

Interannual variability in the relationship between *in situ* primary productivity and somatic crustacean productivity in a temperate fjord

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ABSTRACT: Simultaneous and precise measurements of primary and secondary productivity are required when examining energy transfer from phytoplankton to zooplankton. We examined the relationship between primary and crustacean productivity over 2 yr in Saanich Inlet, British Columbia, Canada, to determine how temporal variations in primary productivity influence crustacean productivity and trophic transfer efficiency (TTE). Primary productivity was measured via the ¹³C tracer method, while the crustacean moulting enzyme chitinase was used to estimate community-level somatic crustacean productivity. Peak primary productivity occurred much earlier in 2010 (late April; 9.17 g C m⁻² d⁻¹) than in 2011 (mid-June; 5.01 g C m⁻² d⁻¹) due to a higher abundance of diatoms. Fatty acid analyses revealed that one of the dominant copepods, *Calanus marshallae*, was feeding on a higher proportion of diatoms than dinoflagellates (lowest DHA:EPA ratios) in May 2010 and June 2011. Crustacean productivity ranged between 0.01 and 0.65 g C m⁻² d⁻¹ over both sampling years. Average TTE was 14% in 2010 and 8% in 2011, indicating that the earlier peak in primary productivity in 2010 resulted in more efficient energy transfer from phytoplankton to crustacean zooplankton compared to 2011. Results from this study highlight the need for incorporating routine field estimates of crustacean productivity into oceanographic studies with the same resolution as primary productivity measurements. Together, these estimates are critical in terms of investigating the impact of a potential increase in the occurrence of mismatches between lower and higher trophic levels in predicted future warming scenarios.

KEY WORDS: Crustacean productivity · Primary productivity · Chitinase · Zooplankton · Trophic transfer efficiency · Saanich Inlet

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INTRODUCTION

Zooplankton provide a key link in marine ecosystems between phytoplankton, at the base of marine food webs, 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 to 24% being observed across most marine ecosystems (Pauly & Christensen 1995). TTE has typi-

cally been calculated on the basis of bulk zooplankton biomass estimates alone, which does not provide information regarding the rate at which the biomass is generated. Furthermore, tropical regions with low overall biomass dominated by small crustaceans with fast growth rates may contribute substantially to overall zooplankton production (Hopcroft & Roff 1998). Temporal mismatches may also 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

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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 and simultaneous estimations 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 remain far less common.

Historically, measurements of secondary productivity have been time-consuming, usually involving incubations of a specific size class of copepods or the development of artificial cohorts (Kimmerer & McKinnon 1987, Peterson et al. 1991). In addition, the egg production method (Kjørboe & Johansen 1986, Poulet et al. 1995) has also been widely accepted due to its feasibility in the field. However, these methods have often been limited to estimating production for only a single (at most a few) copepod species (Poulet et al. 1995). As an alternative, global mathematical models have been used to estimate copepod growth rates (e.g. Huntley & Lopez 1992, Hirst & Lampitt 1998, Hirst & Bunker 2003), thus allowing for broad-scale estimates of copepod production without the need to use more time-consuming traditional field methods. Over the last 20 yr, a method has been developed for obtaining routine estimates of community-level planktonic crustacean productivity involving measurement of the crustacean moulting enzyme chitinase (Oosterhuis et al. 2000, Sastri & Roff 2000, Knotz et al. 2006). Chitinase is one of 2 chitinolytic enzymes responsible for the degradation and reutilization of the exoskeleton during moulting in arthropods (Muzzarelli 1977). Upon moulting, crustaceans release chitinase into the surrounding water, thereby allowing direct estimates of somatic crustacean productivity to be made by measuring the decay rate of chitinase activity. Significant relationships between body length and weight and the rate of production of chitinase activity have already been established (Sastri & Dower 2006) and this analytical method has been validated for *in situ* measurements at sea (Sastri & Dower 2006, 2009).

Whereas growth and productivity in phytoplankton depend mainly on light and nutrient availability, physiological processes in crustacean zooplankton are instead controlled by a combination of other factors such as temperature, oxygen concentration, female body size, food quantity, and food quality. Given that crustacean productivity is a function of biomass and growth rate (Kimmerer 1987, Huntley & Lopez 1992), in addition to egg production (Runge & Roff 2000), changes in the composition of the crusta-

cean community and resulting biomass will undoubtedly impact productivity. Interannual variations in temperature have been shown to influence the species composition of copepod communities (Greve et al. 2004, Mackas et al. 2012). In addition, increasing water temperatures may lead to a mismatch between trophic levels by accelerating the peak timing of naupliar abundance 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 significant 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 rest of the food web (Cushing 1990, Aberle et al. 2012).

Given that the composition and biomass of zooplankton communities are known to exhibit substantial seasonal and interannual variations (Mackas et al. 2001), knowledge about how variations in phytoplankton dynamics influence the crustacean zooplankton community is crucial to understanding the efficiency with which energy is transferred among 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 Strait of Georgia (British Columbia, Canada) (Sastri & Dower 2009). Variability in production rates at critical times (e.g. during the spring) in any given year may have considerable ecological implications throughout the food web in temperate systems. For example, a decline in growth and survival of major juvenile fish species (e.g. Pacific salmon *Oncorhynchus* spp. and Pacific herring *Clupea pallasii*) has recently been linked to low production of their prey during years with unfavourable oceanic conditions in the Strait of Georgia (Beamish et al. 2012, Thomson et al. 2012). Therefore, accurate estimates of crustacean productivity are required to better understand how temporal and spatial variations in this productivity influence fish production, especially in light of the potential impacts of a changing climate on marine ecosystems.

