute substantially to overall production given that they often exhibit faster growth rates (Banse 1982, Arendt et al. 2013). Furthermore, although the majority of the zooplankton in Saanich Inlet are located at around 100 m (Devol 1981), sampling during only daylight hours may have slightly underestimated the contribution of vertically migrating species (e.g. *Metridia*) or individuals with the ability to visually detect nets (e.g. euphausiids) to total abundance and biomass. Regardless, crustacean biomass has been shown to vary significantly with CBA_{nat} (Sastri and Dower 2009). We also found significant relationships between CBA_{nat} and net-based biomass of moulting crustaceans (Fig. 12a; slope = 0.11, $R^2 = 0.37$, $p < 0.05$) and between crustacean productivity and net-based biomass of adult crustaceans (Fig. 12b; slope = 0.26, $R^2 = 0.38$, $p < 0.05$). Given that the majority of copepod productivity estimates in the literature are based on egg production rather than somatic production, future studies would benefit from combining measurements of both community-level crustacean productivity using the chitobiase method and production rates based on female egg production in order to assess the biases associated with using these different methods.

It is important to note that the low crustacean productivity observed on a given day in this study did not necessarily correspond to low crustacean biomass. In 2010, the lowest productivity (during May) were associated with high crustacean biomass (particularly of large, non-moulting adults) and also with the highest abundances of >20 μm diatoms. Koski et al. (2010) found that deleterious compounds produced by phytoplankton may have contributed to high mortality rates of the copepods *Temora longicornis* and *Pseudocalanus elongatus* during periods of high chlorophyll *a*

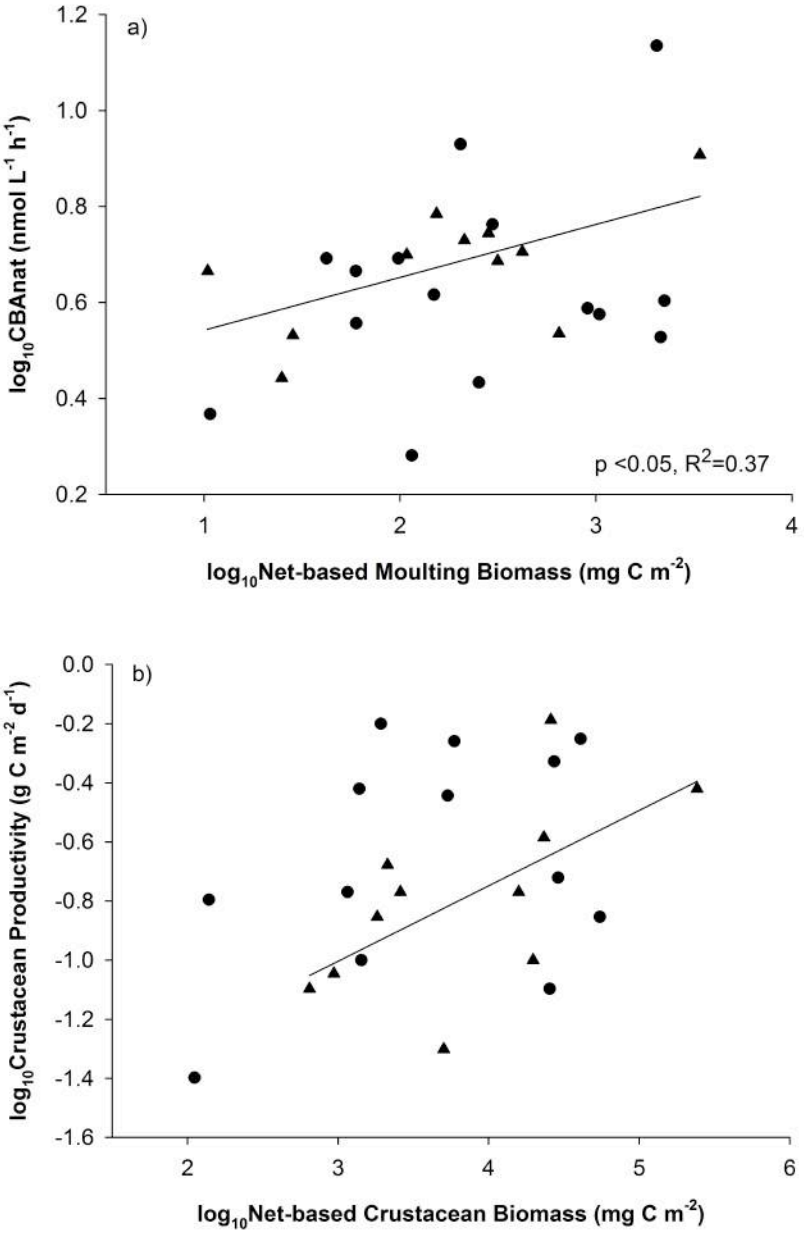


Figure 12. Relationships between (a) CBA_{nat} and net-based moulting biomass and (b) crustacean productivity and net-based adult crustacean biomass across both years (circles: 2010 values; triangles: 2011 values). Solid lines are linear regressions.

concentrations. We also suspect a link between the observed low productivity and what the copepods were feeding on at the time, in agreement with a previous lab study which showed a link between low or even undetectable chitobiase productivity and a diatom-dominated diet (Suchy et al. 2013). Field studies have shown that moulting failure in copepodites resulting in low crustacean productivity was correlated with low DHA:EPA ratios during the 2005 spring bloom in the Strait of Georgia (El-Sabaawi et al. 2009, Sastri and Dower 2009). Nutrient limitation in diatoms may cause a decrease in growth rate and inhibit copepod development altogether (Klein Breteler et al. 2005), which could have negatively influenced crustacean productivity in the Strait of Georgia (Sastri and Dower 2009). Low crustacean productivity in our study occurred on or shortly after the period of lowest nutrient concentrations during the spring bloom, suggesting that nutrient-limited diatoms may have had an adverse effect on copepod growth in Saanich Inlet. In addition, low phytoplankton growth rates were observed just prior to the extremely low crustacean productivity values (Appendix F). Given that diatoms with faster growth rates have been shown to result in increased reproductive success of copepods (Jónasdóttir and Kiørboe 1996), it is possible that productivity was negatively affected when crustaceans were feeding on phytoplankton with slow growth rates. That said, the lowest crustacean productivity in 2011 did coincide with low crustacean biomass, which may have been due to intense predation pressure by cnidarians. Therefore, it is possible that top-down processes may, at times, confound the ability to detect effects of diet on productivity in a natural field setting.

3.4.4 Factors influencing primary and crustacean productivity

Chlorophyll *a* was the strongest variable to influence primary productivity in both years with increasing values of primary productivity occurring during periods of higher phytoplankton biomass. In 2010, this pattern was extended to the zooplankton as we observed a positive relationship between time-averaged values of primary productivity and crustacean productivity (Fig. 13a; slope = 0.06, $R^2 = 0.35$, $p < 0.05$) thereby suggesting bottom-up processes were controlling energy transfer to higher trophic levels. In contrast, a negative relationship was found between primary productivity and crustacean productivity in 2011 (Fig. 13b; slope = -0.14, $R^2 = 0.51$, $p < 0.05$), possibly due to intense grazing pressure on copepods by cnidarian predators. Numerous studies have shown that copepod populations may be largely controlled by the abundance of dominant invertebrate predators (e.g. jellies and chaetognaths) (Davis 1984, Hirst et al. 2007, Ji et al. 2013). Furthermore, top-down control by zooplankton has already been suggested to contribute to periods of low primary productivity in Saanich Inlet (Grundle et al. 2009) and the Strait of Georgia (Harrison et al. 1983). Therefore, it is possible that the phytoplankton community in spring 2010 was more resistant to top-down control due to the high productivity occurring during the spring (Frank et al. 2006). Although bottom-up processes are assumed to be the dominant control for marine ecosystems (see Daewel et al. 2014 for review), these results are often based on broad-scale studies or long-term datasets (Ware and Thomson 2005, Frank et al. 2006). Our results support the suggestion by Frank et al. (2006) that smaller-scale examinations of the temporal variability in ecosystem dynamics may reveal both bottom-up and top-down control.

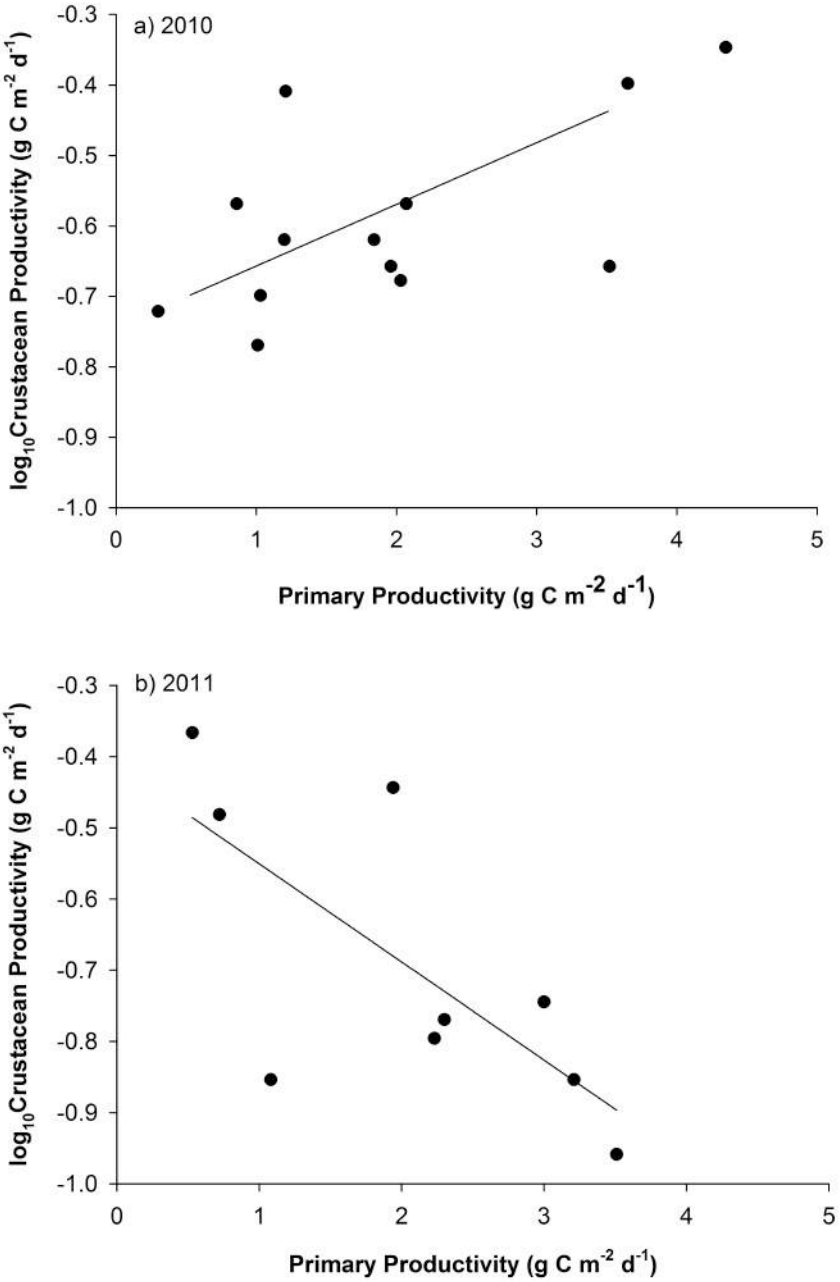


Figure 13. Relationships between time-averaged crustacean productivity and primary productivity in 2010 (a) and 2011 (b). Solid lines are linear regressions.

3.4.5 Trophic transfer efficiency

By coupling time-averaged estimates of primary productivity with chitobiase-based crustacean productivity, our calculations of TTE show that energy is transferred between phytoplankton and zooplankton at an average efficiency of ~20% in Saanich Inlet, thereby falling within the upper range predicted for most marine ecosystems (Pauly and Christensen 1995). The average values of TTE that we calculated are also very close to the 23% transfer efficiency calculated for the nearby Strait of Georgia (Sastri and Dower, 2009). While the assumption of a 10-20% TTE is useful on an annual basis, our results showed substantial variation from early spring to summer. For instance, TTE varied by a factor of 10 (6.33% to 64.24%) in 2010, and by a factor of 26 (3.09% to 81.46%) in 2011. Despite the large range of within-season variability, the high maximum values of TTE we observed in Saanich Inlet are similar to the maximum values of transfer efficiency (74.6%) reported in a similar study for Sagami Bay, Japan (Ara and Hiromi 2007).

Significant, non-linear relationships were found between TTE and primary productivity in Saanich Inlet with TTE decreasing exponentially with increasing primary productivity ($R^2 = 0.82$, $p < 0.0001$) (Fig. 14). Similarly, TTE in Sagami Bay was controlled by primary productivity when productivity was lower than 0.5 g C m^{-2} d^{-1} (Ara and Hiromi 2007). Our results indicate that TTE was relatively constant when primary productivity was higher (>2 g C m^{-2} d^{-1}) because the zooplankton grazers were unable to keep up with primary productivity due to either the crustaceans being food saturated (e.g. 2010) or because of low crustacean biomass during periods of high primary productivity (e.g. 2011). Relationships between TTE and crustacean productivity were not as clear

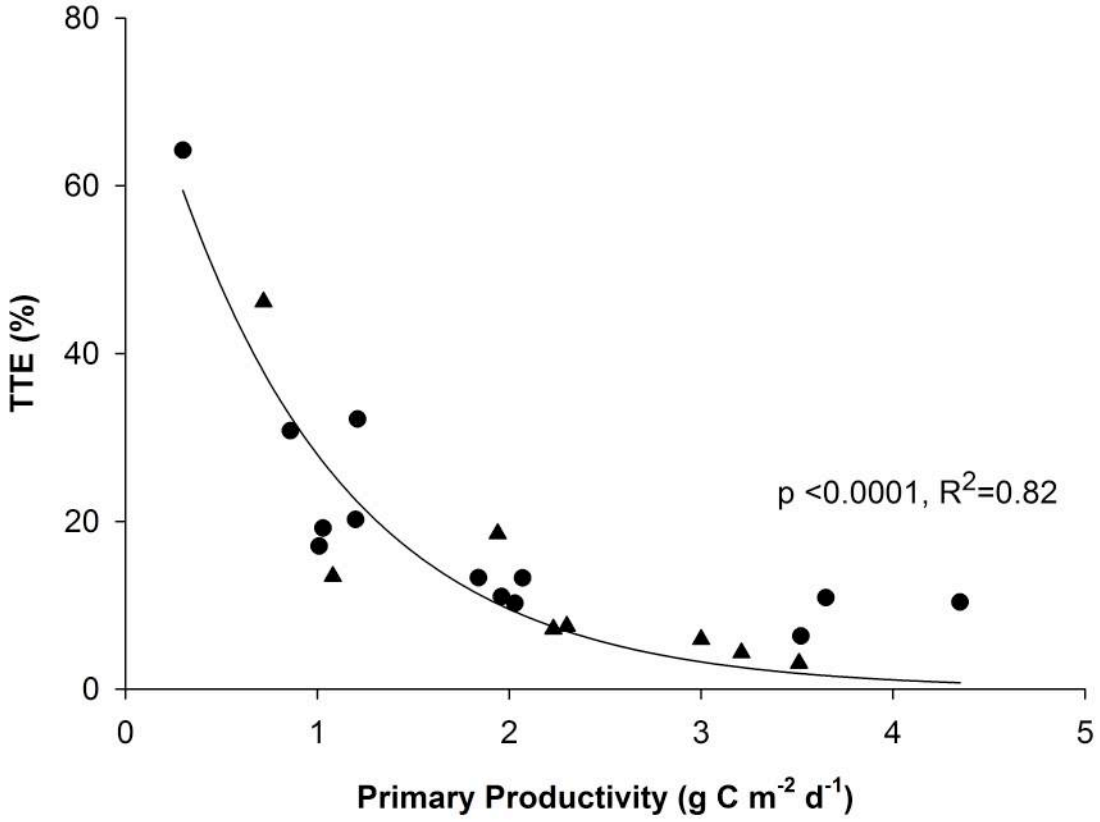


Figure 14. Relationship between time-averaged values of TTE and primary productivity for both years (circles: 2010 values; triangles: 2011 values). The solid line is a non-linear regression.

given that a significant correlation was found in 2011 ($r = 0.67$, $p < 0.01$), but not in 2010. It is important to note that our TTE estimates assume that primary productivity from phytoplankton is the only energy available to support crustacean productivity. However, given that we know heterotrophic protozoa (e.g. ciliates and dinoflagellates) can comprise a significant proportion of copepod diets (Liu et al. 2005, Vargas et al. 2007, Yang et al. 2009) and thus make a significant contribution to copepod productivity, our estimates of TTE may be slightly inflated. Therefore, field studies incorporating crustacean productivity and estimates of both ciliate and primary productivity are necessary in order to determine the relative contributions of the classical food chain and the microbial loop to TTE. Moreover, future studies are needed to extrapolate our results to higher trophic levels in order to determine the impact of variability in TTE at more crucial times during the year, i.e. during the spring when the match/mismatch between predators and their prey is more important (Cushing 1990).

3.4.6 Conclusions

This study is the first of its kind to routinely couple *in situ* production rates for phytoplankton and crustacean zooplankton. Our results show that using biomass estimates alone may lead to misleading interpretations of ecosystem dynamics. For example, the amplitudes of phytoplankton and zooplankton biomass were similar in both 2010 and 2011, yet productivity (particularly primary productivity) estimates for these two trophic levels were different between years. Had we used biomass estimates to calculate productivity in the more traditional sense, the temporal and interannual variability of productivity in Saanich Inlet would have been overlooked. Furthermore, by

using *in situ* productivity estimates, we were able to determine that short-term variations in the transfer of energy to higher trophic levels in Saanich Inlet may be controlled by both bottom-up and top-down processes.

The fact that our chitobiase-based estimates of crustacean productivity and TTE fit with current estimates for coastal marine ecosystems (Pauly and Christensen 1995) provides further support for the use of this method, which is less time-consuming and more practical in terms of obtaining routine estimates in a field setting. However, because the chitobiase method captures all of the enzyme produced in the surrounding water, potential uncertainties in our estimates may exist. For example, we were unable to consider the contribution of chitobiase released from dead or recently preyed upon crustaceans, which may have lead to a slight overestimation in crustacean production and resulting TTE calculations. Chitobiase-based productivity estimates are directly applicable and critical for ecosystem modeling and ecosystem-based fisheries management practices, which have typically relied on bulk zooplankton biomass estimates alone. Previous research has already shown that growth/survival of juvenile fish can be linked to mismatches in the peak timing of copepod prey (Mackas et al. 2007), overall copepod size (Beaugrand et al. 2003), and copepod production rates (based on egg production) (Castonguay et al. 2008). Our results highlight the importance of using field-derived crustacean production rates given that low productivity can occur even when zooplankton biomass is high. Ultimately, accurate estimates of both crustacean productivity and TTE will be particularly critical in terms of investigating the impact of an increasing occurrence of mismatches between lower and higher trophic levels in predicted future warming scenarios.

Chapter 4: Community-level crustacean zooplankton productivity in the tropical Guanabara Bay, Brazil

4.1 Introduction

Examining energy transfer between trophic levels is critical to our understanding of marine food webs. Estimates of zooplankton production, i.e. the rate of biomass generated per unit time, rather than biomass estimates alone, are crucial when examining trophic dynamics (Longhurst 1984) because they reveal how much energy is being transferred from phytoplankton to zooplankton and, ultimately, the amount of energy available to higher trophic levels. Compared to measurements of primary production, there is still a lack of consensus as to how zooplankton production should be measured. Historically, measurements of zooplankton production have relied on time-consuming incubations of specific size classes or cohorts of copepods (Kimmerer and McKinnon 1987, Peterson et al. 1991), which are impractical in a natural field setting. Alternatively, global mathematical models (e.g. Huntley and Lopez 1992, Hirst and Lampitt 1998, Hirst and Bunker 2003) have become increasingly popular for estimating copepod growth rates and production because they allow for broad scale temporal and spatial estimates of zooplankton production with relatively little effort compared to the more traditional methods.