Here, we couple biweekly (every 2 wk) measurements of primary productivity with chitinase-based productivity for the entire crustacean zooplankton community from March to August over 2 yr in a highly productive fjord. Our sampling program spanned the transition from El Niño conditions in early 2010 to La Niña conditions beginning in July 2010 and continuing throughout 2011. Our main goal was to determine

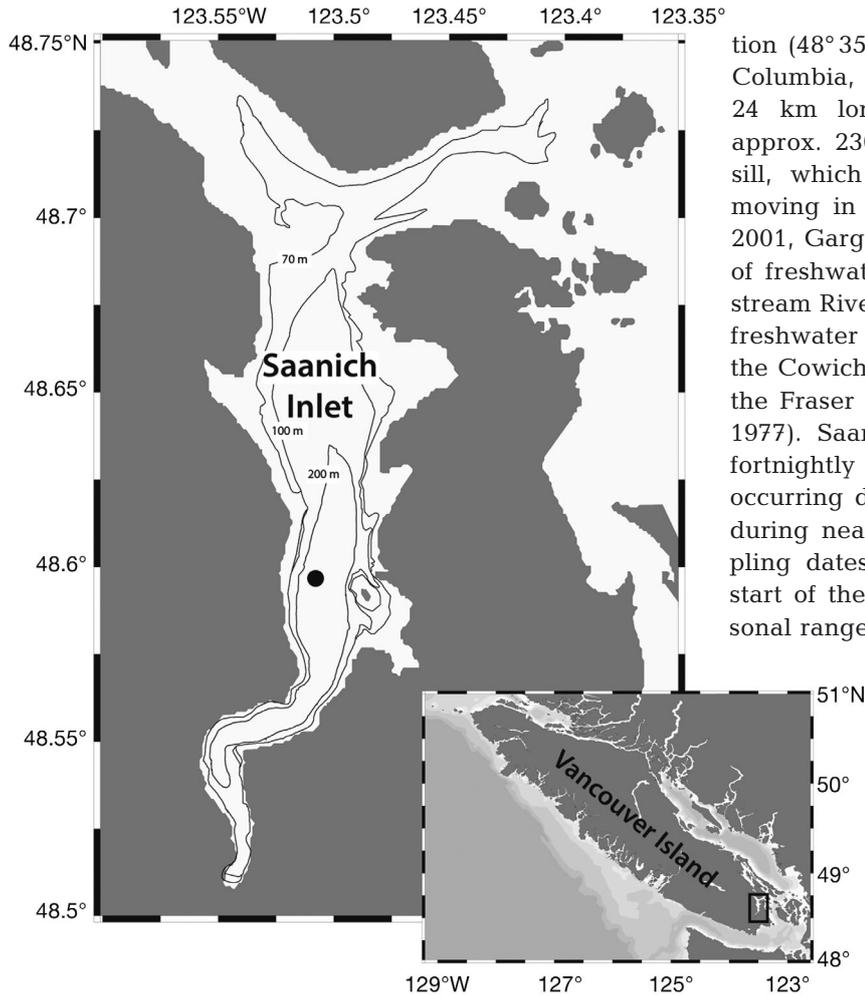


Fig. 1. Location of sampling site (●, 48° 35' N, 123° 30' W) in Saanich Inlet, British Columbia, Canada

how temporal variations in phytoplankton primary productivity influence crustacean productivity and TTE. In addition, we aimed to determine (1) the abiotic and biotic factors that best explained the variation in primary and crustacean productivity, (2) the magnitude of short-term shifts in fatty acid composition for one of the dominant copepod species in the region and whether such diet composition shifts have a significant impact on community-level crustacean productivity, and (3) how temporal variations in phytoplankton and zooplankton productivity influence the transfer of energy between these 2 trophic levels.

MATERIALS AND METHODS

Study site

Sampling took place approx. every 2 wk from March to August in 2010 and 2011 at a single sta-

tion (48° 35' N, 123° 30' W) in Saanich Inlet, British Columbia, Canada (Fig. 1). Saanich Inlet is a 24 km long fjord with a maximum depth of approx. 230 m and a shallow (75 to 80 m deep) sill, which prevents deep water from routinely moving in and out of the inlet (Timothy & Soon 2001, Gargett et al. 2003). Due to the small inputs of freshwater at the head of Saanich Inlet (Goldstream River and Shawnigan Creek), the dominant freshwater sources come from outside the inlet: the Cowichan River during winter and spring, and the Fraser River during summer (Takahashi et al. 1977). Saanich Inlet is strongly influenced by a fortnightly tidal 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 4 d of the start of the spring tide in order to capture a seasonal range of plankton dynamics without the confounding influences of the spring versus neap tides. Seasons were defined as follows: late winter (March), spring (April and May), and summer (June to August).

Physical and chemical measurements

Water temperature, salinity, chlorophyll *a* (chl *a*) fluorescence, and dissolved oxygen were recorded on each sampling date from the surface to approximately 10 m above the bottom (230 m bottom depth) 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 & Jones 1987). A depth of 50 m was chosen to represent the 'functional bottom depth' of the surface layer for our calculations of $\Delta\sigma_t$. Similar results were obtained using other depths (i.e. 20 and 100 m) as most of the variability in Saanich Inlet occurs in the surface layers. Furthermore, the surface mixed layer was almost always within the upper 10 m. 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 (www.climate.weather.gc.ca/) from the meteorological station

at the Victoria International Airport, which is <15 km from our sampling site.

Seawater samples were collected with Niskin bottles at 4 depths corresponding to 100 (surface), 55, 15 and 1% of surface irradiance. Sampling depths were determined from vertical profiles of photosynthetically active radiation (PAR) collected with a Biospherical QSP-200L on the CTD. Samples for dissolved phosphate (PO_4^{3-}), nitrate (NO_3^-) and silicic acid ($\text{Si}(\text{OH})_4$) were filtered through pre-combusted 0.7 μm pore size glass fiber filters into acid-washed 30 ml polypropylene bottles and frozen at 20°C for later analysis. The determination of nutrient concentrations was performed spectrophotometrically using an Astoria II Nutrient Autoanalyzer, following the methods in Barwell-Clarke & Whitney (1996). Nutrient concentrations were trapezoidally integrated from the ocean surface to the bottom of the euphotic zone.

Phytoplankton

Chlorophyll *a*

The concentration of chl *a* was used as a proxy for phytoplankton biomass. Samples of 250 to 500 ml were filtered directly onto 0.7 μm pore size glass fiber filters for the calculation of the chl *a* concentration of the total phytoplankton assemblage. Filters were kept at -20°C until further analysis. Chl *a* was extracted with 90% acetone over 24 h and measured with a Turner Designs 10AU fluorometer as described in Parsons et al. (1984). To calculate phaeopigment interference, the fluorescence was measured again after acidification of the extract with 1.2 N HCl.

Biogenic silica

The concentration of biogenic silica (bSiO_2) was used as a proxy for the biomass of siliceous microphytoplankton (mainly diatoms). Seawater samples (0.75 to 1.5 l) were filtered onto 0.6 μm pore size polycarbonate filters, which were dried at 60°C for at least 48 h, and then stored in a vacuum desiccator at room temperature until further analysis. An alkaline digestion of the material collected on the filters was performed to convert the bSiO_2 into $\text{Si}(\text{OH})_4$ (Brzezinski & Nelson 1989). The concentration of the produced $\text{Si}(\text{OH})_4$ was then measured using a Beckman DU 530 UV/Vis spectrophotometer as described in Brzezinski & Nelson (1986).

Phytoplankton assemblage 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 (Parsons et al. 1984). After gentle homogenization, a 50 ml aliquot was added to an Utermöhl settling chamber and left to settle for 24 h. Abundance of the dominant phytoplankton taxa was then determined using an Olympus inverted microscope (Utermöhl 1958).

Primary productivity

Seawater samples were transferred from the Niskin bottles (from the 4 irradiance depths) into acid-washed 1 l polycarbonate bottles and immediately spiked with both ^{15}N -labelled NaNO_3^- (Cambridge Isotope Laboratories, +98 atom% ^{15}N) and ^{13}C -labelled NaHCO_3 (Cambridge Isotope Laboratories, 99 atom% ^{13}C) with a final enrichment target of ~10% of ambient concentrations. Bottles were placed inside polycarbonate tubes covered with blue and neutral density photographic film to simulate the irradiance and approximate wavelength levels from the sampling depths. Tubes with samples were then placed in a polycarbonate incubator on the ship's deck for 4 h and were held at a constant temperature controlled with flowing surface seawater. The incubation was terminated by filtering the samples onto pre-combusted 0.7 μm pore size glass fiber filters. Filters were then dried at 60°C for ~48 h and stored in a desiccator until further processing.

Isotopic ratios (i.e. $^{14}\text{N}:^{15}\text{N}$ and $^{12}\text{C}:^{13}\text{C}$) and the C and N content of the particulate matter collected on the filters after the incubation period were measured using a PDZ Europa 20-20 isotope mass spectrometer and a PDZ Europa ANCA-GSL elemental analyzer at the Stable Isotope Facility at the University of California Davis, USA. C and NO_3^- uptake rates were calculated following Hama et al. (1983) and Dugdale & Wilkerson (1986), respectively. Depth-integrated uptake rates were trapezoidally integrated from the ocean surface to the bottom of the euphotic zone. New primary production in C units was calculated by multiplying NO_3^- uptake rates by the measured C:N ratio of the particulate matter to determine the relative importance of NO_3^- based primary productivity to total productivity.

Zooplankton

Community composition

Micro- and mesozooplankton samples 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. Zooplankton samples were split (1/64) in the laboratory 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, British Columbia, and Fulton (1968). Copepods were identified to species level when possible while all other zooplankton were identified to major taxonomic group. Prosome lengths of adult copepods were measured at 25 \times magnification using a dissecting microscope with an ocular micrometer. Abundance data (ind. m^{-3}) were converted to biomass (mg m^{-3}) using species-specific length–dry weight relationships from an internal database provided by IOS, and a dry weight to carbon conversion factor of 0.45 (Paffenhöfer & Harris 1976). A full description of the structure and content of the IOS database can be found in Mackas et al. (2013).

Fatty acid analysis

Variations in the dietary fatty acid composition of *Calanus marshallae* (one of the dominant, large copepod species in Saanich Inlet) were examined on each sampling date. For each sample, 10 *C. marshallae* at the fifth copepodite stage (CV) were sorted on board and stored in Cryovials[®] and kept on dry ice until transferred to the lab to be stored at -80°C until analysis. Fatty acid analyses followed the protocols 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 acid extractions were performed following Parrish (1999) and Kainz et al. (2004). Samples were sonicated and vortexed 3 times in a 4:2:1 chloroform:methanol:water mixture. The organic layers were removed, pooled, and samples were then capped off with N_2 to prevent degradation. Fatty acids were analyzed as methyl esters (FAME) formed using

hexane and $\text{BF}_3\text{-CH}_3\text{OH}$ at 85°C for 1 h (Kainz et al. 2004). Esterified fatty acids were analyzed using a gas chromatograph (GC; Varian CP-3800) 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 expressed as percentages of total fatty acids. Given that dinoflagellates have a high proportion of docosahexaenoic acid (22:6n-3, DHA) while diatoms have a high proportion of 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 zooplankton diet (Viso & Marty 1993, Dalsgaard et al. 2003).