More recently, the chitobiase method had been validated for routinely and rapidly estimating *in situ* community-level crustacean productivity at sea (Sastri and Dower 2006, Sastri and Dower 2009). Chitobiase, a crustacean moulting enzyme, is released into the surrounding water upon moulting (Oosterhuis et al. 2000, Sastri and Roff 2000, Knotz et al. 2006). Measurements of the decay rate of chitobiase activity (CBA) in the water

column can then be used to directly estimate crustacean productivity. Significant relationships between body length (and weight) and the rate of production of chitobiase activity have already been established for marine copepods in both temperate (Sastri and Dower, 2006) and subtropical regions (Avila et al. 2011). The major benefit of using this enzymatic approach is that it avoids problems associated with net selectivity in underestimating abundance and biomass, the repeated handling of animals, and the difficulties associated with identifying and measuring small individuals. Therefore, the chitobiase method takes into account the productivity of all planktonic crustaceans – including those that are too small (e.g. nauplii) or too large (e.g. krill) to be effectively sampled by traditional plankton nets.

The majority of zooplankton production estimates comes from mid- to high-latitudes dominated by large-bodied copepods with annual life cycles. Compared to temperate regions, studies of zooplankton dynamics in the tropics are sparse and are often limited to records of species abundance and biomass (e.g. Moore and Sander 1976, Youngbluth 1980, Yoshioka et al. 1985). Given that the copepod assemblage is typically dominated by small (<1 mm) individuals, estimates of abundance and biomass in tropical regions are confounded by the choice of the mesh size used to collect zooplankton, potentially resulting in severe underestimations of the contribution of smaller species (Hopcroft et al. 1998b, Gallienne and Robins 2001, Avila et al. 2012). Furthermore, although tropical regions have lower overall zooplankton biomass, the high growth rates exhibited by small copepods with multi-generational annual life cycles can still contribute to high amounts of production (Hopcroft and Roff 1998b).

Some of the most notable studies in the tropics have provided a comprehensive examination of naupliar (Hopcroft and Roff 1998b), copepodite (Hopcroft et al. 1998b), and general copepod production in the coastal waters of the Caribbean (Hopcroft and Roff 1998a, Hopcroft et al. 1998a, Hopcroft et al. 2001). Studies of copepod production in food-limiting (e.g. oceanic) tropical ecosystems, on the other hand, are even more rare (Webber and Roff 1990, McKinnon and Duggan 2003). While these studies have led to a clearer understanding of copepod production in tropical regions, the magnitude by which production is underestimated because of the historical reliance on traditional incubation methods or net-based biomass estimates is unknown.

The overall aim of this study was to use the chitobiase method to obtain routine estimates of community-level crustacean productivity for the highly eutrophic Guanabara Bay, Rio de Janeiro, Brazil. Chitobiase-based community-level productivity has recently been estimated for Patos Lagoon estuary in southern Brazil (Avila et al. 2011, Avila et al. 2012); however, despite numerous recent studies on population dynamics of zooplankton in Guanabara Bay (e.g. Marazzo and Valentin 2000, Marazzo and Valentin 2001, Schwamborn et al. 2004), crustacean production has yet to be studied in the region. Our main objective was to determine the abiotic and biotic factors most strongly influencing crustacean productivity in Guanabara Bay. Given the dynamic nature of this water body, sampling was conducted across different timescales (monthly, weekly, daily) over a three-month period in order to capture a range of hydrological conditions. In addition, chitobiase-based productivity values were compared with previous estimates for tropical regions and with those derived from global mathematical models using biomass estimates from nets of different mesh sizes. Results from this study provide the first routine

analysis of community-level crustacean productivity for tropical coastal waters. Given that small copepods, including nauplii and copepodites, provide a key link between the microbial food web and fish larvae or other planktivores (Hopcroft et al. 2001, Turner 2004), accurate estimates of productivity are imperative in terms of understanding trophic relationships in tropical regions. Ultimately, these results will provide insight as to how much energy is potentially available to higher trophic levels in tropical regions.

4.2 Methods

4.2.1 Study site

Sampling was conducted over different timescales at a single site (22°54'11" S, 43°09'29" W) near the entrance to Guanabara Bay, Rio de Janeiro, Brazil (Fig. 15) from April to June, 2012. Guanabara Bay is a highly eutrophic, sub-tropical coastal bay characterized by high levels of pollution due to a large input of untreated domestic and industrial waste (Kjerfve et al. 1997). Water is exchanged throughout the bay during the tidal cycles with coastal water being brought in via a Central Channel with a depth of 30 m (Kjerfve et al. 1997, Paranhos et al. 1998, Schwamborn et al. 2004), however, the average water depth of the entire bay is 5.7 m (Kjerfve et al. 1997). Guanabara Bay is influenced by the fortnightly tidal cycle as well as semidiurnal forcing; however, tidal ranges and currents are small in comparison to other coastal bays (Kjerfve et al. 1997).

4.2.2 Physical and biological measurements

Water column temperature, salinity, and dissolved oxygen were recorded on each sampling date from just above the seafloor (~20 m) to the surface with a Seacat SBE-19

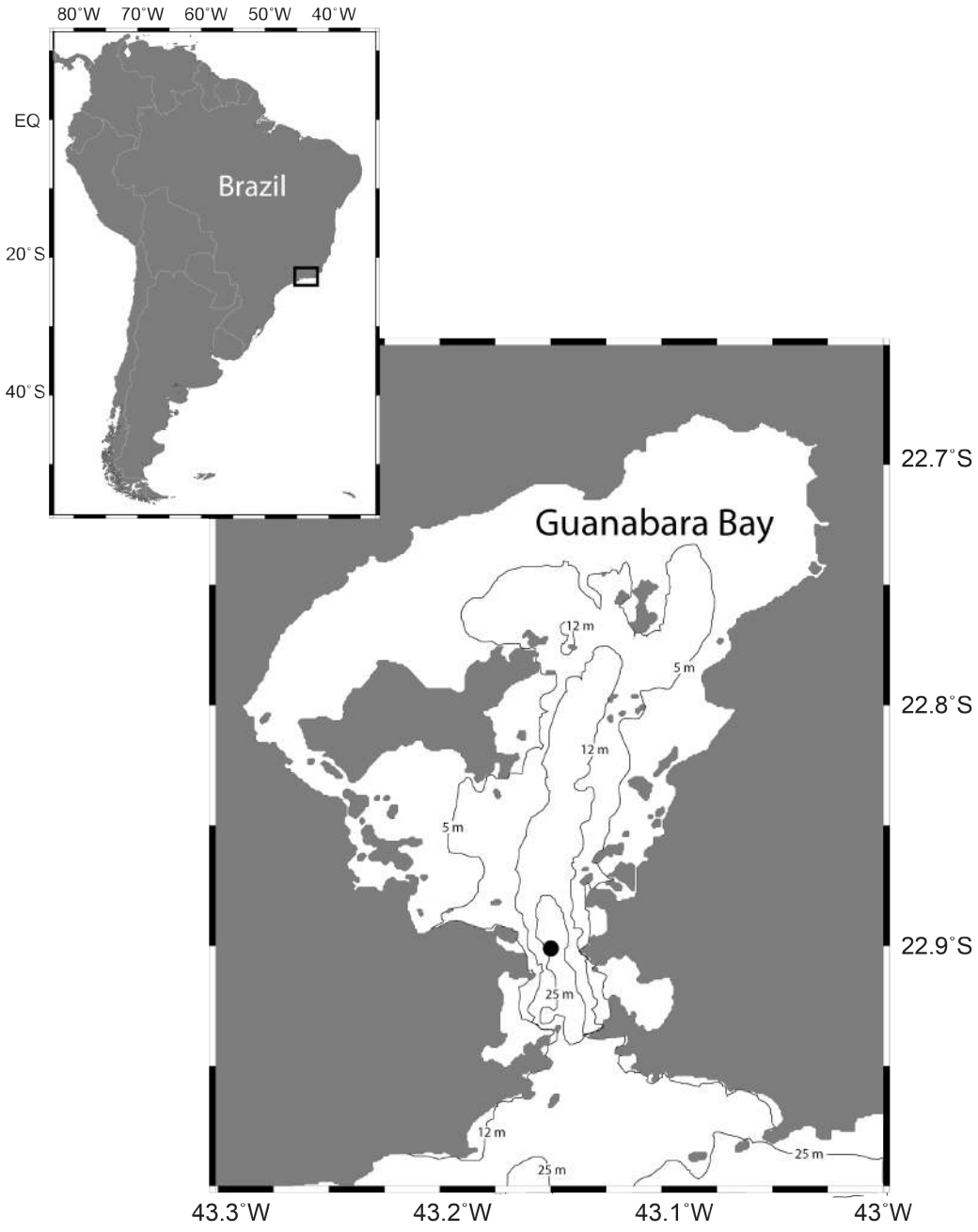


Figure 15. Location of sampling site in Guanabara Bay, Rio de Janeiro, Brazil.

CTD (Seabird Inc.). Average air temperature, wind speed, and precipitation data were provided by the Government of Rio de Janeiro (http://www.simerj.com) for the meteorological stations nearest our sampling site. Hourly tides were obtained from the National Oceanographic Database provided by the Brazilian Marines. Seawater samples for total nitrogen, particular organic carbon (POC), and chlorophyll *a* (a proxy for phytoplankton biomass) were collected from a single depth near the surface (1 m). For POC and total nitrogen, samples were measured on pre-combusted (550°C for 2 h) GF/F filters after filtration of 200 mL of seawater. Filters were analyzed on an elemental analyzer CHN in the Geochemistry Laboratory at the Rio de Janeiro State University.

For chlorophyll *a* analysis, a variable volume (50-300 mL) of total and <20 µm size-fractionated (filtered through Nitex mesh) seawater was filtered onto a 0.7 µm pore size, 47 mm diameter glass fiber filters (GF/F) under low pressure and stored at -20°C until further analysis. Pigments were extracted in 90% acetone and measured with fluorometric techniques on a Varian Cary Eclipse® spectrofluorometer. Chlorophyll and phaeopigments were assessed using a modified version of the Neveux and Lantoine (1993) method. Modifications were as follows: 1) data acquisition was performed by recording the fluorescence emission spectra for each of 31 excitation wavelengths (3-nm increments from 390 to 480 nm). Emission spectra were recorded at 2 nm intervals from 615 to 715 nm, yielding 51 data points for each spectrum. Pigment concentrations were estimated from the resulting 1581 data points, and 2) where the least squares approximation technique was constrained to discard negative solutions as described in Tenório et al. (2005).

4.2.3 Zooplankton community composition

Zooplankton samples were collected using a 64 μm and a 200 μm mesh ring net with an attached flowmeter hauled vertically from just above the sea floor to the surface at 0.3 m s^{-1}. Contents of the non-filtering cod end were immediately preserved in 4% borate-buffered formalin. In the laboratory, three subsamples of 10 mL were counted using a binocular microscope. Copepod measurements (prosome length) were determined using an inverted microscope equipped with a video camera attached to a computer with image analysis software. Zooplankton samples were identified according to taxonomic descriptions provided by (Boltovskoy 1999). Copepods were identified to species when possible. Abundances (individuals m^{-3}) were converted to biomass (mg C m^{-3}) using known length-weight regressions for tropical copepods (Chisholm and Roff 1990a) with a carbon conversion factor of 0.4 (Postel et al. 2000).

4.2.4 Crustacean productivity

Water samples for chitobiase incubations were collected from two depths in the water column (1 m and 15 m). CBA decay rates were estimated from 200 mL seawater samples screened with a 20 μm mesh in order to remove any crustaceans. Approximately 20 mL of the seawater sample from each treatment was immediately filtered (0.2 μm) in order to remove any bacteria and subsequently used to estimate the native *in situ* chitobiase activity (CBA_{nat}). A crude homogenate of approximately 100 small-sized copepods (freshly ground in 3 mL of seawater; 0.2 μm filtered) was used to "spike" the original treatment samples in order to differentiate the decay of CBA from background fluorescence (see Sastri and Dower 2006). Subsamples were taken at regular intervals (i.e. at 2, 4, 6, and 24 hours), 0.2 μm filtered, and stored in the freezer (-20°C) in glass

scintillation vials until assayed. Samples were maintained at ambient seawater temperature over the 24-hour incubation period.

Measurements of chitobiase activity (CBA) followed Sastri and Dower (2006). Briefly, frozen samples were left to thaw at room temperature for two hours prior to analysis. Enzyme assays were initiated by adding the substrate 4-methylumbelliferyl-β-D-glucosaminide (0.1 mmol MBF-NAG; Sigma) to seawater samples. Assays were conducted at 25°C and terminated after 60 minutes with the addition of a 2 M NaOH and 0.4 M EDTA solution. The reaction was buffered to pH 6.0 (optimal for copepods) using a 0.15 M citrate-phosphate buffer. Chitobiase activity (nmol MBF liberated L^{-1} h^{-1}) was estimated by measuring the fluorescence of the liberated MBF using a Turner Designs TD700 fluorometer with a long wavelength bulb (300-400 nm excitation and 410-600 nm emission lenses). Raw fluorescence was converted to nmol MBF using a standard curve of known 4-methylumbelliferone concentrations against fluorescence.

Estimates of CBA decay rate (h^{-1}) were calculated as the slope (k) of the natural logarithm of CBA versus time (Sastri and Dower 2006). The reciprocal of the negative slope ($1/-k$) was used to represent the average stage duration, or the time (T_{CBA}) taken for moulting individuals to produce CBA equivalent to the chitobiase activity (CBA_{nat}). A relationship between CBA and the growth increment of the dominant marine copepods in Guanabara Bay was derived ($\log(g_{inc}) = 0.634 \log (CBA_i) - 2.039$; Table 10, Fig. 16). This relationship was then applied to the average CBA_{nat} in each treatment to calculate the absolute amount of biomass produced (ΔB). Daily crustacean productivity (mg C m^{-3} d^{-1}) was calculated as the biomass production divided by stage duration, or $\Delta B/T_{CBA}$. Net-based productivity rates were calculated using biomass estimates from 64 µm and 200

Table 10. Stage-specific individual body weight (µg DW individual^{-1}) for four common tropical copepod species as reported in the literature.

Species	Stage	Weight (µg ind^{-1})	ΔB (µg ind^{-1})
Acartia tonsa[1]*	NI	0.016	
	NII	0.011	0.005
	NIII	0.019	0.008
	NIV	0.030	0.011
	NV	0.045	0.014
	NVI	0.064	0.019
	CI	0.398	0.334
	CII	0.642	0.245
	CIII	1.235	0.593
	CIV	1.969	0.734
	CV	3.409	1.440
	CVI	6.092	2.689
Temora turbinata[2]*	CI	0.105	
	CII	0.197	0.092
	CIII	0.344	0.147
	CIV	0.587	0.243
	CV	1.204	0.617
	CVI	2.144	0.939
Oithona plumifera[3]*	NI	0.026	
	NII	0.053	0.027
	NIII	0.067	0.014
	NIV	0.111	0.044
	NV	0.149	0.038
	NVI	0.222	0.073
	CI	0.330	0.108
	CII	0.530	0.200
	CIII	0.960	0.430
	CIV	1.380	0.420
	CV	1.710	0.330
	CVI	1.900	0.190
Paracalanus sp.[4]	CI	0.220	
	CII	0.460	0.240
	CIII	0.880	0.420
	CIV	1.430	0.550
	CV	1.740	0.310
	CVI	3.090	1.350

ΔB was calculated as the difference in weight between successive developmental stages. Weights reported for species denoted with an asterisk (*) were calculated from length using the length-weight regressions reported in Ara (2001). [1]Heinle (1966). [2]Ara(2002). [3]Ara(2001). [4]Webber and Roff (1995)

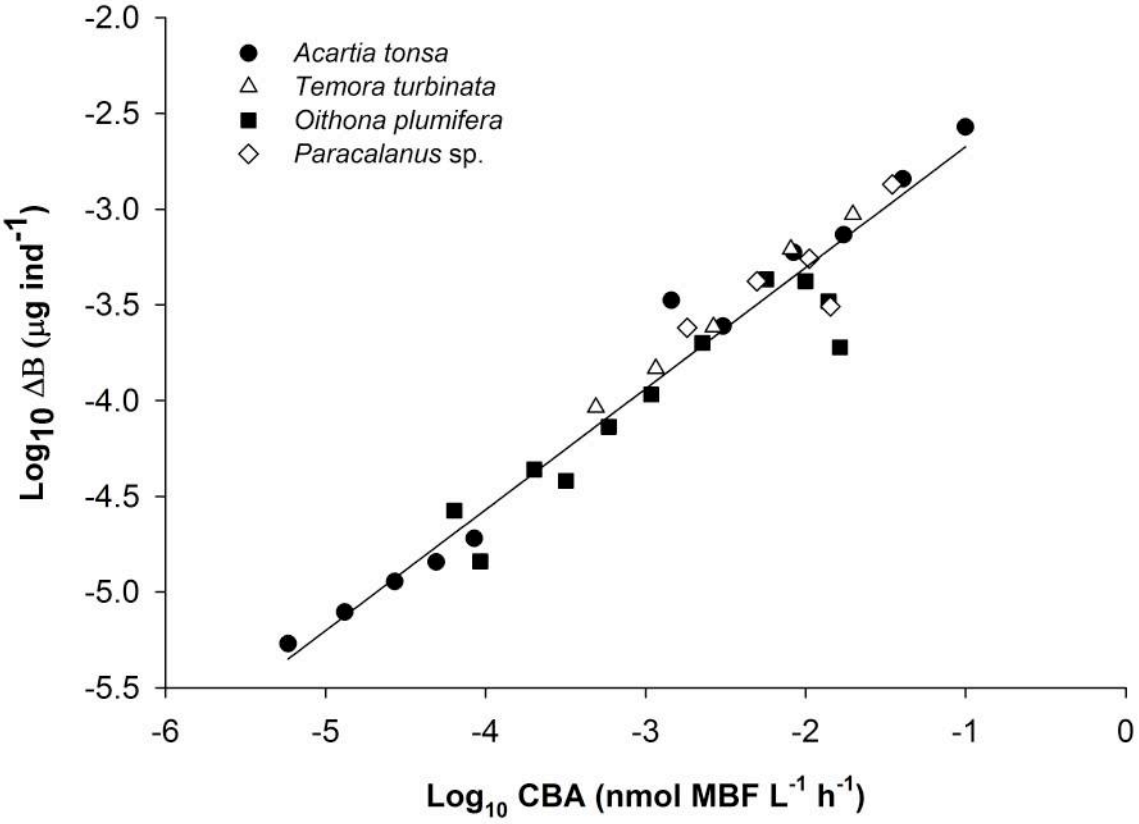


Figure 16. Relationship between change in body weight between successive developmental stages (ΔB, μg ind^{-1}) and individual chitobiase activity (CBA, nmol MBF L^{-1} h^{-1}). CBA was estimated by applying a known relationship between individual chitobiase activity and post-moult body weight (Sastri and Dower 2009) to each of the successive weights presented in Table 10.

μm zooplankton nets by applying the Huntley and Lopez (1992), Hirst and Lampitt (1998), and Hirst and Bunker (2003) global mathematical models, which incorporate temperature, body size, chlorophyll *a*, or a combination of these variables into growth rate equations (Appendix G).

4.2.5 Statistical analysis

Best subsets regression was used to select the best-fitting model to explain our chitobiase- and net-based estimates. All available abiotic and biotic data were used as explanatory variables (chlorophyll *a*, salinity, temperature, oxygen, rainfall, tidal height, and wind speed) and were log-transformed when appropriate. Only statistically significant models with no multicollinearity present were selected based on the highest adjusted R^2 and lowest mean squared error values. Multiple linear regressions were then performed on the explanatory variables represented in the best model. All analyses were performed using Sigmaplot® version 12.3 and R version 3.0.2 (R Development Core Team, 2013).