Crustacean productivity

Planktonic crustacean productivity was measured via the chitobiase enzyme method as described by Sastri & Dower (2006). Water samples (500 ml) for chitobiase incubations were collected from Niskin bottles at 6 depths (5, 15, 25, 50, 75, and 100 m) and screened with a 40 μm mesh in order to remove all crustaceans and other organisms. The process involved (1) measurement of the native *in situ* chitobiase activity (CBA_{nat}) and (2) measurement of chitobiase activity (CBA) over a 24 h period using different aliquots obtained from the same original sample. For (1), approximately 15 ml of the original seawater sample from each depth was immediately filtered (0.2 μm pore size) in order to remove any bacteria and subsequently used to estimate CBA_{nat} . For (2), a crude homogenate of 20 to 30 medium-sized copepods (freshly ground in 3 ml of seawater and passed through a 0.2 μm filter) was used to enhance ('spike') the chitobiase signal of the original samples from each depth. This served to increase the baseline chitobiase level and better differentiate the decay of CBA from background fluorescence (see Sastri & Dower 2006). Spiked seawater samples were sampled just after the homogenate was added ($t = 0$), and then every 3 to 6 h thereafter over the 24 h period. Samples were maintained at ambient surface seawater temperature throughout. Incubations were conducted back on land in the laboratory and the resulting samples were 0.2 μm filtered, and stored at 4°C in glass tubes until assayed in the laboratory. All enzyme assays were conducted within 30 h of sampling.

Enzyme assays to measure CBA were initiated by adding the substrate 4-methylumbelliferyl- β -D-glucosaminide (0.1 mmol MBF-NAG; Sigma) to the

seawater samples. Assays were conducted at 25°C and terminated after 1 h 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 methylumbelliferone (MBF) fluorescence over the 1 h reaction period (Sastri & Dower 2006). The reaction was buffered to pH 6.0 (optimal for copepods; Knotz et al. 2006, Sastri & Dower 2006) 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 filter (365 nm excitation and 450 nm emission). Raw fluorescence values were converted to nmol MBF using a standard curve of known 4-methylumbelliferone concentrations against fluorescence.

The chitobiase method assumes that the planktonic crustacean community is in steady state for the duration of the 24 h incubation and that chitobiase production is therefore balanced by its rate of decay (Sastri & Dower 2009). Estimates of CBA decay rate (h⁻¹) were calculated as the slope (k) of the natural logarithm of CBA versus time (Sastri & Dower 2006). A Q_{10} correction was applied to the slope in order to account for differences between *in situ* temperature and 24 h incubation temperature (Sastri & 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 CBA_{nat} . This equation is based on the assumption that T_{CBA} represents the proportion of animals moulting per hour (Sastri & Dower 2006). 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 ($\log(g_{\text{inc}}) = 0.864 \log(\text{CBA}) - 1.78$) (Sastri & Dower 2006) to the average CBA_{nat} , the equivalent to the average sum of all individual growth, at each depth. Depth-integrated daily planktonic crustacean productivity (mg C m⁻² d⁻¹) was then calculated as the biomass production divided by stage duration, or $\Delta B/T_{\text{CBA}}$. The ratio of daily productivity to the developing biomass (P/B) was estimated from our corrected values of CBA_{nat} and was used as an equivalent to daily growth rate (g , d⁻¹) (Sastri et al. 2012).

Trophic transfer efficiency

Due to the time lags in the relative metabolic rates of phytoplankton and zooplankton, changes in primary productivity are not expected to be immedi-

ately reflected in crustacean productivity. To account for this time lag in energy transfer and for the different timescales required for both processes, we calculated a moving average of productivity values over 2 sampling dates (approx. 4 wk). Time-averaging over a 1 mo period reduced the occurrence of sampling artefacts (e.g. unreasonably high values of TTE) that can result from comparing productivity estimates incorporated over short timescales (Gladyshev et al. 2011). TTE was then calculated as the percent of the time-averaged crustacean production rate divided by time-averaged primary productivity rate. Artefact values of TTE > 35% were excluded from calculations of average TTE (details of TTE calculations can be found in Tables S1 & S2 in the Supplement at www.int-res.com/articles/suppl/m545p091_supp.pdf).

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). Where appropriate, explanatory variables were transformed 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-R² 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-based and plankton net based values, and between our productivity estimates and TTE. All analyses were performed using Sigmaplot® v.12.3 and R v.3.0.2 (R Development Core Team 2013).

RESULTS

Water column characteristics

Differences in local environmental conditions between the 2 sampling years (warmer and drier during El Niño, colder and wetter during La Niña) resulted in slight differences in water column characteristics. Overall, average water column temperatures (ocean surface to 100 m) were somewhat higher in 2010 than in 2011, although both years showed a typical increase in temperature throughout the season, with

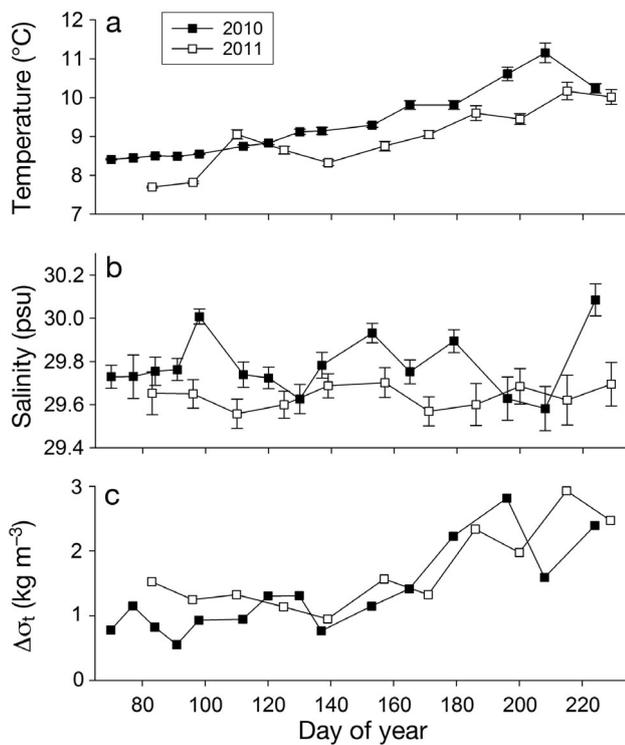


Fig. 2. Mean \pm SE water column (a) temperature, (b) salinity, and (c) Change in density ($\Delta\sigma_t$) from March to August 2010 and 2011 in Saanich Inlet. $\Delta\sigma_t$: difference in density between the surface (average from top 10 m) and 50 m

a maximum occurring near the beginning of August (Days 208 and 215 in 2010 and 2011, respectively; Fig. 2a). Water column salinity was similar throughout both years (Fig. 2b). However, the water column was slightly more stratified in early spring 2011 (Days 83 through 115) as indicated by higher values of the $\Delta\sigma_t$ stratification parameter (values of 0.6 to 2.8 and 0.9 to 2.9 kg m^{-3} for 2010 and 2011, respectively) (Fig. 2c). The depth of the euphotic zone (1% light level) varied between 10 and 20 m throughout the 2010 and 2011 sampling seasons.

Nutrient concentrations

During both years, there was a decrease of all nutrient concentrations after Day 100 (early April) (Fig. 3). Values of NO_3^- and Si(OH)_4 were 24.4 and 44.4 $\mu\text{mol l}^{-1}$, respectively, at the beginning of our sampling program in 2010 (Fig. 3a,c). On our first sampling date in 2011 (end of March; Day 83), concentrations of NO_3^- and Si(OH)_4 were about half of those measured at the end of March (Day 84) in 2010 (Fig. 3a,c). By comparison, PO_4^{3-} was only slightly lower on our first sampling date in 2011 (Fig. 3b).

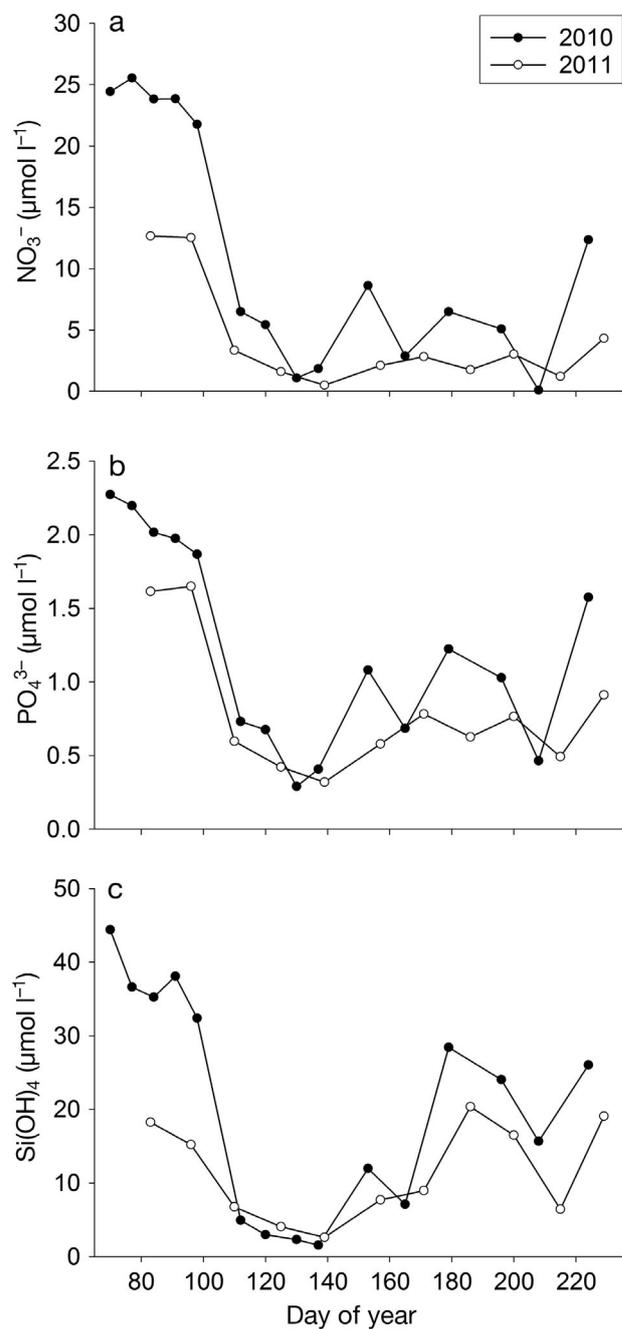


Fig. 3. Concentrations of dissolved (a) nitrate, (b) phosphate, and (c) silicic acid from March to August 2010 and 2011 in Saanich Inlet

Phytoplankton

Assemblage composition

In both years flagellates/coccoid cells ($<5 \mu\text{m}$) and dinoflagellates dominated the community in late winter (before Day 85) and summer (after Day 150),