4.3 Results

4.3.1. Physical and biological measurements

Water column temperature in Guanabara Bay followed a typical seasonal pattern showing a gradual decrease from 25°C to less than 22°C from Day 101 to 170 (April to June) (Fig. 17a). Salinity values showed little variation during the study period, ranging between 31.8 and 33.0 (Fig. 17b). Oxygen was lowest between Days 108 and 123, ranging between 3 and 5 mg L^{-1} throughout the sampling period (Fig. 17c). The

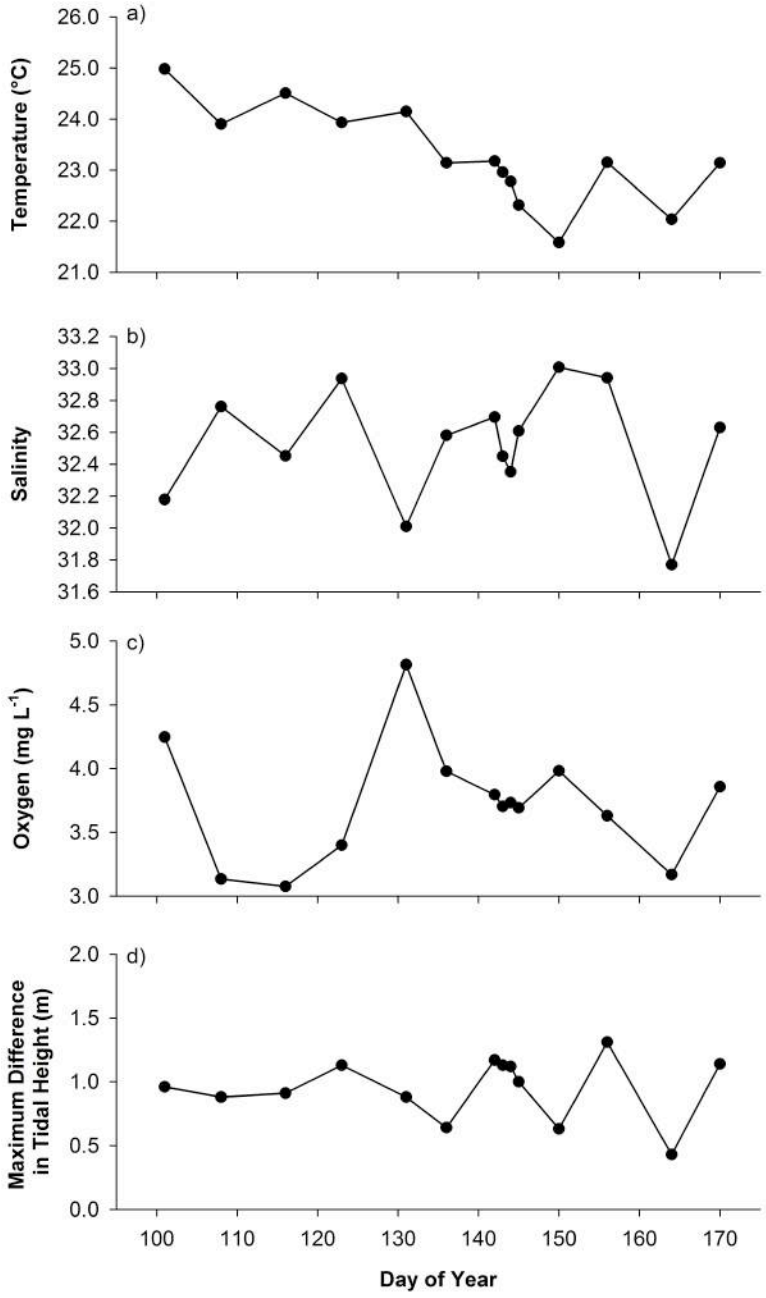


Figure 17. Average water column a) temperature (°C), b) salinity, c) oxygen (mg L^{-1}), and d) maximum difference in tidal height (m) from the lowest to highest measured tide on the sampling day from April to June, 2012.

maximum difference in tidal height, which was the difference between the highest and lowest measured tides on our sampling days, was fairly constant in April (approximately 1 m) compared to the fluctuations observed after mid-May (Fig. 17d). Average rainfall amounts were substantially higher at the beginning of our sampling program (i.e. Days 101, 123, and 136) compared to other sampling dates (Fig. 18a). In addition, mean daily wind speeds were higher in May and June than they were in April (Fig. 18b). Slight variations in temperature, salinity, and mean wind speed were observed during our daily sampling in May (Days 142 to 145), whereas the maximum difference in tidal height and rainfall amount were fairly constant.

POC ranged between 1.4 and 12.2 mg L^{-1} (Fig. 19a). Maximum POC concentrations were observed in early May (8.3 and 12.2 mg L^{-1} on Days 130 and136, respectively). Total nitrogen ranged between 0.6 and 2.2 mg L^{-1} and peaked on the same dates as POC in early May (Fig. 19a). In terms of phytoplankton biomass, the <20 µm size fraction dominated in Guanabara Bay, however, >20 µm cells (diatoms) also contributed to the total chlorophyll concentration in May (Fig. 19b). Spearman's Rank Order correlations revealed significant, negative correlations between surface salinity (1 m) and both <20 µm chlorophyll *a* ($r = -0.73$, $p < 0.01$) and POC ($r = -0.66$, $p < 0.05$). A negative, but non-significant correlation was found between surface salinity and total nitrogen ($r = -0.51$, $p = 0.07$).

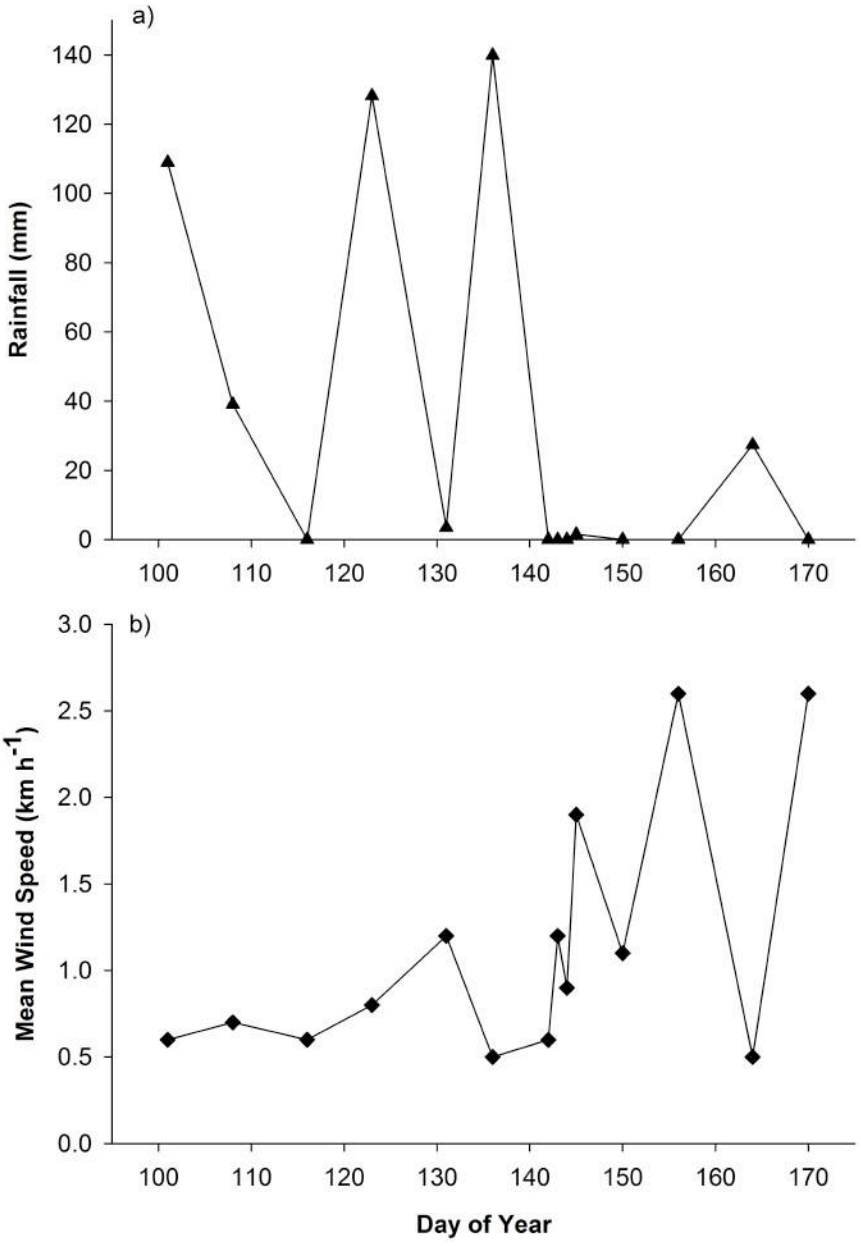


Figure 18. Average rainfall (mm) for the 48 hours prior to sampling (a) and mean wind speed (km h^{-1}) on the sampling day (b) from April to June, 2012.

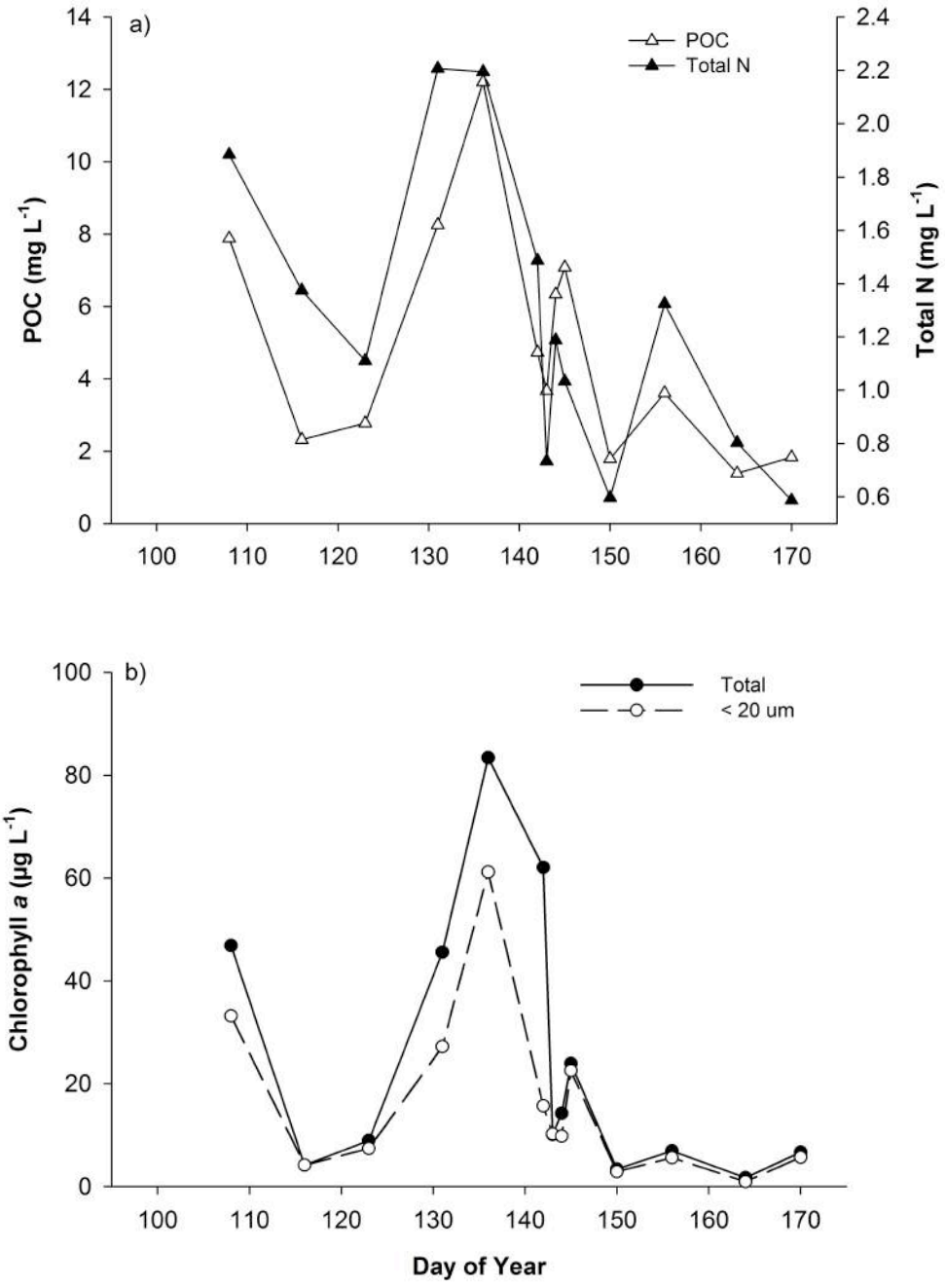


Figure 19. a) POC (mg L^{-1}; left axis) and Total N (mg L^{-1}; right axis) and b) Total and < 20 µm chlorophyll *a* (µg L^{-1}) from April to June, 2012.

4.3.2 Zooplankton abundance and biomass

The amount of zooplankton collected in the 200 µm mesh net was consistently lower than that collected with the 64 µm mesh net. Crustacean abundance from the 64 µm mesh net ranged between 36,000 and 315,000 individuals m^{-3} (mean 112,000 ind. m^{-3}) with the highest abundance occurring on the second sampling date in April (Fig. 20a). Nauplii contributed substantially to overall crustacean abundance on most of the sampling dates. Although *Acartia tonsa* and *Oithona hebes* were the dominant copepods throughout our study, *Paracalanus* sp., *Temora turbinata*, and *Oncaea venusta* were also abundant in most of the zooplankton samples. In contrast to abundance, peak copepod biomass calculated from the 64 µm mesh net occurred in early June when biomass was 61.2 mg C m^{-3} (Fig. 20b) due to a high abundance of *T. turbinata*. Mean biomass estimated from the 64 µm mesh net was 18.1 mg C m^{-3}. Biomass estimates from the 200 µm mesh net were consistently lower and often underestimated the biomass values obtained from the 64 µm mesh net by over 80%. Crustacean abundance showed little variation during our daily sampling in May; however, biomass in the 64 µm mesh net decreased substantially on day 143 before increasing again on days 144 and 145.

4.3.3 Chitobiase-based crustacean productivity

For our chitobiase-based measurements, average CBAnat was significantly higher in April (14.38 nmol L^{-1} h^{-1}) compared to May and June (Student's *t*-test, $p < 0.0001$) (Table 11). In addition, average TCBA, or stage duration, was significantly longer in April (approximately 1.3 days) compared to stage durations in May and June (less than 0.5 days) (Student's *t*-test, $p < 0.01$). Overall, crustacean productivity ranged between

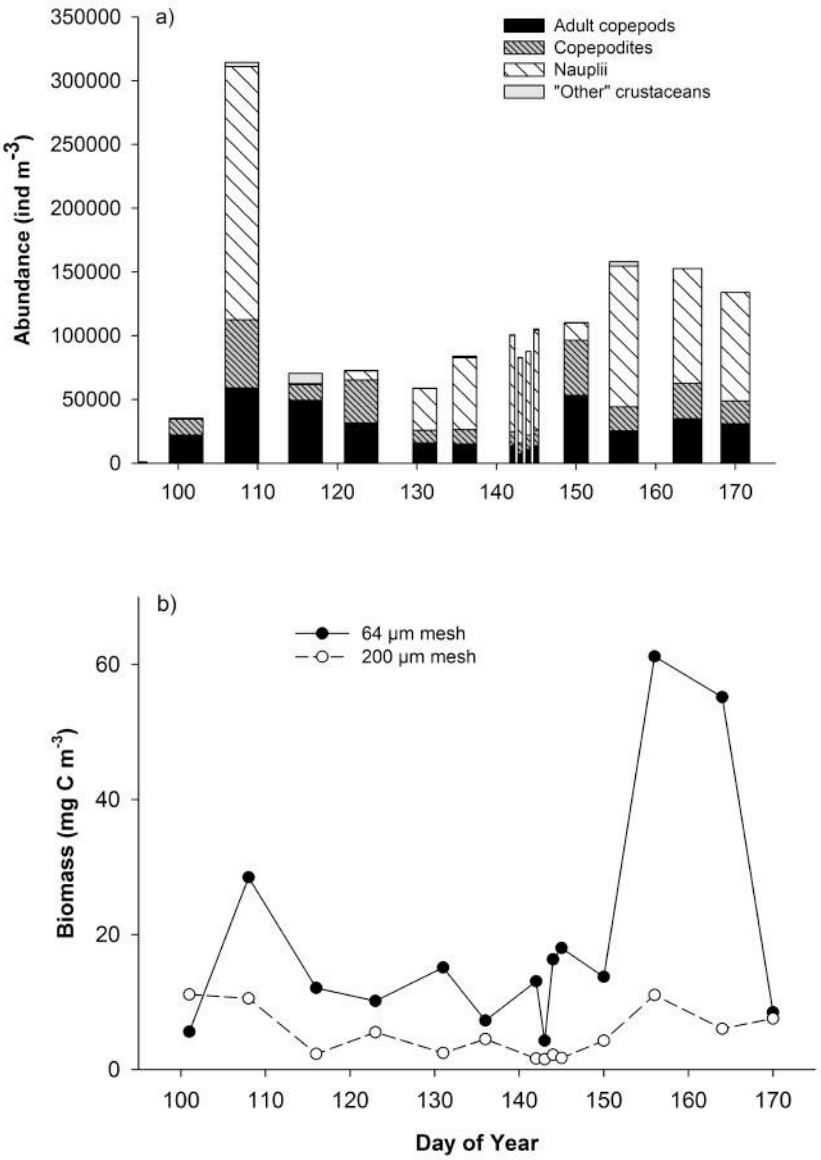


Figure 20. a) Net-based crustacean abundance (ind m^{-3}) from the 64 µm mesh zooplankton net and b) Net-based crustacean biomass (mg C m^{-3}) estimated from 64 µm and 200 µm mesh zooplankton nets from April to June, 2012. Note: the four narrow bars after Day 140 represent daily samples.

Table 11. Average monthly chitobiase-based estimates of CBAnat (nmol L^{-1} h^{-1}), and TCBA (stage duration; days). Range of values is also presented.

	CBAnat (nmol L^{-1} h^{-1})		TCBA (days)	
April	14.38	(12.19-16.77)	1.27	(0.75-2.23)
May	3.93	(1.33-12.82)	0.42	(0.28-0.71)
June	3.53	(2.04-4.86)	0.37	(0.31-0.42)

9.95 and 29.33 mg C m^{-3} d^{-1} and were slightly higher in April than in May/June, varying with the spring/neap tidal cycle (Fig. 21). Specifically, crustacean productivity was higher during the neap tides in April, whereas in May and June productivity was higher during the spring tides. As a result, crustacean productivity varied more on a weekly timescale compared to the daily timescale examined in May. Average monthly chitobiase-based crustacean productivity was substantially higher than productivity estimated from the mathematical models on all but one occasion (i.e. June value from the 64 µm mesh net using the Huntley and Lopez model) (Table 12). In addition, productivity estimated by the mathematical models based on the contents of the 64 µm mesh net were consistently higher than those determined from the 200 µm mesh net.

Best subset regression analysis was used to determine which explanatory variables best explained variations in net-based and chitobiase-based productivity estimates (Tables 13,14). In terms of net-based estimates, relationships between explanatory variables and either abundance or biomass varied depending on the size of the mesh (Table 13). For example, crustacean abundance estimated from the 64 µm mesh net was negatively related to oxygen (Adj $R^2 = 0.23$, $p < 0.05$). For the 200 µm mesh net, the only significant relationships in the model were between crustacean abundance and <20 µm chlorophyll *a* (negative, $p < 0.05$) and between abundance and salinity (positive, $p < 0.01$) (Adj $R^2 = 0.60$). In terms of copepod biomass, the model explained none of the variability in biomass calculated from the 64 µm mesh net. In contrast, a significant relationship was found between 200 µm mesh net copepod biomass and rainfall ($p < 0.05$) and nearly significant relationships were found with both <20 µm chlorophyll *a* and

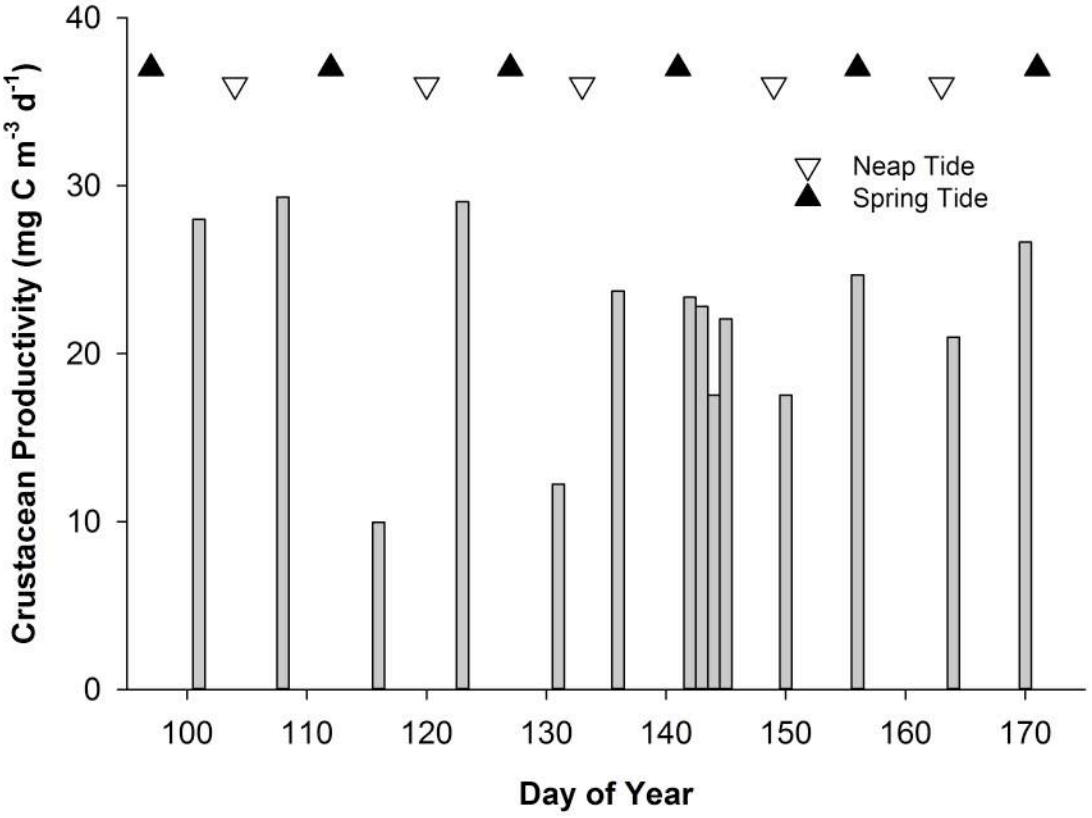


Figure 21. Chitobiase-based crustacean productivity (mg C m^{-3} d^{-1}) from April to June, 2012. Timing of the spring versus neap tides is indicated at the top of the graph.

Table 12. Comparison of chitobiase-based crustacean productivity (mg C m^{-3} d^{-1}) derived from the global predictive models of Huntley and Lopez (1992), Hirst and Lampitt (1998), and Hirst and Bunker (2003). Model values were produced using biomass estimates from both the 64 µm and 200 µm mesh zooplankton nets for comparison.

	Chitobiase	Huntley and Lopez		Hirst and Lampitt		Hirst and Bunker	
		64 µm	200 µm	64 µm	200 µm	64 µm	200 µm
April	22.43	10.04	5.37	6.78	1.25	8.54	3.35
May	21.04	6.98	1.71	4.57	0.47	9.21	1.54
June	24.11	22.95	4.63	12.12	1.08	12.12	1.54

Table 13. Results of multiple linear regressions and the significance of the models chosen by Best Subsets regression best describing the explanatory variables influencing 64 µm mesh and 200 µm mesh net-based estimates of crustacean abundance and copepod biomass. Significant values are indicated in bold.

			Coefficient	SE	*P*-value
64 µm	Log_{10}Crustacean Abundance	Constant	5.981	0.433	
	N = 13	Oxygen	-0.265	0.115	**0.041**
	R^2 = 0.305				
	Adj R^2 = 0.247				
	Log_{10}Copepod Biomass	Constant	1.277	0.901	
	N = 13	Log_{10}<20 µm Chl	-0.295	0.234	0.244
	R^2 = 0.310	Oxygen	-0.227	0.268	0.422
	Adj R^2 = 0.000	Log_{10}Rainfall	0.094	0.082	0.282
		Log_{10}Wind	1.279	1.121	0.287
200 µm	Log_{10}Crustacean Abundance	Constant	-20.481	6.167	
	N = 13	Log_{10}<20 µm Chl	-0.418	0.132	**0.020**
	R^2 = 0.800	Salinity	0.618	0.171	**0.011**
	Adj R^2 = 0.599	Temp	0.172	0.076	0.063
		Log_{10}Rainfall	0.106	0.052	0.088
		Tidal Height	-0.553	0.316	0.131
		Log_{10}Wind	1.051	0.558	0.109
	Log_{10}Copepod Biomass	Constant	-13.963	7.199	
	N = 13	Log_{10}<20 µm Chl	-0.383	0.179	0.065
	R^2 = 0.530	Salinity	0.431	0.219	0.085
	Adj R^2 = 0.295	Log_{10}Rainfall	0.169	0.066	**0.033**
		Log_{10}Wind	1.120	0.779	0.188

Table 14. Results of multiple linear regressions and the significance of the models chosen by Best Subsets regression best describing the explanatory variables influencing chitobiase-based estimates of crustacean productivity, CBAnat and TCBA. Significant values are indicated in bold.

		Coefficient	SE	*P*-value
Log_{10}Crustacean Productivity	Constant	0.639	3.232	
N = 14	Salinity	0.069	0.090	0.469
$R^2 = 0.646$	Temperature	-0.086	0.038	0.053
Adj $R^2 = 0.488$	Log_{10}Rainfall	0.088	0.024	**0.006**
	Tidal height	0.429	0.166	**0.030**
Log_{10}CBAnat	Constant	-15.348	3.053	
N = 14	Salinity	-0.309	0.088	**0.007**
$R^2 = 0.943$	Temperature	0.311	0.034	**<0.001**
Adj $R^2 = 0.918$	Oxygen	-0.358	0.066	**<0.001**
	Log_{10}Rainfall	-0.059	0.022	**0.022**
Log_{10}TCBA	Constant	-11.090	2.366	
N = 14	Salinity	0.164	0.065	**0.036**
$R^2 = 0.943$	Temperature	0.286	0.027	**<0.001**
Adj $R^2 = 0.907$	Oxygen	-0.186	0.044	**0.003**
	Log_{10}Rainfall	-0.062	0.017	**0.007**
	Tidal Height	-0.551	0.118	**0.002**

salinity. Chitobiase-based estimates were best predicted by a linear combination of salinity, oxygen (CBA_{nat} and TCBA), temperature, rainfall, and tidal height (productivity and TCBA) (Table 14). Of these variables, only rainfall ($p < 0.01$) and tidal height ($p < 0.05$) showed positive, significant relationships with crustacean productivity. In contrast to crustacean productivity, temperature was the best predictor ($p < 0.001$) for both CBA_{nat} and TCBA. However, all variables in the models for CBA_{nat} and TCBA were significant (Table 14).

4.4 Discussion

Despite the fact that tropical regions are generally viewed as having low zooplankton biomass and thus low productivity throughout the year compared to temperate regions (Webber and Roff 1995), the eutrophic nature of Guanabara Bay supports high crustacean abundance, biomass, and productivity. Our values for mean crustacean abundance (112,000 ind. m^{-3}) and mean biomass (18.1 mg C m^{-3}) are similar to those observed in Kingston Harbour, Jamaica (92,500 m^{-3} and 22.1 mg m^{-3} for mean abundance and biomass, respectively) (Hopcroft et al. 1998a). In addition, mean biomass in Guanabara Bay was only slightly higher relative to that observed in Patos Lagoon estuary in southern Brazil (13.64 mg C m^{-3}) (Avila et al. 2012). Furthermore, although nauplii dominated our samples numerically, copepodite stages likely contributed to the majority of biomass in the 64 µm mesh net (Hopcroft et al. 2001, McKinnon and Duggan 2003).

In order to compare our productivity estimates with previous studies in the region, we converted our chitobiase-based productivity estimates into energy density using a

conversion factor of 25 kJ g^{-1} (Chisholm and Roff 1990b). Across the entire sampling period, average crustacean productivity in the present study was 550 J m^{-3} d^{-1}, which is substantially higher than the average copepod community productivity of 307 J m^{-3} d^{-1} estimated for Kingston Harbour by Hopcroft et al. (1998a). In contrast, in tropical regions where food may become limiting to copepods (i.e. oceanic or shelf regions), productivity is lower than the values reported in our study due to a decreased availability of food (Webber and Roff 1995, McKinnon and Duggan 2003). Therefore, the high productivity observed in Guanabara Bay can be attributed to a combination of high copepod abundance, fast growth rates, and higher food concentrations. As a result, productivity values in Guanabara Bay and other eutrophic tropical regions may be comparable to, or even higher than, those observed in temperate water bodies (Hopcroft et al. 1998a).

Crustacean productivity in Guanabara Bay was most strongly influenced by rainfall and tidal height. Higher rainfall amounts and an increase in water column mixing associated with high tides results in larger inputs of organic material to the bay originating from riverine and sewage wastes (Guenther and Valentin 2008, Guenther et al. 2008). This organic material fuels the microbial loop in the inner waters of the bay supporting high bacterial production (Guenther and Valentin 2008), thereby recovering the fixed carbon (DOM, POM) which would otherwise be lost in the food web (Sherr and Sherr 1988, Pomeroy et al. 2007). Heterotrophic microflagellates, in turn, feed on bacteria (Azam et al. 1983) and provide a food source for small copepods. Thus, higher crustacean productivity during certain times of the tidal cycle can be attributed to conditions providing a better food source (i.e. smaller food items from the microbial loop) for the small copepods that dominate Guanabara Bay.

Rainfall has also been shown to affect the hydrological structure of the bay due to its association with the passage of cold fronts, which result in advective changes to the zooplankton (Valentin and Marazzo 2003). Horizontal advection due to tidal currents influences cladoceran (Marazzo and Valentin 2000) and copepod (Gomes et al. 2004) dynamics in the region with an exportation of zooplankton during spring tides and a subsequent importation during neaps tides (Júnior et al. 2007). Our productivity estimates may be explained, in part, by the timing of our sampling date during the tidal cycle. By sampling on different timescales (weekly, daily), our results indicate that the weekly tidal changes resulted in larger variations in crustacean productivity compared to the variations on a daily timescale. During April, crustacean productivity was higher during the neap tides, which are strongly influenced by waters from the inner bay (i.e. low salinity, high temperature, high chlorophyll *a*) (Guenther et al. 2008). In contrast, during May and June high productivity was associated with the spring tides. Although productivity might be expected to be lower during the spring tides due to the potential export of zooplankton, our sampling times occurred close to the lowest tide of the day, and was thus influenced by inner bay waters as opposed to the coastal waters that enter the bay during high tides. Unfortunately, our sampling dates were limited based on the availability of equipment and personnel; however, future sampling in this region should be conducted at the same time during the tidal cycle in order to avoid confounding influences of the tides.

The relationship between environmental variables and net-based estimates of both abundance and biomass varied depending on the mesh size of the net. For example, crustacean abundance in the 64 µm mesh net was only related significantly to oxygen. Although low oxygen concentrations (<1 mg L^{-1}) are known to result in low abundances

of copepods (Roman et al. 1993), oxygen concentrations in our study were never below 3 mg L^{-1}. Thus, it is more likely that the negative relationship between crustacean abundance and oxygen resulted from the association between low oxygen concentrations and inner bay waters. In contrast, abundance from the 200 µm mesh showed significant relationships with both <20 µm chlorophyll *a* (negative) and salinity (positive). The lack of a relationship between chlorophyll *a* and biomass from the 64 µm mesh net is likely a result of the size range of animals typically collected in nets with different mesh sizes. Specifically, the 64 µm mesh net would have a higher density of nauplii and smaller copepodites, which feed predominantly on smaller particles via the microbial loop (McKinnon and Duggan 2003). On the other hand, the larger-bodied individuals collected with the 200 µm mesh net would feed on larger phytoplankton at the upper end of the <20 µm size fraction of chlorophyll *a*. In contrast to our results with the 200 µm mesh net, Hopcroft et al. (1998a) found no significant correlations between chlorophyll and copepod abundance or biomass (even with a time lag) in the eutrophic Kingston Harbour, Jamaica. Our results suggest that phytoplankton biomass may have been briefly regulating the abundance and biomass of larger zooplankton in Guanabara Bay.

In contrast to abundance, biomass, and productivity estimates, CBAnat (the average sum of all individual growth) and TCBA (stage duration) were influenced by abiotic factors (temperature, salinity, and oxygen). For instance, both CBAnat and stage duration increased with the higher water temperatures and lower oxygen concentrations associated with the inner bay waters. In general, we would expect that stage durations would be shorter at higher temperatures (Landry 1975). However, chitobiase-based stage durations represent the average stage duration of all moulting individuals present in the

water column. Therefore, the larger size range of moulting individuals associated with inner bay waters may have contributed to the longer stage durations observed (Peterson et al. 1991) despite the slight increase temperature. In addition, CBAnat was higher while stage durations were shorter at lower salinities. These results suggest that more individuals were moulting and that the average individual was spending less time in a given stage (either due to their smaller size or because development rates were faster) at lower salinities.

In terms of food availability and quality, the lower salinities inside the bay, associated with higher POC and chlorophyll *a* concentrations, provide a better food source for small individuals than the diluted phytoplankton concentrations found in the more saline coastal water. It is important to note that previous studies have shown *A. tonsa* growth rates may be limited at salinities higher than 30 (Cervetto et al. 1999). Even though *A. tonsa* was not always the dominant copepod in our samples, the higher salinities observed during our study may have decreased activity of the chitobiase enzyme and thus limited growth in certain copepod species with narrower salinity tolerances (Avila et al. 2011). Nonetheless, these results suggest that although abiotic factors might not be directly linked to productivity estimates, they can still influence the growth rates and development times of copepods thereby indirectly influencing crustacean production.

Average monthly chitobiase-based crustacean productivity in our study was consistently higher (i.e. 2 to 5 times) than productivity estimates from global predictive models. Recent production rates calculated for Patos Lagoon using the chitobiase method were also consistently higher than those generated by the Huntley and Lopez (1992) and

Hirst and Bunker (2003) mathematical models (Avila et al. 2012). Our productivity values derived from the mathematical models only considered copepod productivity because these were the only organisms for which we had measurements. As a result, "other" crustaceans were excluded from these calculations, which may have contributed to the underestimation of the models compared to chitobiase productivity. Avila et al. (2012) found that productivity estimates from the Huntley and Lopez (1992) temperature-based model were generally higher than those produced by the Hirst and Bunker (2003) model, which also included the variables of body size and chlorophyll concentration. Our results also showed that productivity values derived from the Huntley and Lopez (1992) model were higher during June; however, productivity estimates for the models were similar during April and May. Studies that have used the chitobiase method in coastal temperate waters on the West Coast of Canada, have shown that both the Huntley and Lopez (1992) and Hirst and Lampitt (1998) models underestimate productivity when crustacean growth rates are higher than >0.25 d^{-1} (Sastri and Dower unpublished, Suchy et al. in prep). Thus, given the high growth rates of copepods in tropical regions, it is likely that the use of these models will consistently underestimate productivity due to their reliance on net-based biomass estimates. Furthermore, the Hirst and Bunker (2003) model, which takes chlorophyll *a* into account, might not be as applicable in eutrophic tropical regions wherein copepods are feeding mainly on the microbial loop.

One of the major drawbacks of using biomass to calculate productivity values is the bias associated with net selectivity. Net selectivity is an issue of major concern in oceanographic studies because mesh size can influence abundance, biomass, and productivity estimates (Gallienne and Robins 2001). Coarse (>200 μm mesh) nets have

been shown to undersample a large portion of the zooplankton community (Hopcroft et al. 2001), while the use of smaller mesh sizes (<100 µm) may result in a higher abundance and biomass of small-bodied zooplankton compared to larger individuals (see Turner 2004 for review). Moreover, Hopcroft et al. (1998a) found that a 200 µm mesh net estimated only 48% of the productivity estimated by a 64 µm mesh net and that over half of the biomass is missed when sampling with a 200 µm mesh net (Hopcroft et al. 2001). In contrast to these results, Avila et al. (2012) found that mean biomass of copepods was higher from the 200 µm mesh net compared to a 64 µm mesh net in Patos Lagoon, Brazil. Nevertheless, these results highlight the importance of taking net selectivity into consideration when estimating abundance, biomass, and productivity, particularly in tropical regions. Consistency in the size of nets used would help limit the contrasting results between studies and allow for a better comparison of such estimates across studies.

4.5 Conclusions

Despite the lack of a consistent method for estimating zooplankton production as of yet, the fact that our productivity estimates were comparable with previous estimates for tropical regions suggests that the chitobiase method is a reliable tool for estimating *in situ* productivity without the need for repeatedly handling or incubating animals. Furthermore, this method estimates productivity for all moulting crustaceans in the water column, including nauplii and smaller copepodites that are often missed in net collections. However, it is important to note that we were unable to consider the contribution of chitobiase by moulting benthic crustaceans, which may have lead to

slight overestimations in planktonic crustacean production due to the shallow nature of Guanabara Bay. Results from the present study indicate that small copepods may contribute a substantial amount of crustacean production in eutrophic tropical estuaries, thus deserving more attention. Furthermore, these results suggest that crustacean productivity in tropical waters may be substantially higher than is generally assumed. Therefore, routine estimates of crustacean productivity should be incorporated into sampling regimes. Ideally, such measurements should be made on a weekly or biweekly basis, preferably during the same time in the tidal cycle, in order to effectively estimate how much energy is available to higher trophic levels in dynamic tropical water bodies.

Currently, the amount of energy available from zooplankton to higher trophic levels remains largely unknown, and this particularly true for tropical ecosystems. Routine estimates of crustacean productivity will allow oceanographers to more accurately characterize patterns of spatial and temporal variability in production across marine ecosystems. With the same resolution as well-documented estimates of primary productivity, routine crustacean productivity estimates will provide insight into how variations in productivity may impact fish production in different ecosystems. Ultimately, reliable productivity estimates, such as those established in the present study, are necessary in terms of future ecosystem modeling and management practices.

Chapter 5: Conclusions and Significance of Research

The main objective of this thesis was to quantitatively link routine estimates of community-level crustacean productivity to changes in food availability, food quality, and primary productivity. Routine estimates of crustacean productivity allow us to examine some of the key assumptions commonly made in oceanographic studies. Results from Chapter 2 show that copepods fed a poor food item (the dinoflagellate *Amphidinium carterae*) took a longer time to develop through early stages, had lower daily growth rates, and exhibited lower productivity. Chapter 3 is the first study to temporally link patterns of variability in primary productivity and community-level crustacean productivity in a marine setting. This work revealed that if we rely on biomass estimates alone, we fail to capture important variations in the rate in which crustacean biomass is generated throughout the season. Furthermore, while a positive relationship was found between primary productivity and crustacean productivity in 2010, there was a negative relationship between primary productivity and crustacean productivity in 2011, which may be due to top-down control by cnidarian predators. When values of primary productivity and crustacean productivity were time-averaged, substantial interannual variations in trophic transfer efficiency (TTE) were observed despite the fact that average TTE was ~20% for both years. Finally, Chapter 3 showed that high crustacean productivity in Guanabara Bay was associated with factors that contributed to a better food source for small copepods. In addition, this work reveals that community-level crustacean productivity in tropical regions dominated by the microbial food loop can be as high as, if not higher than,

productivity measured in temperate regions. By building on the framework established by more traditional methods of estimating crustacean productivity, this work provides insight into how accurate productivity estimates can improve our understanding of zooplankton dynamics in both laboratory and field settings in different oceanic regions.

5.1 Chitobiase versus traditional crustacean productivity estimates

The lack of a consistent method for estimating crustacean productivity makes it difficult to compare the results from the current study to previous work. That said, the field estimates of chitobiase-based productivity obtained for both temperate and tropical regions in this thesis were consistently higher (by up to four times) than previously reported productivity estimates for copepod communities. For example, chitobiase-based crustacean productivity in Saanich Inlet (Chapter 3) ranged between 0.05 and 15.61 mg C m^{-3} d^{-1} (mean 4.71 mg C m^{-3} d^{-1}), or depth-integrated values of 0.01 to 0.65 g C m^{-2} d^{-1} (mean 0.24 g C m^{-2} d^{-1}) (Table 15). In comparison, Peterson et al. (1991) found that the range of community-level productivity obtained using incubation methods in the Skagerrak region over the summer period was slightly lower (3.0-8.0 mg C m^{-3} d^{-1}) than productivity in Saanich Inlet. Maximum depth-integrated copepod productivity for a coastal region in Denmark (0.16 g C m^{-2} d^{-1}) (based on the egg production method) was substantially lower than our chitobiase-based estimates (maximum of 0.65 g C m^{-2} d^{-1}) (Kiørboe and Nielsen, 1994).

Table 15. Comparison of community-level chitobiase-based productivity with estimates based on traditional methods in temperate and tropical regions.