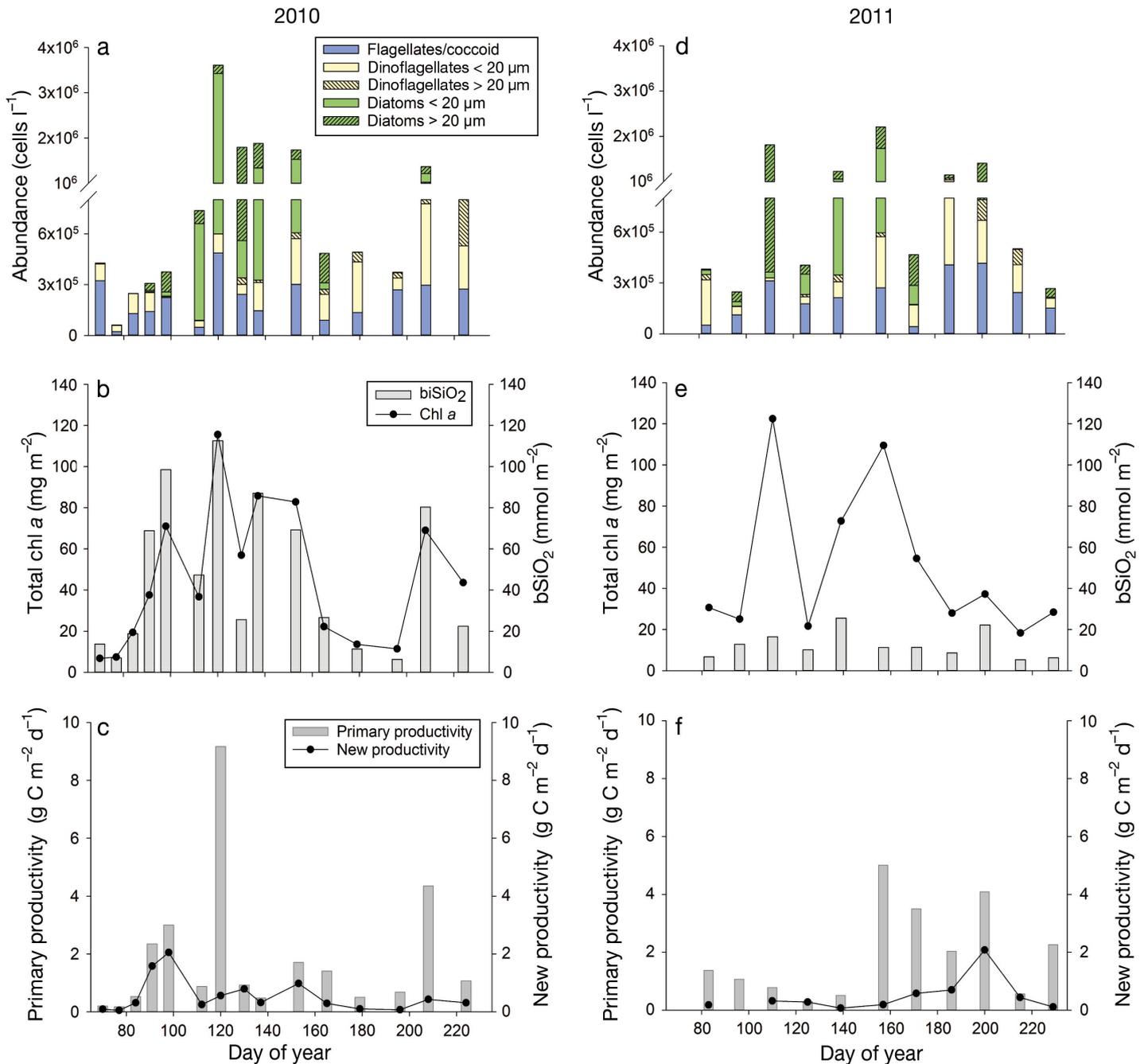


Fig. 4. (a,d) Abundance of major taxonomic groups comprising the phytoplankton assemblage, (b,e) depth-integrated total chl a and biogenic silica (bSiO₂), and (c,f) depth-integrated primary productivity and new primary productivity throughout the euphotic zone from March to August 2010 and 2011 in Saanich Inlet. Note: no data available for new productivity on the second sampling date in 2011

whereas diatoms were the most abundant phytoplankton group during spring (Days 90–140) (Fig. 4a,d). In late spring 2010 (Days 112–153), diatoms <20 μm were more abundant than diatoms >20 μm (with the exception of Day 130, May 10; Fig. 4a). In 2011, the highest abundance of large

diatoms occurred a few weeks earlier on Day 110 (April 20) (Fig. 4d). The most abundant diatom genera were *Chaetoceros*, *Thalassiosira*, *Cylindrotheca*, *Pseudonitzschia*, and *Skeletonema*, while the most common dinoflagellate genera were *Gymnodinium*, *Prorocentrum*, and *Protoperidinium*.

Biomass as chl *a* and bSiO₂

The depth of the chl *a* maximum was typically between 5 and 10 m throughout the sampling period for both years (data not shown). The first peak in total depth-integrated chl *a* occurred on Day 98 (April 8) in 2010 and Day 110 (April 20) in 2011 (Fig. 4b,e), corresponding to an increase in salinity and temperature (Fig. 2) in 2010 and 2011, respectively. During both years, this initial peak in chl *a* was followed by a substantial decrease 2 wk later. Large phytoplankton (>20 µm) were the dominant size fraction in Saanich Inlet at all times, representing a larger proportion of the total chl *a* biomass in the spring of 2011 than in the spring of 2010 (data not shown). Depth-integrated bSiO₂ was substantially higher in 2010 compared to 2011 (Fig. 4b,e), with mean values of 46.36 ± 9.40 mmol m⁻² in 2010 and 12.40 ± 1.97 mmol m⁻² in 2011.