	Region	Method	Productivity		Study
			Range	Mean	
Temperate:	Saanich Inlet	Chitobiase	0.05-15.61 mg C m^{-3} d^{-1} or 0.01-0.65 g C m^{-2} d^{-1}	4.71 mg C m^{-3} d^{-1} or 0.24 g C m^{-2} d^{-1}	Present Study
	Skagerrak	Incubation	3.0-8.0 mg C m^{-3} d^{-1}	4.6 mg C m^{-3} d^{-1}	Peterson et al. 1991
	Coastal region, Denmark	Egg production	0.01-0.16 g C m^{-2} d^{-1}	n/a	Kiørboe and Nielsen 1994
	Sagami Bay, Japan	Hirst and Lampitt (1998) model	0.09-7.77 mg C m^{-3} d^{-1}	0.94 mg C m^{-3} d^{-1}	Ara and Hiromi 2007
Tropical:	Guanabara Bay	Chitobiase	9.95-29.33 mg C m^{-3} d^{-1}	21.99 mg C m^{-3} d^{-1}	Present Study
	Kingston Harbour, Jamaica	Incubation	n/a	12 mg C m^{-3} d^{-1}	Hopcroft et al. 1998a*
	Discovery Bay, Jamaica	Incubation	0.05-0.23 mg C m^{-3} d^{-1}	n/a	Webber and Roff 1995*

*Note: productivity values were originally reported in J m^{-3} d^{-1} and were converted to mg C m^{-3} d^{-1} for comparison with our chitobiase-based results by applying a 25 kJ g^{-1} energy density conversion factor (Chisholm and Roff 1990b).

Using yet another method, Ara and Hiromi (2007) estimated productivity based on the Hirst and Lampitt (1998) model in Sagami Bay, Japan, and determined that daily productivity for the entire copepod community varied between 0.097 and 7.77 mg C m^{-3} d^{-1}.

Productivity in Guanabara Bay (Chapter 4), on the other hand, ranged between 9.95 and 29.33 mg C m^{-3} d^{-1} (mean 21.99 mg C m^{-3} d^{-1}) (Table 15). Comparable productivity estimates are even more rare in tropical regions than in temperate regions with only two previous studies providing estimates for the entire copepod community. Average copepod community productivity in Guanabara Bay was about twice as high as that reported for Kingston Harbour, Jamaica (12 mg C m^{-3} d^{-1}) (Hopcroft et al. 1998a). Furthermore, the range of daily productivity (0.05-0.23 mg C m^{-3} d^{-1}) for all copepod species in the upper 60 m of the water column in the more food-limited region of Discovery Bay, Jamaica, did not even fall within the lower limits of productivity we observed in Guanabara Bay (Webber and Roff 1995).

The scarcity of results against which our chitobiase-based productivity estimates can be compared highlights the need for routine community-level crustacean productivity estimates in the field of zooplankton ecology. Without a consensus on a consistent method, akin to the ^{14}C method commonly employed for primary productivity estimates, it remains difficult to determine whether or not observed differences in crustacean productivity are real or merely artifacts of the sampling method(s) chosen. Given that larval fish and other consumers eat nauplii, copepodites, and adult copepods (see Turner 2004), the chitobiase method provides a more accurate estimate of the production available to higher trophic levels compared

to traditional methods, which typically exclude naupliar stages and small copepods in general. Furthermore, chitobiase-based estimates incorporate the productivity of all moulting crustaceans including decapod larvae, amphipods, and euphausiids, which are key prey items for many juvenile fish species, e.g. chinook and coho salmon (Schabetsberger et al. 2003). Therefore, although copepods are assumed to numerically dominate the zooplankton in most marine ecosystems, chitobiase estimates are particularly useful in capturing total production in regions wherein euphausiids and other non-copepod crustaceans can often constitute a large proportion of the zooplankton (e.g. Saanich Inlet, BC). The field of zooplankton ecology would benefit greatly from an inter-calibration experiment involving concurrent estimates using traditional methods (incubation techniques, egg production method) and the chitobiase method in order to quantify the biases associated with using these different methods to estimate productivity.

5.2 Chitobiase-based productivity versus global predictive models

The use of global predictive models to estimate copepod growth rates and productivity has grown in popularity because such models require relatively little effort compared to more traditional methods. However, these models have not been rigorously tested and can give unrealistic estimates (Hirst and Sheader 1997). Indeed, results from this thesis highlight the fact that mathematical models can under- or overestimate productivity depending on copepod growth rates. Chitobiase-based values of daily production to biomass (P:B), which is the equivalent of a daily mean-specific growth rate (Sastri et al. 2012) in Saanich Inlet ranged between 0.01 and 0.42

d^{-1} (mean 0.11 d^{-1}), whereas growth rates in Guanabara Bay ranged between 0.15 and 1.2 d^{-1} (mean 0.76 d^{-1}). Although these growth rates fell within the ranges previously reported in temperate (e.g. Peterson et al. 2002, Sastri and Dower 2009) and tropical (e.g. Hopcroft et al. 1998b) regions, the application of global predictive models, on average, tended to overestimate productivity by 595% (ranging from 129 to >1000%) in Saanich Inlet, particularly when growth rates were lower than 0.05 d^{-1} (e.g. during May; Fig. 22a,b). Moreover, the temperature-dependent Huntley and Lopez (1992) model consistently overestimated chitobiase productivity in Saanich Inlet during the slightly warmer year of 2010 (Fig. 22a). In contrast, the models, on average, underestimated productivity by 41% (ranging from 28 to 66%) in Guanabara Bay when growth rates were higher than 0.25 d^{-1} (Fig. 22c). Peterson et al. (2002) found good agreement between productivity estimates off the coast of Oregon and the Hirst and Lampitt (1998) model when growth rates were <0.10 d^{-1}; however, the model underestimated growth at rates >0.10 d^{-1} because many of the copepods were growing at maximum rates not represented by the "global" averages used in the models.

The discrepancies between chitobiase-based productivity estimates and estimates derived from the global models in this thesis can be attributed to a few key reasons. First, the Huntley and Lopez (1992) model is not representative of "community-level" planktonic crustacean productivity because it is derived from global measurements that are only applicable to a specific size range of planktonic copepods representing the 'average' global growth rate. Therefore, larger individuals with slower growth rates such as euphausiids and other non-copepod crustaceans (e.g. amphipods, crab zoea, etc.) are excluded from the model. Chitobiase estimates of

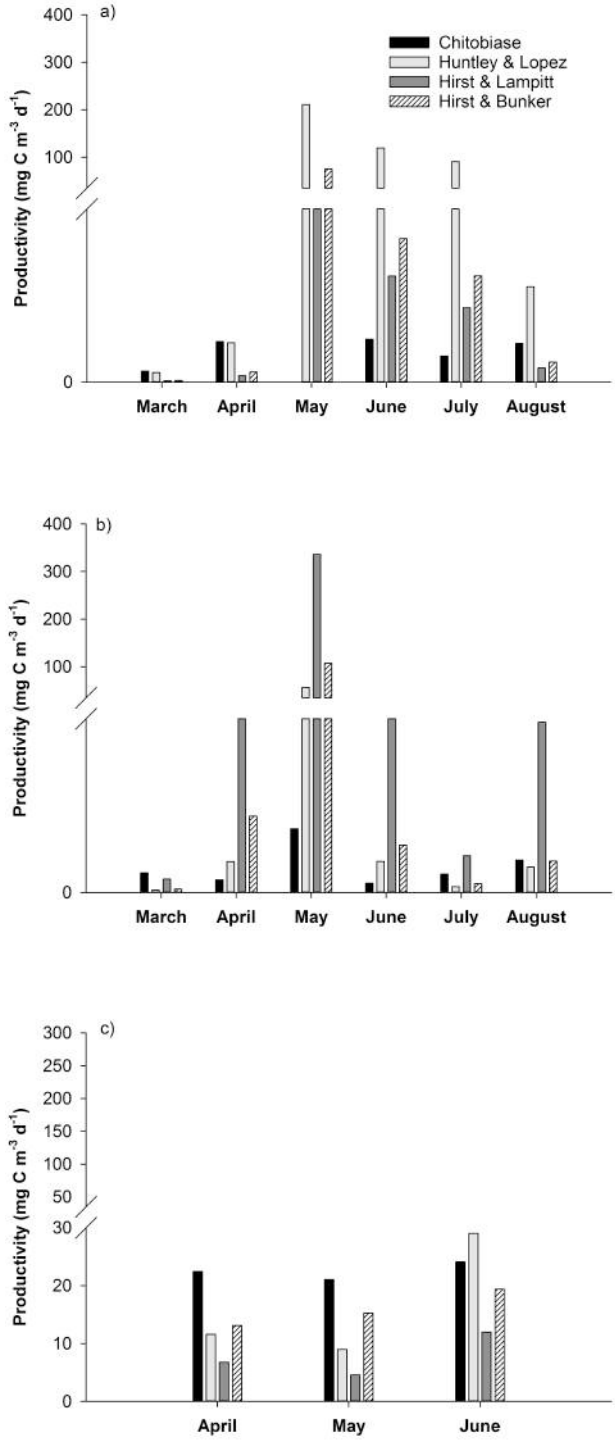


Figure 22. Comparison of chitobiase-based productivity estimates with global predictive models for a) 2010 and b) 2011 in Saanich Inlet, Canada and b) Guanabara Bay, Brazil.

daily P:B and productivity, on the other hand, include the growth rates of all crustaceans, including the smallest and largest individuals, thereby making it more of a "community-level" estimate. In addition, because the models rely on biomass estimates to calculate productivity, the numerous biases associated with net selection come into play (Hopcroft et al. 2001, Turner et al. 2004), which may result in greater variations in productivity estimates compared to variations observed in the growth rates predicted by the models themselves. As a result, the lack of consistency as to how biomass estimates are obtained (e.g. different mesh sizes of zooplankton nets) undoubtedly makes it difficult to compare model-based productivity estimates with other productivity estimates across oceanic regions. Furthermore, although the Hirst and Bunker (2003) model includes food availability (represented by chlorophyll *a* concentration) as a model parameter, none of the models ever includes food quality. Therefore, it is possible that some of the divergences in our chitobiase-productivity estimates compared to the model results may be due to variations in food *quality* as opposed to food quantity, *per se*.

5.3 Moving beyond the use of chlorophyll *a* as a proxy for food availability

Despite the fact that relationships between chlorophyll *a* and zooplankton biomass, growth rates, and productivity remain a contentious issue, many oceanographic studies still continue to use chlorophyll *a* as a proxy for food availability (e.g. Henriksen et al. 2012, Head et al. 2013). Although chlorophyll *a* is used to determine whether or not food is a limiting factor for copepod growth and development, copepods and other crustaceans may always be able to find sufficient

food in coastal and/or eutrophic regions such as those examined in this thesis. In addition, chlorophyll *a* varies on timescales much shorter than zooplankton growth and thus measurements of chlorophyll *a* concentration at the time of zooplankton capture may be somewhat meaningless. Moreover, even though measurements of size-fractionated chlorophyll *a* give a rough idea as to the size of phytoplankton particles available to consumers, chlorophyll *a* does not provide information on the quality of food available to copepods and other crustaceans. Indeed, crustacean abundance, biomass, and chitobiase-based productivity estimates were unrelated to measurements of chlorophyll *a* in Guanabara Bay (Chapter 4). These results thus emphasize the fact that chlorophyll *a* is an inadequate proxy for food availability, particularly in regions dominated by the microbial loop, given that many crustaceans also feed on non-photosynthetic organisms such as ciliates and heterotrophic nanoflagellates, which are not represented by measurements of chlorophyll *a*.

Our estimates of crustacean productivity fueled by the microbial loop in Guanabara Bay were often higher than those reported for Saanich Inlet. These results raise questions about the potential contribution of the microbial loop to the overall production of the planktonic crustacean community in regions dominated by the classical food web. A few studies have shown that the microbial food web may, at times, be the main food source for copepods (Kleppel et al. 1991) and thus intermittently contribute to copepod production (e.g. Kleppel et al. 1991, Kiørboe and Nielsen 1994), even in coastal upwelling regions (Vargas et al. 2007). Yet, it remains unknown as to how much of the crustacean productivity observed in temperate regions is supported directly by primary productivity versus production from the

microbial loop. Future studies would benefit from a comparison of chitobiase-based crustacean productivity with estimates of both ciliate and primary productivity. Specifically, the simultaneous use of fatty acid analyses would help to determine what crustaceans are feeding on, and thus the relative contributions of the classical food chain and the microbial loop to total community-level crustacean productivity in a given region.

5.4 Does food quality matter for crustaceans in a natural field setting?

One of the major contributions of this work is that it shows chitobiase-based productivity estimates can be used to directly test whether or not changes in diet/food quality affect community-level crustacean productivity. In the feeding experiment (Chapter 2), we found that the chitobiase method is sensitive enough to detect variations in growth rates, stage durations, and productivity for a single copepod species in response to different phytoplankton diets. Nevertheless, the true test of these observations hinges on whether or not simple lab manipulations of diet can be extended to a field setting where consumers are exposed to a much wider prey field of differing nutritional quality. To date, poor food quality has been shown to contribute to low copepod growth rates (Klein Breteler et al. 2005) and crustacean productivity (El-Sabaawi et al., 2009, Sastri and Dower, 2009). However, these relationships have thus far only been determined for single copepod species as opposed to the entire planktonic crustacean community. Therefore, Chapter 3 explicitly tested this relationship in a field setting in order to investigate the impact of short-term shifts in food quality on the productivity of the entire crustacean zooplankton community.

Low crustacean productivity was associated with high abundances of diatoms in both the feeding experiment (Chapter 2) and in Saanich Inlet (Chapter 3) on several occasions. During our feeding experiment, chitobiase productivity was undetectable on a few sampling dates when *Tigriopus californicus* was reared on diets containing the diatom *Thalassiosira weissflogii*. Similarly, the lowest productivity observed in Saanich Inlet during both years coincided with high abundances of large (>20 μm) diatoms. The results from Saanich Inlet were further supported by the presence of low DHA:EPA ratios (indicative of a diatom-dominated diet) in the fatty acid content of *Calanus marshallae* on these sampling dates (Chapter 3, Fig. 7). In addition, low nutrient concentrations in the water column just prior to the periods of low productivity indicate that herbivorous crustaceans were likely feeding on nutrient-stressed diatoms on, or just prior to, these sampling dates, which has been shown to result in slower growth rates and even developmental arrest in copepods (Klein Breteler et al. 2005). Furthermore, low phytoplankton growth rates on these days indicate that the diatoms were senescent and likely of poorer quality as previous studies have shown that both egg production and hatching success of copepods decreases with the age of the algae being consumed (Jónasdóttir 1994, Jónasdóttir and Kiørboe 1996).

Although low or undetectable crustacean productivity occurred during periods of high diatom abundance, the exact mechanisms by which food quality may have affected crustacean productivity in our studies remains unclear. Overall, *T. californicus* and *C. marshallae* abundance were positively correlated with diatom abundance and thus the high abundances of diatoms and low DHA:EPA ratios

observed in our studies did not appear to affect juvenile growth. Aside from biochemical requirements (e.g. fatty acids), mineral requirements (e.g. phosphorus and nitrogen) appear to be more important for the growth and development of zooplankton (Boersma 2000). Previous studies have shown that imbalances in nutrient stoichiometry affect growth in *Daphnia magna* (Boersma 2000) and *Acartia tonsa* (Malzahn and Boersma 2012). However, given that low productivity estimates were also observed in the feeding experiment wherein diatoms were always nutrient replete and in the exponential growth phase, it is unlikely that these factors were the limiting factors for growth in our studies.

During both the feeding experiment and in Saanich Inlet, the lowest crustacean production rates occurred around the period of highest adult copepod abundance when adults were likely investing energy into egg production rather than growth. As a result, high diatom abundance may have resulted in an unfavourable ratio of essential fatty acids required for egg production and/or hatching success (Jónasdóttir and Kiørboe, 1996, Jónasdóttir et al. 2005). In addition, although neither egg production nor hatching success was examined in this study, it is possible that inhibitory compounds produced by diatoms may have delayed hatching during periods of peak diatom abundance (Ban et al. 1997, Vargas et al. 2006). Short-term exposure to food of poor quality has previously been shown to reduce secondary production (Malzahn and Boersma 2012). Moreover, a previous lab study showed that the high EPA content associated with diatoms resulted in lower growth rates of larval North Sea cod (St. John et al. 2001). Regardless, any limitation of food quality in our studies appeared to be short-lived given that low crustacean productivity

subsequently increased in both the feeding experiment and in Saanich Inlet. Future studies combining measurements of egg production with chitobiase-based productivity are needed to determine whether or not these intermittent periods of low production are related directly to egg production or hatching success during periods of high diatom abundance.

5.5 The need for routine field estimates of planktonic crustacean productivity

5.5.1 Trophic transfer efficiency

After over 60 years of routinely estimating primary productivity to determine how much energy is available to zooplankton consumers, biological oceanographers still generally rely on estimates of zooplankton biomass (or abundance) to estimate the food available to higher trophic levels (e.g. Frederiksen et al. 2006, Nicolas et al. 2014). This, despite the fact that we know that not everything caught in a zooplankton net is suitable food for larval fish and other consumers. Moreover, temporal mismatches occur when instantaneous estimates of zooplankton biomass are used to represent the food available to larval fish given that larval fish condition is a result of what was being consumed days to weeks prior to the time of fish capture. Recently, the collapse of zooplankton populations and resulting poor food productivity have been linked to a decline in growth and survival of major juvenile fish species during years of unfavourable oceanic conditions in the Strait of Georgia, Canada (Beamish et al. 2012, Thomson et al. 2012). Our results from Chapter 3 show that low crustacean productivity can occur even when zooplankton biomass is high and thus highlight the importance of using field-derived crustacean productivity estimates rather than biomass estimates alone. For example, the presence of many late-stage or adult

crustaceans in the water column could result in high biomass estimates despite the fact that the rate at which herbivore biomass is added to the potential energy pool is low. In contrast, low biomass estimates during times of high productivity may occur when many nauplii and early-stage juveniles, which zooplankton nets often fail to capture, are present, thereby resulting in high crustacean growth rates and, ultimately, high rates of energy being transferred up to higher trophic levels.

Furthermore, Chapter 3 also stresses the importance of using crustacean productivity estimates for calculations of trophic transfer efficiency (TTE). Our results indicate that using zooplankton biomass, alone, would have resulted in a failure to capture the interannual and seasonal variability in energy transfer, which may have been underappreciated in the past. This variability, in turn, may have a direct impact on higher trophic levels during critical periods such as the spring and early summer when a match/mismatch between larval fish and their zooplankton prey is more important (Cushing 1990). Although the growth and survival of juvenile fish has already been linked to the peak timing of copepod abundance (e.g. Mackas et al. 2007), future studies should directly examine the link between crustacean *productivity*, periods of high TTE, and the recruitment of juvenile fish. Ideally, field studies examining crustacean productivity and TTE in a given year should be linked to the survival/return of fish stocks with a time lag of one to three years (e.g. Beamish et al. 2012, Thomson et al. 2012). For example, our data from 2011 in Saanich Inlet revealed a match between the timing of the highest TTE values and the hatching of larval Pacific herring, which typically begin feeding in mid-April (Hay et al. 2009). In contrast, the highest TTE values in 2010 occurred at the beginning of March and then

increased again towards the end of May, which may have resulted in a mismatch in terms of when larval fish began feeding in the region. Based on these results, we might expect to see lower returns of fish stocks a few years after a 'mismatch' between peaks in TTE and larval fish hatching. Ultimately, routine estimates of field-derived crustacean productivity will allow oceanographers to determine how temporal and spatial variations in crustacean productivity influence fish production in oceanic regions worldwide. Ideally, productivity estimates should be measured on a weekly or bi-weekly basis in order to obtain reliable annual estimates of crustacean production.

5.5.2 Direct applications of routine crustacean productivity estimates

Routine estimates of planktonic crustacean productivity have direct application to both ecosystem modeling and management practices. For example, while zooplankton biomass has often been integrated into nutrient-phytoplankton-zooplankton (NPZ) models (e.g. Ayden et al. 2005, Daewel et al. 2008), specific processes such as growth and productivity are typically not included, thereby making the energy flow from phytoplankton to higher trophic levels in marine ecosystems difficult to interpret (Zhou et al. 2010). In addition, community-level estimates of productivity can replace zooplankton biomass estimates for a more accurate approach to ecosystem-based fisheries management (deYoung et al. 2004, Ayden et al. 2005, Gaichas et al. 2010). Ecopath, originally described by Polovina (1984), is one of the most widely used software packages for modeling food-web dynamics (Pauly et al. 2000). While Ecopath-based models use biomass and primary production estimates as phytoplankton parameters, zooplankton are still represented only by biomass and a

production/biomass ratio regulated by a mortality rate (e.g. Persad and Webber 2009). Consequently, Ecopath-based models are recommended as a means of predicting the productivity of aquatic ecosystems rather than as a tool for implementing fisheries management practices due to their inability to predict energy efficiency between primary producers and zooplankton (Christensen and Walters 2004). Therefore, incorporating community-level crustacean production estimates as a parameter in future models will provide more accurate estimates of transfer efficiency in a given ecosystem, thereby making them more reliable for ecosystem management purposes. Furthermore, models incorporating estimates of community-level crustacean production can then be coupled with global climate models to better predict the fate of plankton communities in response to variations in climate (Richardson 2008). The ability to track energy flow to zooplankton in response to changes in temperature, food quantity, food quality, and primary production and, ultimately, the impact this could have on a ecosystem level, is one of the greatest challenges biological oceanographers will face in light of the future warming scenarios predicted by global climate change.

Bibliography

Aberle N, Bauer B, Lewandowska A, Gaedke U, Sommer U (2012) Warming induces shifts in microzooplankton phenology and reduces time-lags between phytoplankton and protozoan production. Marine Biology 159:2441-2453

Allen SE, Wolfe MA (2013) Hindcast of the timing of the spring phytoplankton bloom in the Strait of Georgia, 1968-2010. Progress in Oceanography 115:6-13

Ara K (2001) Length-weight relationships and chemical content of the planktonic copepods in the Cananéia Lagoon estuarine system, São Paulo, Brazil. Plankton Biology and Ecology 48:121-127

Ara K (2002) Temporal variability and production of *Temora turbinata* (Copepoda: Calanoida) in the Cananéia Lagoon estuarine system, São Paulo, Brazil. Scientia Marina 66:399-406

Ara K, Hiromi J (2007) Temporal variability in primary and copepod production in Sagami Bay, Japan. Journal of Plankton Research 29:i85-i96

Arendt KE, Jónasdóttir SH, Hansen PJ, Gärtner S (2005) Effects of dietary fatty acids on the reproductive success of the calanoid copepod *Temora longicornis*. Marine Biology 146:513-530

Arendt KE, Juul-Pedersen T, Mortensen J, Blicher ME (2013) A 5-year study of seasonal patterns in mesozooplankton community structure in a sub-Arctic fjord reveals dominance of *Microsetella norvegica* (Crustacea, Copepoda). Journal of Plankton Research 35:105-120

Avila TR, Machado AAS, Bianchini A (2011) Chitobiase of planktonic crustaceans from South Atlantic coast (Southern Brazil): characterization and influence of abiotic parameters on enzyme activity. Journal of Experimental Marine Biology and Ecology 407:323-329

Avila TR, Machado AAS, Bianchini A (2012) Estimation of zooplankton secondary production in estuarine waters: Comparison between the enzymatic (chitobiase) method and mathematical models. Journal of Experimental Marine Biology and Ecology 416:144-152

Aydin KY, McFarlane GA, King JR, Megrey BA, Myers KW (2005) Linking oceanic food webs to coastal production and growth rates of Pacific salmon (*Oncorhynchus* spp.), using models on three scales. Deep-Sea Research Part II 52:757-780

Azam F, Fenchel T, Field JG, Gray JS, Meyer-Reil LA, Thingstad F (1983) The ecological role of water-column microbes in the sea. Marine Ecology Progress Series 10:257-263

Ban S, Burns C, Castel J, Chaudron Y, Christou E, Escribano R, Fonda Umani S, Gasparini S, Guerrero Ruiz F, Hoffmeyer M, Ianora A, Kang H, Laabir M, Lacoste A, Miralto A, Ning X, Poulet S, Rodriguez V, Runge J, Shi J, Starr M, Uye S, Wang Y (1997) The paradox of diatom-copepod interactions. Marine Ecology Progress Series 157:287-293

Banse K (1982) Mass-scaled rates of respiration and intrinsic growth in very small invertebrates. Marine Ecology Progress Series 9:281-297

Banse K, Mosher S (1980) Adult body mass and annual production/biomass relationships of field populations. Ecological Monographs 50:355-379

Barwell-Clarke J, Whitney F (1996) Institute of Ocean Sciences nutrient methods and analysis. Canadian Technical Report of Hydrography 182:43

Beamish RJ, Neville C, Sweeting R, Lange K (2012) The synchronous failure of juvenile Pacific salmon and herring production in the Strait of Georgia in 2007 and the poor return of sockeye salmon to the Fraser River in 2009. Marine and Coastal Fisheries 4: 403-414

Beaugrand G, Brander KM, Lindley JA, Souissi S, Reid PC (2003) Plankton effect on cod recruitment in the North Sea. Nature 426:661-664

Bell MV, Dick JR, Anderson TR, Pond DW (2007) Application of liposome and stable isotope tracer techniques to study polyunsaturated fatty acid biosynthesis in marine zooplankton. Journal of Plankton Research 29:417-422

Berggreen U, Hansen B, Kiørboe T (1988) Food size spectra, ingestion and growth of the copepod *Acartia tonsa* during development: implications for determination of copepod production. Marine Biology 99:341-352

Boersma M (2000) The nutritional quality of P-limited algae for *Daphnia*. Limnology and Oceanography 45:1157-1161

Boltovskoy D (1999) South Atlantic Zooplankton. Leiden, Backhuys. 1706p

Bonnet D, Carlotti F (2001) Development and egg production in *Centropages typicus* (Copepoda: Calanoida) fed different food types: a laboratory study. Marine Ecology Progress Series 224:133-148

Brzezinski MA, Nelson DM (1986) A solvent extraction method for the colorimetric determination of nanomolar concentrations of silicic acid in seawater. Marine Chemistry 19:129-151

Brzezinski MA, Nelson DM (1989) Seasonal changes in the silicon cycle within a Gulf Stream warm-core ring. Deep Sea Research Part A:1009-1020

Calbet A, Trepat I, Arin L (2000) Naupliar growth versus egg production in the calanoid copepod *Centropages typicus*. Journal of Plankton Research 22:1393-1402

Campbell RG, Wagner MM, Teegarden GJ, Boudreau CA, Durbin EG (2001) Growth and development rates of the copepod *Calanus finmarchicus* reared in the laboratory. Marine Ecology Progress Series 221:161-183

Castonguay M, Plourde S, Robert D, Runge JA, Fortier L (2008) Copepod production drives recruitment in a marine fish. Canadian Journal of Fisheries and Aquatic Sciences 65:1528-1531

Cervetto G, Gaudy R, Pagano M (1999) Influence of salinity on the distribution of *Acartia tonsa* (Copepoda, Calanoida). Journal of Experimental Marine Biology and Ecology 239:33-45

Chisholm LA, Roff JC (1990a) Size-weight relationships and biomass of tropical neritic copepods off Kingston, Jamaica. Marine Biology 106:71-77

Chisholm LA, Roff JC (1990b) Abundances, growth rates, and production of tropical neritic copepods off Kingston, Jamaica. Marine Biology 106:79-89

Christensen V, Walters CJ (2004) Ecopath with Ecosim: methods, capabilities and limitations. Ecological Modelling 172:109-139

Collins AK, Allen SE, Pawlowicz R (2009) The role of wind in determining the timing of the spring bloom in the Strait of Georgia. Canadian Journal of Fisheries and Aquatic Sciences 66:1597-1616

Crain JA, Miller CB (2001) Effects of starvation on intermolt development in *Calanus finmarchicus* copepodites: a comparison between theoretical models and field studies. Deep-Sea Research Part II 48:551-566

Cushing DH (1990) Plankton production and year-class strength in fish populations: an update of the match/mismatch hypothesis. Advances in Marine Biology 26:249-293

Daase M, Soreide JE, Martynova D (2011) Effects of food quality on naupliar development in *Calanus glacialis* at subzero temperatures. Marine Ecology Progress Series 429:111-124

Daewel U, Peck MA, Schrum C, St John MA (2008) How best to include the effects of climate-driven forcing on prey fields in larval fish individual-based models. Journal of Plankton Research 30:1-5

Daewel U, Hjøllo SS, Huret M, Ji R, Maar M, Niiranen S, Travers-Trolet M, Peck MA, van de Wolfshaar KE (2014) Predation control of zooplankton dynamics: a review of observations and models. ICES Journal of Marine Science 71:254-271

Dalsgaard J, St John MA, Kattner G, Muller-Navarra D, Hagen W (2003) Fatty acid trophic markers in the pelagic marine environment. Advances in Marine Biology 46:225-340

Davis CS (1984) Predatory control of copepod seasonal cycles on Georges Bank. Marine Biology 82:31-40

Davis, CS, Alatalo P (1992) Effects of constant and intermittent food supply on life-history parameters in a marine copepod. Limnology and Oceanography 37:1618-1639

Devol AH (1981) Vertical distribution of zooplankton respiration in relation to the intense oxygen minimum zones in two British Columbia fjords. Journal of Plankton Research 3:593-602

DeYoung B, Heath M, Werner F, Chai F, Megrey B, Monfray P (2004) Challenges of modeling ocean basin ecosystems. Science 304:1463-1466

Drinkwater KF, Jones EP (1987) Density stratification, nutrient and chlorophyll distributions in the Hudson Strait region during summer and their relation to tidal mixing. Continental Shelf Research 7:599-607

Dugdale RC, Wilkerson F (1986) The use of ^{15}N to measure nitrogen uptake in eutrophic oceans: experimental considerations. Limnology and Oceanography 31:673-689

Edwards M, Richardson AJ (2004) Impact of climate change on marine pelagic phenology and trophic mismatch. Nature 430:881-884

El-Sabaawi R, Dower JF, Kainz M, Mazumder A (2009) Interannual variability in fatty acid composition of the copepod *Neocalanus plumchrus* in the Strait of Georgia, British Columbia. Marine Ecology Progress Series 382:151-161

Espie PJ, Roff JC (1995a) Characterization of chitobiase from *Daphnia magna* and its relation to chitin flux. Physiological Zoology 68:727-748

Espie PJ and Roff JC (1995b) A biochemical index of duration of the molt cycle for planktonic Crustacea based on the chitin-degrading enzyme, chitobiase. Limnology and Oceanography 40:1028-1034

Evjemo JO, Tokle N, Vadstein O and Olsen Y (2008) Effect of essential dietary fatty acids on egg production and hatching success of the marine copepod *Temora longicornis*. Journal of Experimental Marine Biology and Ecology 365:31-37

Frank KT, Petrie B, Shackell NL, Choi JS (2006) Reconciling differences in trophic control in mid-latitude marine ecosystems. Ecology Letters 9:1096-1105

Frederiksen M, Edwards M, Richardson AJ, Halliday NC, Wanless S (2006) From plankton to top predators: bottom-up control of a marine food web across four trophic levels. Journal of Animal Ecology 75:1259-1268

Fulton J (1968) A laboratory manual for the identification of British Columbia marine zooplankton. No. 55. Queen's Printer

Gaichas SK, Aydin KY, Francis RC (2010) Using food web model results to inform stock assessment estimates of mortality and production for ecosystem-based fisheries management. Canadian Journal of Fisheries and Aquatic Sciences 67:1490-1506

Gallienne CP, Robins DB (2001) Is *Oithona* the most important copepod in the world's oceans? Journal of Plankton Research 23:1421-1432

Gargett AE, Stucchi D, Whitney F (2003) Physical processes associated with high primary production in Saanich Inlet, British Columbia. Estuarine Coastal and Shelf Science 56:1141-1156

Gladyshev MI, Sushchik NN, Anishchenko OV, Makhutova ON, Kolmakov VI, Kalachova GS, Komalova AA, Dubovskaya OP (2011) Efficiency of transfer of essential polyunsaturated fatty acids versus organic carbon from producers to consumers in a eutrophic reservoir. Oecologia 165:521-531

Gomes CL, Marazzo A, Valentin JL (2004) The vertical migration behavior of two calanoid copepods, *Acartia tonsa* Dana, 1849 and *Paracalanus parvus* (Claus, 1863) in a stratified tropical bay in Brazil. Crustaceana 77:941-954

Graeve M, Kattner G, Hagen W (1994a) Diet-induced changes in the fatty acid composition of Arctic herbivorous copepods: Experimental evidence of trophic markers. Journal of Experimental Marine Biology and Ecology 182:97-110

Graeve M, Hagen W, Kattner G (1994b) Herbivorous or omnivorous? On the significance of lipid compositions as trophic markers in Antarctic copepods. Deep-Sea Research Part I 41:915-924

Greve W, Reiners F, Nast J, Hoffmann S (2004) Helgoland Roads meso- and macrozooplankton time-series 1974 to 2004: lessons from 30 years of single spot, high frequency sampling at the only off-shore island of the North Sea. Helgoland Marine Research 58:274-288

Grundle DS, Timothy DA, Varela DE (2009) Variations of phytoplankton productivity and biomass over an annual cycle in Saanich Inlet, a British Columbia fjord. Continental Shelf Research 29:2257-2269

Guenther M, Paranhos R, Rezende CE, Gonzalez-Rodriguez E, Valentin JL (2008) Dynamics of bacterial carbon metabolism at the entrance of a tropical eutrophic bay influenced by tidal oscillation. Aquatic Microbial Ecology 50:123-133

Guenther M, Valentin JL (2008) Bacterial and phytoplankton production in two coastal systems influenced by distinct eutrophication processes. Oecologia Australis 12:172-178

Hama T, Miyazaki T, Ogawa Y, Iwakuma T, Takahashi M, Otsuki A, Ichimura, S. (1983) Measurement of photosynthetic production of a marine phytoplankton population using a stable ^{13}C isotope. Marine Biology 73:31-36

Harrison PJ, Fulton JD, Taylor FJR, Parson TR (1983) Review of the biological oceanography of the Strait of Georgia: pelagic environment. Canadian Journal of Fisheries and Aquatic Sciences 40:1064-1094

Hay DE, McCarter PB, Daniel KS, Schweigert JF (2009) Spatial diversity of Pacific herring (*Clupea pallasi*) spawning areas. ICES Journal of Marine Science 66:1662-1666

Head EJ, Harris LR, Ringuette M, Campbell RW (2013) Characteristics of egg production of the planktonic copepod, *Calanus finmarchicus*, in the Labrador Sea: 1997-2010. Journal of Plankton Research 35:281-298

Heinle DR (1966) Production of a calanoid copepod, *Acartia tonsa*, in the Patuxent River Estuary. Chesapeake Science 7:59-74

Henriksen MV, Jung-Madsen S, Nielsen TG, Møller EF, Henriksen KV, Markager S, Hansen BW (2012) Effects of temperature and food availability on feeding and egg production of *Calanus hyperboreus* from Disko Bay, western Greenland. Marine Ecology Progress Series 447:109-126

Hirst AG, Bonnet D, Harris RP (2007) *Calanus helgolandicus* over two years at a station in the English Channel. Marine Ecology Progress Series 340:189-205

Hirst AG, Bunker AJ (2003) Growth of marine planktonic copepods: Global rates and patterns in relation to chlorophyll *a*, temperature, and body weight. Limnology and Oceanography 45:1988-2010

Hirst AG, Lampitt RS (1998) Towards a global model of in situ weight-specific growth in marine planktonic copepods. Marine Biology 132:247-257

Hirst AG, Sheader M (1997) Are *in situ* weight-specific growth rates body-size independent in marine planktonic copepods? A re-analysis of the global syntheses and a new empirical model. Marine Ecology Progress Series 154:155-165

Hirst AG, Peterson WT, Rothery P (2005) Errors in juvenile copepod growth rate estimates are widespread: problems with the Moult Rate method. Marine Ecology Progress Series 296:263-279

Hopcroft RR, Roff JC (1998a) Zooplankton growth rates: the influence of female size and resources on egg production of tropical marine copepods. Marine Biology 132:79-86

Hopcroft RR, Roff JC (1998b) Zooplankton growth rates: the influence of size in nauplii of tropical marine copepods. Marine Biology 132:87-96

Hopcroft RR, Roff JC, Lombard D (1998a) Production of tropical copepods in Kingston Harbour, Jamaica: the importance of small species. Marine Biology 130:593-604

Hopcroft RR, Roff JC, Webber MK, Witt JDS (1998b) Zooplankton growth rates: the influence of size and resources in tropical marine copepodites. Marine Biology 132:67-77

Hopcroft RR, Roff JC, Chavez FP (2001) Size paradigms in copepod communities: a re-examination. Hydrobiologia 453:133-141

Hovekamp S (1989) Avoidance of nets by *Euphausia pacifica* in Dabob Bay. Journal of Plankton Research 11:907-924

Huntley M, Boyd C (1984) Food-limited growth of marine zooplankton. The American Naturalist 124:455-478

Huntley ME, Lopez MDG (1992) Temperature-dependent production of marine copepods: a global synthesis. The American Naturalist 140:201-242

Hutchings L, Verheye HM, Mitchell-Innes BA, Peterson WT, Huggett JA, Painting SJ (1995) Copepod production in the southern Benguela system. ICES Journal of Marine Science 52:439-455

Irvine JR, Crawford WR (2012) State of the physical biological, and selected fishery resources of Pacific Canadian marine ecosystems in 2011. Department of Fisheries and Oceans Canadian Science Advisory Secretariat Research Document 2012/072. xi+142 p

Ismar SMH, Hansen T, Sommer U (2008) Effect of food concentration and type of diet on *Acartia* survival and naupliar development. Marine Biology 154:335-343

Ji R, Stegert C, Davis CS (2013) Sensitivity of copepod populations to bottom-up and town-down forcing: a modeling study in the Gulf of Maine region. Journal of Plankton Research 35:66-79

Jónasdóttir SH (1994) Effects of food quality on the reproductive success of *Acartia tonsa* and *Acartia hudsonica*: laboratory observations. Marine Biology 121:67-81

Jónasdóttir SH, Kiørboe T (1996) Copepod recruitment and food composition: do diatoms affect hatching success? Marine Biology 125:743-750

Jónasdóttir SH, Trung NH, Hansen F, Gärtner S (2005) Egg production and hatching success in the calanoid copepods *Calanus helgolandicus* and *Calanus finmarchicus* in the North Sea from March to September 2001. Journal of Plankton Research 27:1239-1259

Jónasdóttir SH, Visser AW, Jespersen C (2009) Assessing the role of food quality in the production and hatching of *Temora longicornis* eggs. Marine Ecology Progress Series 382:139-150

Júnior MM, Paranaguá MN, Schwamborn R, Neumann Leitão S, Ekau W (2007) Fluxes of zooplankton biomass between a tidal estuary and the sea in Northeastern Brazil. Brazilian Journal of Oceanography 55:239-249

Kahan D, Berman Y, Bar-El T (1988) Maternal inhibition of hatching at high population densities in *Tigriopus japonicas* (Copepoda, Crustacea). Biological Bulletin 174:139-144

Kainz M, Arts MT, Mazumder A (2004) Essential fatty acids in the planktonic food web and their ecological role for higher trophic levels. Limnology and Oceanography 49:1784-1793

Kimmerer WJ (1987) The theory of secondary production calculations for continuously reproducing populations. Limnology and Oceanography 32:1-13

Kimmerer WJ, McKinnon AD (1987) Growth, mortality, and secondary production of the copepod *Acartia tranteri* in Westernport Bay, Australia. Limnology and Oceanography 32:14-28

Kiørboe T, Johansen K (1986) Studies of a larval herring (*Clupea harengus* L.) patch in the Buchan area. IV. Zooplankton distribution and productivity in relation to hydrographic features. Dana 6:37-51

Kiørboe T, Nielsen TG (1994) Regulation of zooplankton biomass and production in a temperate, coastal ecosystem. 1. Copepods. Limnology and Oceanography 39:493-507

Kjerfve B, Ribeiro CHA, Dias GTM, Filippo AM, Quaresma VS (1997) Oceanographic characteristics of an impacted coastal bay: Baía de Guanabara, Rio de Janeiro, Brazil. Continental Shelf Research 17:1609-1643

Klein Breteler WCM, Schogt N, Gonzalez SR (1990) On the role of food quality in grazing and development of life stages, and genetic change of body size during cultivation of pelagic copepods. Journal of Experimental Marine Biology and Ecology 55:177-189

Klein Breteler WCM, Schogt N, Rampen S (2005) Effect of diatom nutrient limitation on copepod development: role of essential lipids. Marine Ecology Progress Series 291:125-133

Kleppel GS (1993) On the diets of calanoid copepods. Marine Ecology Progress Series 99:183-195

Kleppel GS, Burkart CA (1995) Egg production and the nutritional environment of *Acartia tonsa*: the role of food quality in copepod nutrition. ICES Journal of Marine Science 52:297-304

Kleppel GS, Davis CS, Carter K (1996) Temperature and copepod growth in the sea: a comment on the temperature-dependent model of Huntley and Lopez. The American Naturalist 148:397-406

Knotz S, Boersma M, Saborowski R (2006) Microassays for a set of enzymes in individual small marine copepods. Comparative Biochemistry and Physiology Part A 145:406-411