Primary productivity

Primary productivity peaked on Day 120 (late April; 9.17 g C m⁻² d⁻¹) in 2010 (Fig. 4c). In 2011, however, the peak was not observed until Day 160 (mid-June; 5.01 g C m⁻² d⁻¹, Fig. 4f). Mean primary productivity over the sampling period was quite similar for both years with values of 1.83 ± 0.60 g C m⁻² d⁻¹ in 2010 and 1.95 ± 0.48 g C m⁻² d⁻¹ in 2011. New primary productivity values were also similar between years ranging between 0.05 and 2.05 g C m⁻² d⁻¹ in 2010 and between 0.07 and 2.08 g C m⁻² d⁻¹ in 2011 (Fig. 4c,f).

Zooplankton

Abundance

Abundances of the copepod genera *Calanus* and *Corycaeus* were substantially higher in 2010 than in 2011 (Fig. 5a,b). Mean abundance of *Calanus* was 308 and 90 ind. m⁻³ in 2010 and 2011, respectively, whereas mean abundance of *Corycaeus* was 718 and 228 ind. m⁻³ in 2010 and 2011, respectively. In contrast, the abundance of euphausiids was relatively higher between Days 120 and 140 in 2011 (Fig. 5a,b). The abundances of other numerically dominant crustaceans (copepods *Oithona* and *Metridia*) were similar for both years. The initial peak in copepod abundance occurred on about Day 110 in 2010 compared to the first peak in 2011, which did not happen

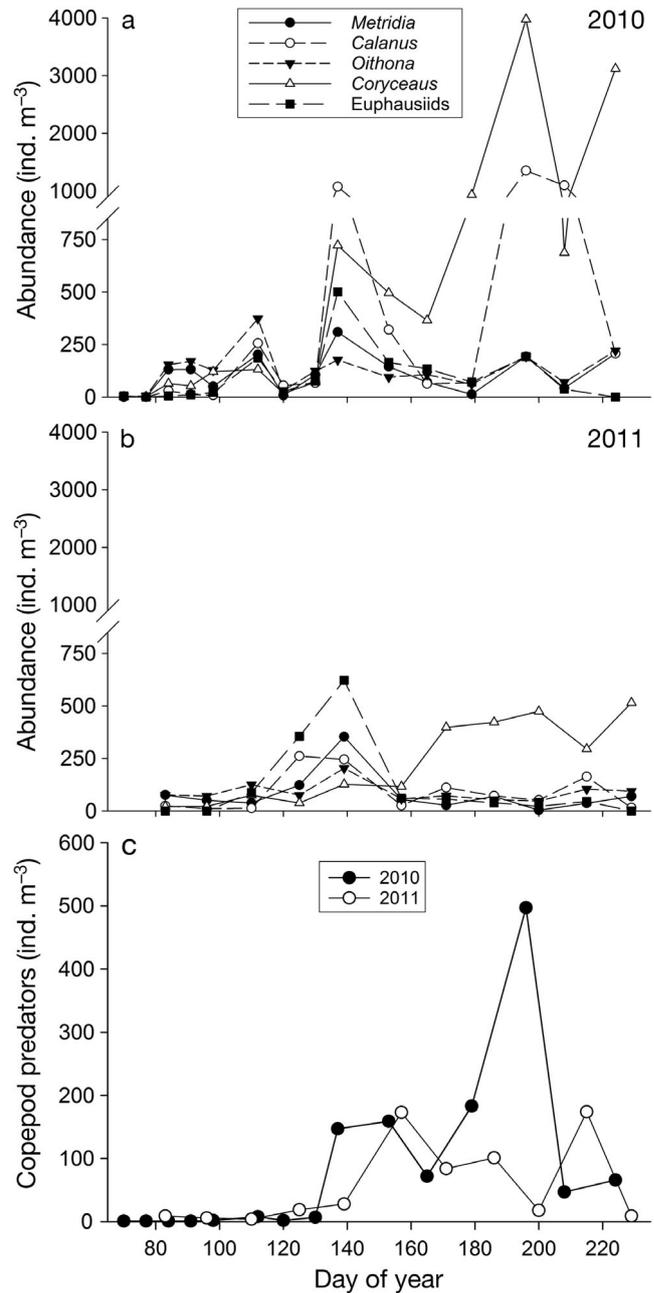


Fig. 5. Abundance of dominant crustaceans in (a) 2010 and (b) 2011, and (c) copepod predators (chaetognaths, cnidarians, and other jellies including siphonophores, ctenophores, and salps) in 2010 and 2011 in Saanich Inlet. All animals were collected from 0–100 m depth vertical net hauls

until after Day 140 (Fig. 5a,b). *Calanus* and, to a lesser extent, *Metridia*, peaked multiple times in 2010 compared to a single peak in 2011. Copepods <2 mm in length (e.g. *Oithona*) had a lower, albeit steady, abundance throughout our study. In terms of seasonal variation, *Calanus*, *Metridia*, and euphausiids were more abundant in spring, whereas *Corycaeus*