Koski M, Klein Breteler WCM (2003) Influence of diet on copepod survival in the laboratory. Marine Ecology Progress Series 264:73-82

Koski M, Klein Breteler W, Schogt N (1998) Effect of food quality on rate of growth and development of the pelagic copepod *Pseudocalanus elongatus* (Copepod, Calanoida). Marine Ecology Progress Series 170:169-187

Koski M, Klein Breteler W, Schogt N, Gonzalez S, Jakobsen HH (2006) Life-stage-specific differences in exploitation of food mixtures: diet mixing enhances

copepod egg production but not juvenile development. Journal of Plankton Research 28:919-936

Koski M, Dutz J, Klein Breteler W, Rampen S, Noordeloos A (2010) Seasonal changes in food quantity and quality of the common North Sea copepods *Temora longicornis* and *Pseudocalanus elongatus*: a bioassay approach. Marine Ecology Progress Series 399:141-155

Landry MR (1975) The relationship between temperature and the development of life stages of the marine copepod *Acartia clausi* Giesbr. Limnology and Oceanography 20:854-857

Landry MR (1978) Population dynamics and production of a planktonic marine copepod, *Acartia clausii*, in a small temperate lagoon on San Juan Island, Washington. International Review of Hydrobiology 63:77-119

Lear DW, Oppenheimer C (1962) Consumption of microorganisms by the copepod *Tigriopus californicus*. Limnology and Oceanography 7:63-65

Leggett WC, Deblois E (1994) Recruitment in marine fishes: Is it regulated by starvation and predation in the egg and larval stages? Netherlands Journal of Sea Research 32:119-134

Lewis AG, Chatters L, Raudsepp M (1998) Feeding structures and their functions in adult and preadult *Tigriopus californicus* (Copepoda: Harpacticoida). Journal of the Marine Biological Association of the UK 78:451-466

Lindeman RL (1942) The trophic-dynamic aspect of ecology. Ecology 23:399-417

Liu H, Dagg MJ, Strom S (2005) Grazing by the calanoid copepod *Neocalanus cristatus* on the microbial food web in the coastal Gulf of Alaska. Journal of Plankton Research 27:647-662

Liu H, Hopcroft RR (2006) Growth and development of *Neocalanus flemingeri/plumchrus* in the northern Gulf of Alaska: validation of the artificial-cohort method in cold waters. Journal of Plankton Research 28:87-101

Liu H, Hopcroft RR (2007) A comparison of seasonal growth and development of the copepods *Calanus marshallae* and *C. pacificus* in the northern Gulf of Alaska. Journal of Plankton Research 29:569-581

Liu H, Hopcroft RR (2008) Growth and development of *Pseudocalanus* spp. in the northern Gulf of Alaska. Journal of Plankton Research 30:923-935

Longhurst A (1984) Importance of measuring rates and fluxes in marine ecosystems. In: Fasham MJR (ed) Flows of energy in marine ecosystems. Plenum Press, London. pp 3-22

Lonsdale DJ, Levinton JS (1985) Latitudinal differentiation in copepod growth: an adaptation to temperature. Ecology 66:1397-1407

Mackas DL, Tsuda A (1999) Mesoplankton in the eastern and western subarctic Pacific: community structure, seasonal life histories, and interannual variability. Progress in Oceanography 43:335-363

Mackas DL, Thomson RE, Galbraith M (2001) Changes in the zooplankton community of the British Columbia continental margin, 1985-1999, and their covariation with oceanographic conditions. Canadian Journal of Fisheries and Aquatic Sciences 58:685-702

Mackas DL, Batten S, Trudel M (2007) Effects on zooplankton of a warmer ocean: recent evidence from the Northeast Pacific. Progress in Oceanography 75:223-252

Mackas DL, Greve W, Edwards M, Chiba S, Tadokoro K, Eloire D, Mazzocchi MG, Batten S, Richardson AJ, Johnson C, Head E, Conversi A, Peluso T (2012) Changing zooplankton seasonality in a changing ocean: comparing time series of zooplankton phenology. Progress in Oceanography 97:31-62

Malzahn AM, Boersma M (2012) Effects of poor food quality on copepod growth are dose dependent and non-reversible. Oikos 121:1408-1416

Marazzo A, Valentin JL (2000) Daily variation of marine cladoceran densities in a tropical bay – Brazil. Hydrobiologia 428:205-208

Marazzo A, Valentin JL (2001) Diel changes in embryonic maturation in two species of marine cladocerans in Guanabara Bay, Rio de Janeiro, Brazil. Revista Brasileira de Zoologia 18:353-356

McKinnon AD, Duggan S (2003) Summer copepod production in subtropical waters adjacent to Australia's North West Cape. Marine Biology 143:897-907

McLaren IA, Corkett CJ (1981) Temperature-dependent growth and production by a marine copepod. Canadian Journal of Fisheries and Aquatic Sciences 38:77-83

Miller AJ (1990) Subset selection in regression. Edited by AJ Miller. Chapman and Hall. London, England; New York.

Moore E, Sander F (1976) Quantitative and qualitative aspects of the zooplankton and breeding patterns of copepods at two Caribbean coral reef stations. Estuarine and Coastal Marine Science 4:589-607

Murray MM, Marcus NH (2002) Survival and diapause egg production of the copepod *Centropages hamatus* raised on dinoflagellate diets. Journal of Experimental Marine Biology and Ecology 270:39-56

Muzzarelli RAA (1977) Chitinases and related enzymes. In Chitin. Edited by RAA Muzzarelli. Pergamon, Oxford, UK. pp. 155-177

Neveux J, Lantoine F (1997) Spectrofluorometric assay of chlorophylls and phaeopigments using the least squares approximation technique. Deep-Sea Research I 40:1747-1765

Nicolas D, Rochette S, Llope M, Licandro P (2014) Spatio-temporal variability of the North Sea cod recruitment in relation to temperature and zooplankton. PLoS ONE 9:e88447

Nielsen TG, Andersen CM (2002) Plankton community structure and production along a freshwater-influenced Norwegian fjord system. Marine Biology 141:707-724

Oosterhuis, SS, Baars MA, Klein Breteler WCM (2000) Release of the enzyme chitobiase by the copepod *Temora longicornis*: characteristics and potential tool for estimating crustacean biomass production in the sea. Marine Ecology Progress Series 196:195-206

Paffenhöfer G-A (1984) Food ingestion by the marine planktonic copepod *Paracalanus* in relation to abundance and size distribution of food. Marine Biology 80:323-333

Paffenhöfer G-A, Harris RP (1976) Feeding, Growth and Reproduction of the Marine Planktonic Copepod *Pseudo-Calanus Elongatus* Boeck. Journal of the Marine Biological Association of the UK 56:327-344

Paranhos R, Pereira AP, Mayr LM (1998) Diel variability of water quality in a tropical polluted bay. Environmental Monitoring and Assessment 50:131-141

Parrish C (1999) Determination of total lipids, lipid classes and fatty acids in aquatic samples. In: Arts M, Wainmann BC (ed) Lipids in freshwater ecosystems. Springer, New York

Parsons TR, Maita Y, Lalli CM (1984) A manual of biological and chemical methods for seawater analysis. Pergamon Press, Oxford

Pauly D, Christensen V (1995) Primary production required to sustain global fisheries. Nature 374:255-257

Pauly D, Christensen V, Walters C (2000) Ecopath, Ecosim, and Ecospace as tools for evaluating ecosystem impact of fisheries. ICES Journal of Marine Science 57:697-706

Persad G, Webber M (2009) The use of Ecopath software to model trophic interactions within the zooplankton community of Discovery Bay, Jamaica. The Open Marine Biology Journal 3:95-104

Peters G, Saborowski R, Buchholz F and Mentlein R (1999) Two distinct forms of the chitin-degrading enzyme *N*-acetyl-ß-D-glucosaminidase in the Antarctic krill: specialists in digestion and moult. Marine Biology 134:697-703

Peterson WT, Tiselius P, Kiørboe T (1991) Copepod egg production, moulting and growth rates, and secondary production, in the Skagerrak in August 1988. Journal of Plankton Research 13:131-154

Peterson WT, Gómez-Gutiérrez J, Morgan CA (2002) Cross-shelf variation in calanoid copepod production during summer 1996 off the Oregon coast, USA. Marine Biology 141:353-365

Polovina JJ (1984) Model of a coral reef ecosystem. I: the ECOPATH model and its application to French Frigate Schoals. Coral Reefs 3:1-11

Pomeroy LR (1974) The ocean's food web, a changing paradigm. BioScience 24:499-504

Pomeroy LR, Williams PJ, Azam F, Hobbie JE (2007) The microbial loop. Oceanography 20:28-33

Postel L, Fock H, Hagen W (2000) Biomass and abundance. In: Harris RP, Weibe PH, Lenz J, Skjoldal HR, Huntley M (Eds), ICES Zooplankton Methodology Manual. Academic Press, San Diego, pp. 83-192

Poulet SA, Ianora A, Laabir M, Klein Breteler WCM (1995) Towards the measurement of secondary production and recruitment in copepods. ICES Journal of Marine Science 52:359-368

Powlik JJ, Lewis AG, Spaeth M (1997) Development, body length, and feeding of *Tigriopus californicus* (Copepoda, Harpacticoida) in laboratory and field populations. Crustaceana 70:324-343

Richardson AJ (2008) In hot water: zooplankton and climate change. ICES Journal of Marine Science 65:279-295

Rigler FH, Downing JA (1984) The calculation of secondary production. In: Downing JA, Rigler FH (eds) A manual on the methods for the assessment of secondary

production in fresh waters, 2nd edn. Blackwell Scientific Publications, Oxford, pp 19-58

Roff JC, Kroetsch JT, Clarke AJ (1994) A radiochemical method for secondary production in planktonic crustacean based on rate of chitin synthesis. Journal of Plankton Research 16:961-976

Roman MR, Gauzens AL, Rhinehart WK, White JR (1993) Effects of low oxygen waters on Chesapeake Bay zooplankton. Limnology and Oceanography 38:1603-1614

Ryther JH (1969) Photosynthesis and fish production in the sea. Science 166:72-76

Saiz E, Calbet A, Trepat I, Irigoien X, Alcaraz M (1997) Food availability as a potential source of bias in the egg production method for copepods. Journal of Plankton Research 19:1-14

Sastri AR, Roff JC (2000) Rate of chitobiase degradation as a measure of development rate in planktonic Crustacea. Canadian Journal of Fisheries and Aquatic Sciences 57:1965-1968

Sastri AR, Dower JF (2006) Field validation of an instantaneous estimate of in situ development and growth for marine copepod communities. Canadian Journal of Fisheries and Aquatic Sciences 63:2639-2647

Sastri A, Dower JF (2009) Interannual variability in chitobiase-based production rates of the crustacean zooplankton community in the Strait of Georgia, British Columbia, Canada. Marine Ecology Progress Series 388:147-157

Sastri AR, Nelson RJ, Varela DE, Young KV, Wrohan I, Williams WJ (2012) Variation of chitobiase-based estimates of crustacean production rates in high latitude waters. Journal of Experimental Marine Biology and Ecology 414:54-61

Schabetsberger R, Morgan CA, Brodeur RD, Potts CL, Peterson WT, Emmett RL (2003) Prey selectivity and diel feeding chronology of juvenile chinook (*Oncorhynchus tshawytscha* and coho (*O. kisutch*) salmon in the Columbia River plume. Fisheries Oceanography 12:523-540

Schwamborn R, Bonecker SLC, Galvão IB, Silva TA, Neumann-Leitão S (2004) Mesozooplankton grazing under conditions of extreme eutrophication in Guanabara Bay, Brazil. Journal of Plankton Research 26:983-992

Sherr E, Sherr B (1988) Role of microbes in pelagic food webs: a revised concept. Limnology and Oceanography 33:1225-1227

Shreeve RS, Ward P, Whitehouse MJ (2002) Copepod growth and development around South Georgia: relationships with temperature, food and krill. Marine Ecology Progress Series 233:169-183

Sommer U, Stibor H, Katechakis A, Sommer F, Hansen T (2002) Pelagic food web configurations at different levels of nutrient richness and their implications for the ratio fish production:primary production. Hydrobiologia 484:11-20

Sommer U, Aberle N, Engel A, Hansen T, Lengfellner K, Sandow M, Wohlers J, Zöllner E, Riebesell U (2007) An indoor mesocosm system to study the effect of climate change on the late winter and spring succession of Baltic Sea phyto- and zooplankton. Oecologia 150:655-667

Steeman-Nielsen E (1952) The use of radio-active carbon (14C) for measuring organic production in the sea. Journal du Conseil/Conseil Permanent International pour l'Exploration de la Mer. 18:117-140

Stevens CJ, Deibel D, Parrish CC (2004) Species-specific differences in lipid composition and omnivory indices in Arctic copepods collected in deep water during autumn (North Water Polynya). Marine Biology 144:905-915

St John MA, Clemmesen C, Lund T, Köster T (2001) Diatom production in the marine environment: implications for larval fish growth and condition. ICES Journal of Marine Science 58:1106-1113

Suchy KD, Dower JF, Sastri AR, Neil MC (2013) Influence of diet on chitobiase-based production rates for the harpacticoid copepod Tigriopus californicus. Journal of Plankton Research 35:657-667

Takahashi M, Seibert DL, Thomas WH (1977) Occasional blooms of phytoplankton during summer in Saanich Inlet, BC, Canada. Deep-Sea Research 24:775-780

Tenório MMB, Le Borgne R, Rodier M, Neveux J (2005) The impact of terrigeneous inputs on the Bay of Ouinné (New Caledonia) phytoplankton communities: A spectrofluorometric and microscopic approach. Estuarine and Coastal Shelf Science 64:531-545

Thomson RE, Beamish RJ, Beacham TD, Trudel M, Whitfield PH, Hourston RAS (2012) Anomalous ocean conditions may explain the recent extreme variability in Fraser River sockeye salmon production. Marine and Coastal Fisheries 4:415-437

Timothy DA, Soon MYS (2001) Primary production and deep-water oxygen content of two British Columbian fjords. Marine Chemistry 73:37-51

Tommasi DAG, Routledge RD, Hunt BPV, Pakhomov EA (2013a) The seasonal development of the zooplankton community in a British Columbia (Canada) fjord

during two years with different spring bloom timing. Marine Biology Research 9:129-144

Tommasi, D, Hunt BPV, Pakhomov EA, Mackas DL (2013b) Mesozooplankton community seasonal succession and its drivers: insights from a British Columbia, Canada, fjord. Journal of Marine Systems 115:10-32

Turner JT (2004) The importance of small planktonic copepods and their roles in pelagic marine food webs. Zoological Studies 43:255-266

Utermöhl H (1958) Zur Vervollkommnung der quantitativen Phytoplankton – Methodik. Mitteilung Internationale Vereinigung fuer Theoretische unde Amgewandte Limnologie 9:1-38

Uye S (1982) Length-weight relationships of important zooplankton from the Inland Sea of Japan. Journal of the Oceanographical Society of Japan 38:149-158

Uye S (1988) Temperature-dependent development and growth of *Calanus sinicus* (Copepoda: Calanoida) in the laboratory. Hydrobiologia 167:285-293

Valentin JL, Marazzo A (2003) Modelling the population dynamics of *Penilia avirostris* (Branchiopoda, Ctenopoda) in a tropical bay. Acta Oecologica 24:S369-S376

Vargas CA, Escribano R, Poulet S (2006) Phytoplankton food quality determines time windows for successful zooplankton reproductive pulses. Ecology 87:2992-2999

Vargas CA, Martinez RA, Cuevas LA, Pavez MA, Cartes C, González HE, Escribano R, Daneri G (2007) The relative importance of microbial and classical food webs in a highly productive coastal upwelling area. Limnology and Oceanography 52:1495-1510

Vargas CA, Martinez RA, Escribano R, Lagos NA (2010) Seasonal relative influence of food quantity, quality, and feeding behaviour on zooplankton growth regulation in coastal food webs. Journal of the Marine Biological Association of the UK 90:1189-1201

Vidal J (1980a) Physioecology of zooplankton. I. Effects of phytoplankton concentration, temperature and body size on the growth rate of *Calanus pacificus* and *Pseudocalanus* sp. Marine Biology 56:111-134

Vidal J (1980b) Physioecology of zooplankton. II. Effects of phytoplankton concentration, temperature, and body size on the development and molting rates of *Calanus pacificus* and *Pseudocalanus* sp. Marine Biology 56:135-146

Viso AC, Marty JC (1993) Fatty acids from 28 marine microalgae. Phytochemistry 34:1525-1533

Wagner M, Durbin E, Buckley L (1998) RNA:DNA ratios as indicators of nutritional condition in the copepod *Calanus finmarchicus*. Marine Ecology Progress Series 162:173-181

Ware DM (1975) Relation between egg size, growth and natural mortality of larval fish. Journal of the Fisheries Research Board of Canada 32:2503-2512

Ware DM, Thomson RE (2005) Bottom-up ecosystem trophic dynamics determine fish production in the Northeast Pacific. Science 308:1280-1284

Webber MK, Roff JC (1995) Annual biomass and production of the oceanic copepod community off Discovery Bay, Jamaica. Marine Biology 123:481-495

Yang EJ, Kang H-K, Yoo S, Hyun J-H (2009) Contribution of auto- and heterotrophic protozoa to the diet of copepods in the Ulleung Basin, East Sea/Japan Sea. Journal of Plankton Research 31:647-659

Yebra L, Hernández-León S (2004) Aminoacyl-tRNA synthetases activity as a growth index in zooplankton. Journal of Plankton Research 26:351-356

Yebra L, Harris RP, Smith T (2005) Comparison of five methods for estimating growth of *Calanus helgolandicus* later developmental stages (CV-CVI). Marine Biology 147:1367-1375

Yoshioka PM, Owen GP, Pesante D (1985) Spatial and temporal variations in Caribbean zooplankton near Puerto Rice. Journal of Plankton Research 6:733-751

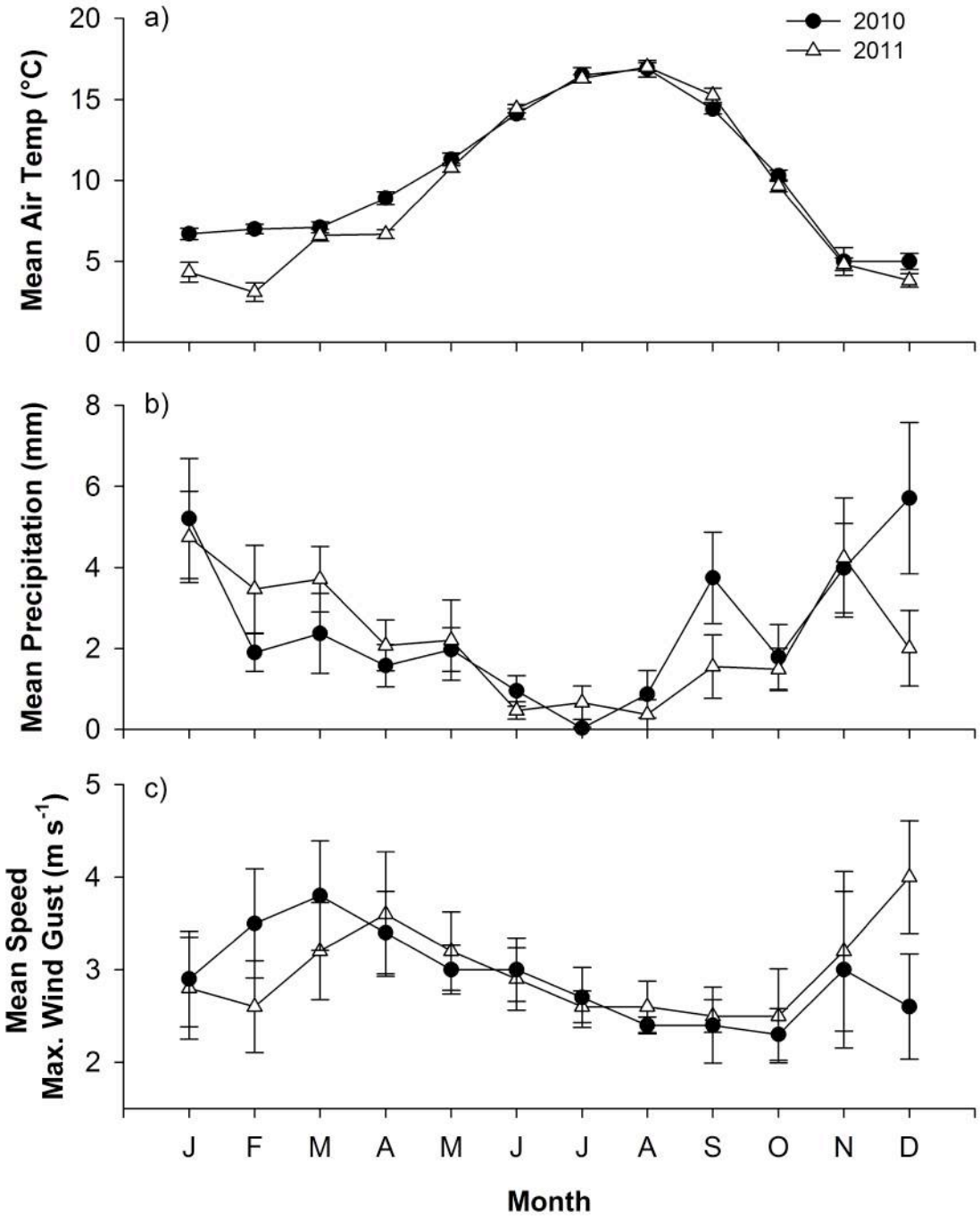
Youngbluth MJ (1980) Daily, seasonal, and annual fluctuations among zooplankton populations in an unpolluted tropical embayment. Estuarine and Coastal Marine Science 10:265-287

Zhou M, Carlotti F, Zhu Y (2010) A size-spectrum zooplankton closure model for ecosystem modelling. Journal of Plankton Research 32:1147-1165

Appendix A: Terminology and equations associated with the chitobiase method

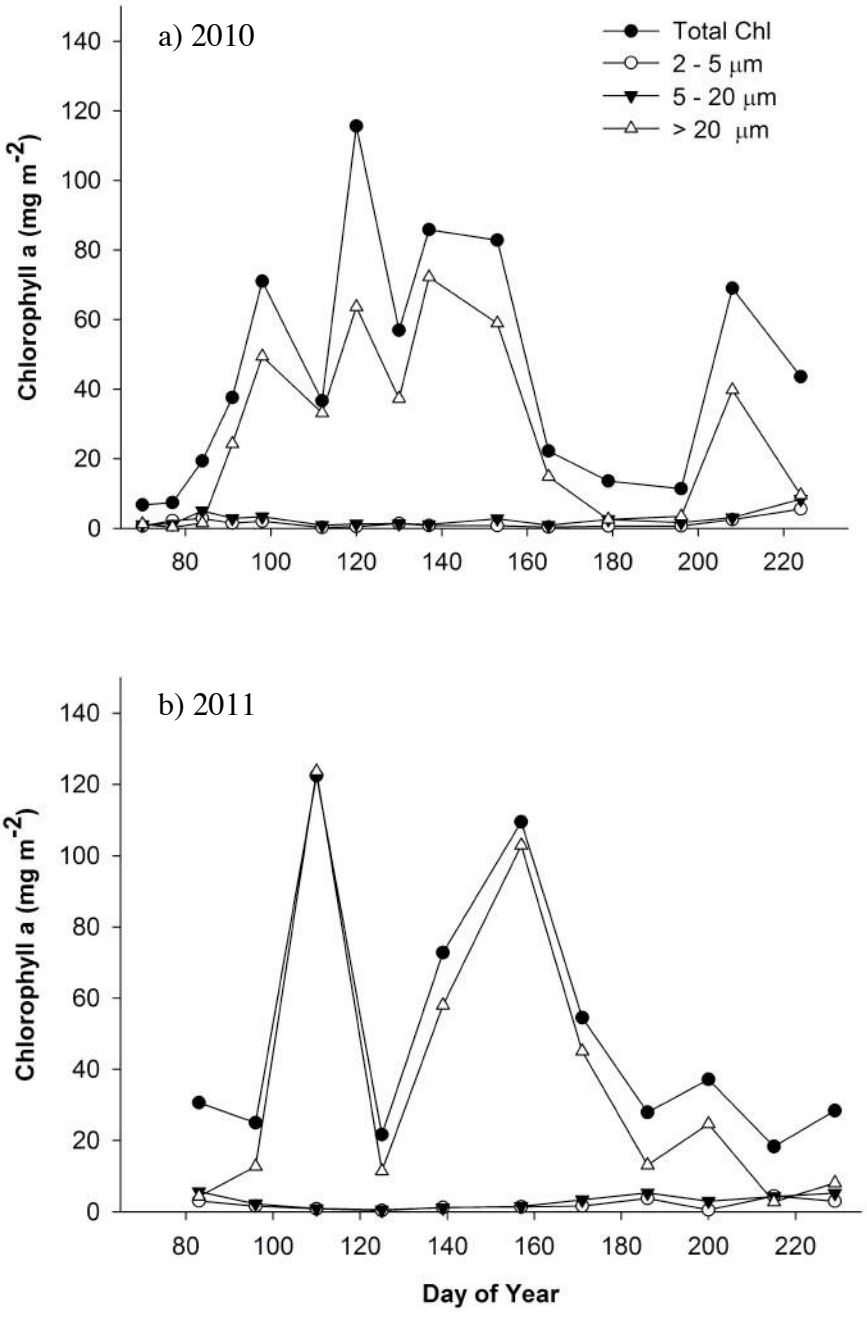
Chitobiase Terminology	Definition	Equation
CBA (nmol MBF L^{-1} h^{-1})	Chitobiase activity liberated into the water by moulting crustaceans.	
CBA_{nat} (nmol MBF L^{-1} h^{-1})	The native *in situ* chitobiase activity measured before seawater samples are "spiked" with homogenate.	
TCBA or stage duration (days)	The time taken for moulting individuals to produce CBA equivalent to the CBA_{nat}.	[1/(-k)]/24 where k = slope of lnCBA vs. time
ΔB or g_{inc} (mg)	The absolute amount of biomass produced by crustaceans calculated using a known relationship between CBA and the growth increment of marine copepods (Sastri and Dower 2006).	$\log (g_{inc}) = 0.864 \log(CBA_i) - 1.78$ where $CBA_i = CBA_{nat}$
Crustacean productivity (mg C m^{-3} d^{-1})	The rate of increase in the biomass of crustaceans.	$\Delta B/TCBA$
CBA-biomass (mg C m^{-3})	Chitobiase-based biomass calculated using a known relationship between individual body weight and CBA (Sastri and Dower 2009).	$\log_e(CBA) = 1.55 \log_e(DW) + 5.60$
Daily P:B	Ratio of daily production to biomass	Crustacean productivity/CBA-biomass

Appendix B: Monthly mean air temperature, precipitation, and mean wind speed for Chapter 3

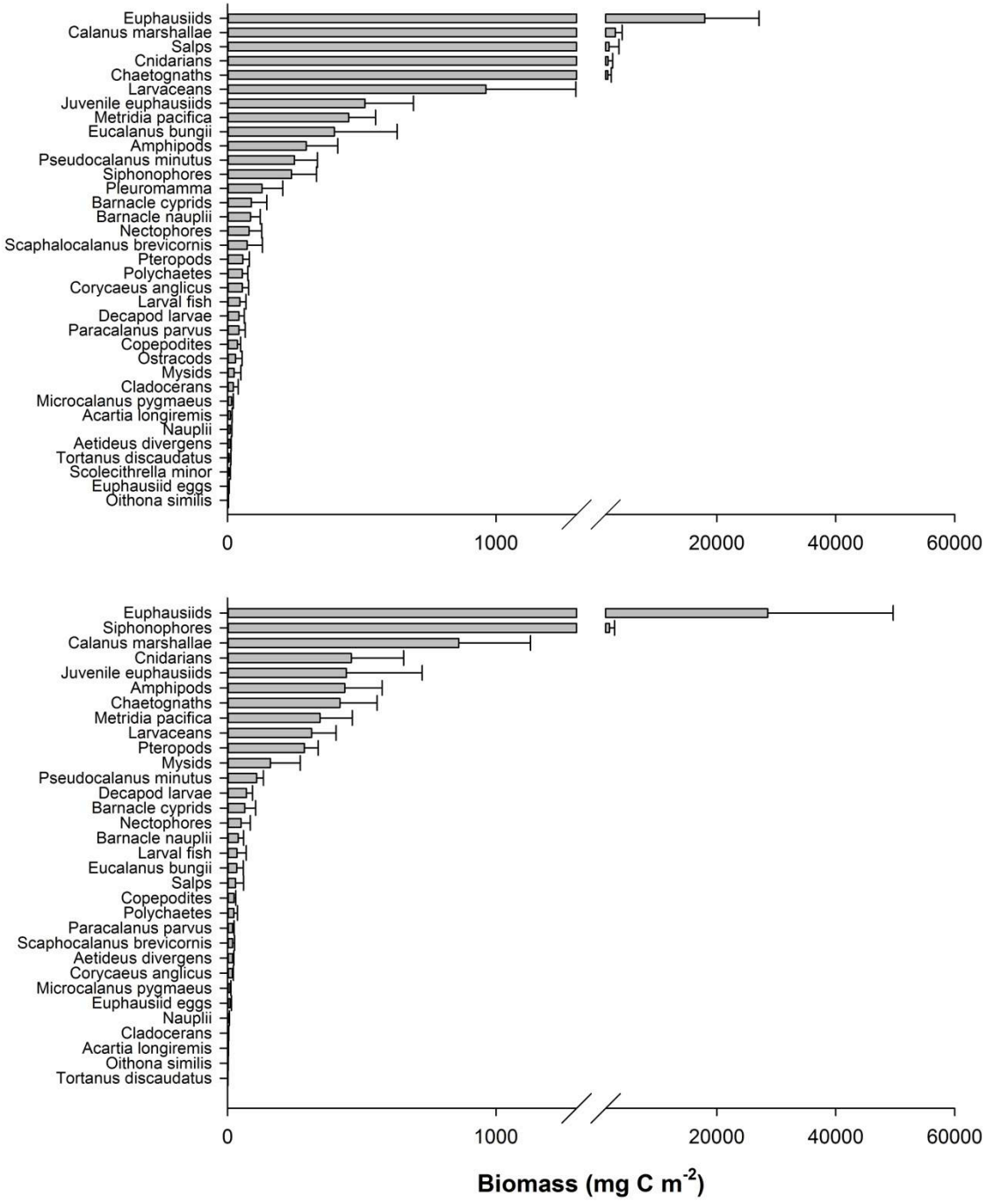


Error bars are ± SE. Data provided by the Environment Canada station at the Victoria International Airport near our sampling location in Saanich Inlet, BC.

Appendix C: Depth-integrated total and size-fractionated chlorophyll *a* for Chapter 3



Appendix D: Average depth-integrated biomass over the entire sampling season for Chapter 3



Appendix E: Fatty acid composition of *Calanus marshallae* from Chapter 3

2010	11-Mar	18- Mar	25-Mar	01-Apr	08-Apr	22-Apr	30-Apr
Saturated fatty acids							
14:00	13.0±n/a	9.4±n/a	7.7±0.6	6.9±1.3	8.6±1.0	7.2±n/a	14.7±2.6
16:00	24.1±n/a	24.4±n/a	25.4±0.0	24.4±1.4	27.0±2.6	25.2±n/a	16.0±0.4
18:00	5.1±n/a	6.5±n/a	12.3±4.0	11.3±0.7	16.2±1.9	27.0±n/a	7.9±1.4
20:01	3.3±n/a	9.3±n/a	5.0±1.7	0.6±0.1	1.6±1.1	0.7±n/a	3.8±1.3
Sub-total	45.5	49.6	50.4	43.2	53.4	60.1	42.4
Monounsaturated fatty acids							
16:1n-7 (diatoms)	8.9±n/a	5.9±n/a	5.5±0.2	7.1±0.4	5.8±0.3	4.9±n/a	7.2±1.1
18:1n-9 (carnivory/omnivory)	0.5±n/a	1.0±n/a	5.8±1.4	7.7±2.5	2.4±2.0	0.7±n/a	6.2±0.3
Sub-total	9.4	6.9	11.3	14.8	8.2	5.6	13.4
Polyunsaturated fatty acids							
18:2n-6 (green algae)	0.5±n/a	1.0±n/a	3.3±2.3	2.2±1.6	0.5±0.1	0.7±n/a	7.7±0.4
20:5n-3 (diatoms)	19.5±n/a	17.4±n/a	13.5±0.3	20.0±1.2	19.8±2.2	18.1±n/a	24.3±0.8
22:6n-3 (dinoflagellates)	25.1±n/a	25.3±n/a	21.5±1.7	19.7±1.0	18.1±0.3	15.6±n/a	12.2±0.2
Sub-total	45.1	43.7	38.3	42.0	38.4	34.3	44.2

2010 (cont'd)	10-May	17-May	02-Jun	14-Jun	28-Jun	12-Jul	27-Jul	12-Aug
Saturated fatty acids								
14:00	12.8±n/a	10.3±0.5	14.5±1.0	15.0±1.6	12.8±1.2	15.5±5.8	4.3±1.8	5.6±0.4
16:00	13.4±n/a	15.5±1.0	13.1±0.1	13.4±1.2	20.7±2.6	23.4±2.7	31.2±0.4	28.9±0.7
18:00	3.9±n/a	6.5±1.1	3.9±0.3	3.2±0.3	7.0±0.8	8.3±1.6	17.7±2.6	14.7±2.0
20:01	6.1±n/a	2.9±0.4	6.8±0.7	5.5±0.9	4.7±1.4	2.6±2.3	0.7±0.1	0.7±0.0
Sub-total	36.1	35.2	38.4	37.2	45.1	49.8	54.0	49.8
Monounsaturated fatty acids								
16:1n-7 (diatoms)	13.4±n/a	12.1±0.3	9.3±0.2	12.1±0.6	8.9±1.4	6.9±2.5	0.0±0.0	5.6±0.2
18:1n-9 (carnivory/omnivory)	5.5±n/a	4.4±0.2	6.2±0.1	6.4±0.5	5.1±0.5	4.2±1.9	0.7±0.1	1.7±1.0
Sub-total	18.9	16.5	15.5	18.5	14.0	11.1	0.7	7.3
Polyunsaturated fatty acids								
18:2n-6 (green algae)	7.2±n/a	5.5±0.3	6.4±0.0	6.1±0.7	3.1±1.6	3.2±1.4	3.1±1.1	0.7±0.0
20:5n-3 (diatoms)	28.2±n/a	31.4±1.8	29.8±1.8	28.2±1.9	20.9±0.9	17.2±2.2	17.5±1.7	20.5±1.0
22:6n-3 (dinoflagellates)	9.6±n/a	11.4±0.6	9.8±0.0	9.9±2.1	16.9±2.9	18.7±4.0	24.8±2.0	21.7±0.7
Sub-total	45.0	48.3	46.1	44.3	40.8	39.1	45.3	42.8

2011		24-Mar	06-Apr	20-Apr	05-May	19-May	06-Jun
Saturated fatty acids							
14:00		21.4±2.5	11.8±1.1	21.2±3.4	13.2±1.6	14.7±0.5	19.2±0.7
16:00		19.6±1.2	25.8±1.5	24.5±1.9	18.3±1.1	14.0±0.4	11.5±1.2
18:00		4.3±0.6	4.6±0.8	3.6±0.5	2.6±0.2	1.6±0.1	1.2±0.1
20:01		5.4±0.9	1.7±0.5	0.8±0.2	2.9±0.7	4.7±0.4	7.8±0.5
	Sub-total	50.6	43.9	50.1	36.9	34.9	39.6
Monounsaturated fatty acids							
16:1n-7 (diatoms)		10.3±0.9	7.4±0.6	5.8±0.2	11.1±0.5	12.9±0.9	13.9±0.7
18:1n-9 (carnivory/omnivory)		3.0±0.3	2.7±0.6	3.9±0.1	4.8±0.1	4.3±0.3	6.4±0.2
	Sub-total	13.3	10.2	0.7	15.9	17.2	20.3
Polyunsaturated fatty acids							
18:2n-6 (green algae)		0.5±0.1	0.6±0.2	3.9±0.9	2.9±0.4	0.8±0.5	0.1±0.0
20:5n-3 (diatoms)		16.7±0.9	21.8±1.3	17.6±0.3	28.4±0.6	30.8±1.0	28.7±1.4
22:6n-3 (dinoflagellates)		18.8±1.2	23.5±0.7	18.6±2.6	16.0±1.5	16.8±0.7	11.2±1.8
	Sub-total	36.0	45.9	40.1	47.2	47.9	40.1

2011 (cont'd)		20-Jun	05-Jul	19-Jul	03-Aug	17-Aug
Saturated fatty acids						
14:00		14.6±0.9	8.8±4.1	9.7±2.4	13.1±2.2	3.5±0.7
16:00		16.0±0.9	24.0±4.7	21.7±1.8	18.2±1.9	22.6±0.1
18:00		1.9±0.2	2.7±0.6	4.9±0.6	4.0±1.1	9.9±0.4
20:01		4.2±1.5	3.4±2.6	1.5±0.3	3.3±1.8	0.6±0.1
	Sub-total	36.8	38.9	37.8	38.7	36.5
Monounsaturated fatty acids						
16:1n-7 (diatoms)		11.8±0.6	6.6±2.6	5.8±0.2	13.8±1.2	3.1±0.4
18:1n-9 (carnivory/omnivory)		7.4±1.2	4.2±0.2	2.2±0.1	4.8±0.5	3.6±0.5
	Sub-total	19.3	10.9	8.0	18.6	6.7
Polyunsaturated fatty acids						
18:2n-6 (green algae)		0.3±0.0	1.3±0.9	1.0±0.0	0.3±0.0	0.7±0.0
20:5n-3 (diatoms)		25.7±2.1	21.9±2.2	19.6±1.8	20.7±1.8	14.9±0.3
22:6n-3 (dinoflagellates)		18.0±1.8	27.0±5.9	33.5±2.2	21.8±2.8	41.2±0.2
	Sub-total	43.9	50.2	54.1	42.8	56.8

Appendix F: Depth-integrated particulate concentrations and ratios for Chapter 3

	Day of Year	$bSiO_2$ (mmol m^{-2})	POC (mmol m^{-2})	PON (mmol m^{-2})	$bSiO_2$:POC	$bSiO_2$:PON
2010						
11-Mar	70	13.72	137.66	24.31	0.10	0.56
18-Mar	77	6.97	94.63	16.71	0.07	0.42
25-Mar	84	18.79	187.66	33.18	0.10	0.57
01-Apr	91	68.84	347.80	55.30	0.20	1.24
08-Apr	98	98.55	377.99	60.38	0.26	1.63
22-Apr	112	47.25	460.61	78.75	0.10	0.60
30-Apr	120	112.58	977.59	111.59	0.12	1.01
10-May	130	25.67	674.19	48.27	0.04	0.53
17-May	137	87.00	578.11	91.75	0.15	0.95
02-Jun	153	69.20	541.98	85.76	0.13	0.81
14-Jun	165	26.58	513.43	76.52	0.05	0.35
28-Jun	179	11.30	312.75	60.90	0.04	0.19
15-Jul	196	6.26	302.69	54.52	0.02	0.11
27-Jul	208	80.36	819.95	105.85	0.10	0.76
12-Aug	224	22.37	330.40	50.66	0.07	0.44
2011						
24-Mar	83	6.71	255.65	24.91	0.03	0.27
06-Apr	96	12.79	217.57	28.66	0.06	0.45
20-Apr	110	16.46	981.23	17.75	0.02	0.93
05-May	125	10.14	324.07	20.53	0.03	0.49
19-May	139	25.45	543.08	24.94	0.05	1.02
06-Jun	157	11.21	693.85	73.28	0.02	0.15
20-Jun	171	11.32	472.48	67.65	0.02	0.17
05-Jul	186	8.63	377.28	153.94	0.02	0.06
19-Jul	200	22.16	402.06	93.61	0.06	0.02
03-Aug	215	5.29	347.86	264.93	0.02	0.02
17-Aug	229	6.22	460.16	79.63	0.01	0.08

Appendix G: Equations for global predictive models used to calculate growth rates and productivity

Equation	Reference
$g = 0.0445e^{0.111T}$	Huntley & Lopez 1992
$\log_{10}g = 0.0208[T]-0.3221[\log_{10}BW]-1.1408$	Hirst & Lampitt 1998
$\log_{10}g = 0.0186[T]-0.288[\log_{10}BW]+0.417[\log_{10}\text{Chl } a]-1.209$	Hirst & Bunker 2003

T = temperature (ºC)
BW = body weight (µg C ind^{-1})
Chl *a* = chlorophyll *a* (µg Chl *a* L^{-1})