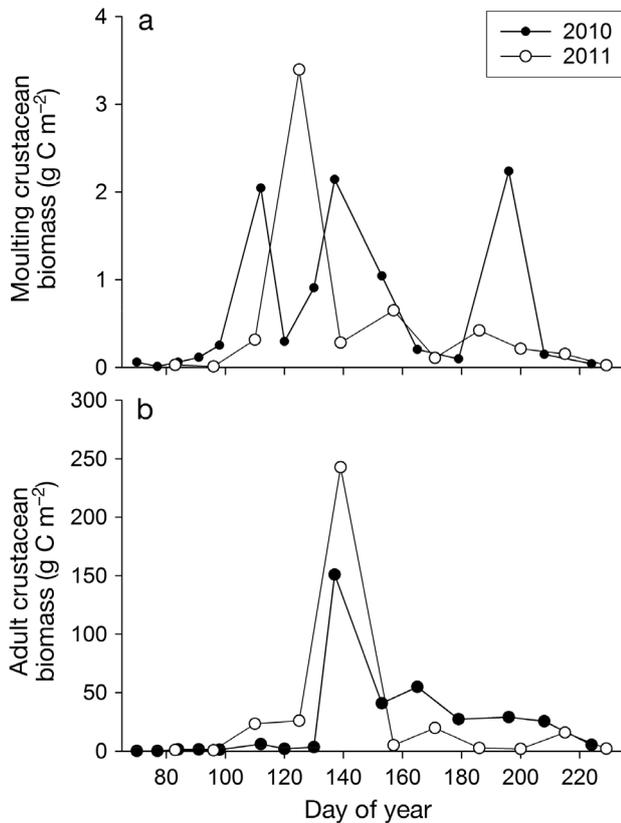


Fig. 6. Depth-integrated biomass of (a) moulting crustaceans (larvae and juveniles), and (b) adult (i.e. non-moulting) crustaceans from March to August in 2010 and 2011 in Saanich Inlet

was the most abundant crustacean in the summer of both years. The abundance of copepod predators such as chaetognaths, cnidarians, and other jellies (siphonophores, ctenophores, salps) showed a single, high peak in summer 2010 compared to the 2 smaller peaks in 2011 (late spring and summer) (Fig. 5c).

Biomass

Overall, zooplankton biomass in Saanich Inlet was dominated by euphausiids, cnidarians (and other jellies), chaetognaths, larvaceans, and the copepods *Calanus*, and *Metridia* (data not shown). Biomass of moulting crustaceans (larvae and juveniles) followed similar patterns to crustacean abundance, with multiple peaks in biomass occurring in 2010 compared to a single peak in 2011 (Fig. 6a). Although the initial peak in biomass of moulting crustaceans occurred slightly earlier in 2010, the highest adult crustacean biomass occurred at the same time during both years (on Day 140, mid-May; Fig. 6b).

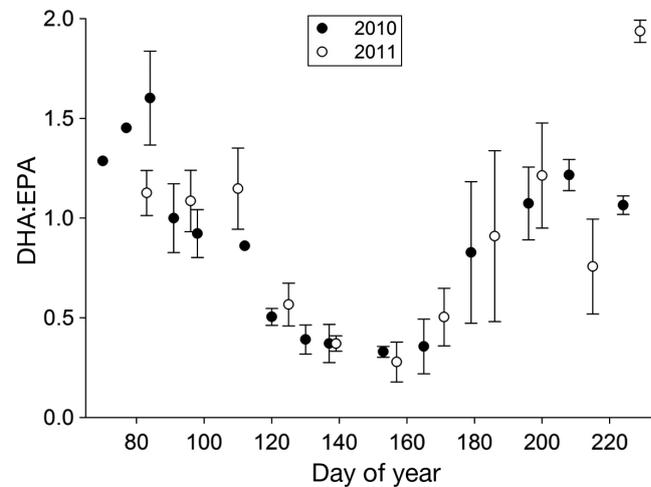


Fig. 7. Seasonal variability (mean \pm SE) in the DHA:EPA ratio of *Calanus marshallae* from March to August in 2010 and 2011 in Saanich Inlet

Fatty acid content

In general, the DHA:EPA ratios in *Calanus marshallae* showed a similar pattern in both years—with a higher ratio of DHA:EPA occurring in late winter/early spring and late summer (Fig. 7). In contrast, the lowest DHA:EPA ratios were observed during the late spring/early summer (between Days 120 and 170). A higher DHA:EPA ratio on Day 80 in 2010 compared to 2011 implies that *C. marshallae* was feeding on a higher proportion of dinoflagellates in spring 2010 (Fig. 7). 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 marginally significant negative correlation was found for DHA:EPA and *Calanus* biomass ($r = -0.36$, $p = 0.07$). It should be noted, however, that the power of the latter correlation was quite low (0.44) and it is thus possible that we failed to detect a significant relationship between DHA:EPA and *Calanus* biomass when one actually existed.

Crustacean productivity

The highest estimates of CBA_{nat} were recorded in April and June of 2010, and in May and August of 2011 (Table 1). The longest mean stage durations (T_{CBA}) occurred in May and June 2010, and in March 2011. The shortest stage duration of 4.6 d in 2010 occurred in late March (Day 84), whereas the shortest stage duration in 2011 occurred in May (Days 125 and 139). Daily P/B was higher in spring 2010 (March

Table 1. Estimates of native *in situ* chitobiase activity (CBA_{nat}), time (stage duration) taken for moulting individuals to produce CBA (T_{CBA}), and ratio of daily productivity to developing biomass (Daily P/B) from March to August in 2010 and 2011 in Saanich Inlet, British Columbia, Canada

Sampling date	Day of year	CBA_{nat} ($nmol\ l^{-1}\ h^{-1}$)	T_{CBA} (d)	Daily P/B
2010				
11 Mar	70	4.63	29.5	0.07
18 Mar	77	2.33	48.4	0.03
25 Mar	84	3.6	4.6	0.19
01 Apr	91	1.91	15.3	0.12
08 Apr	98	2.71	11.8	0.09
22 Apr	112	13.65	8.2	0.17
30 Apr	120	5.79	44.8	0.33
10 May	130	3.87	98.3	0.01
17 May	137	3.37	13.4	0.04
02 Jun	153	3.76	6.2	0.42
14 Jun	165	8.51	95.7	0.04
28 Jun	179	4.92	5.9	0.17
15 Jul	196	4.01	12.1	0.08
27 Jul	208	4.13	30.9	0.03
12 Aug	224	4.92	7.7	0.13
2011				
24 Mar	83	3.4	49.8	0.04
06 Apr	96	4.63	23.8	0.05
20 Apr	110	4.86	13.8	0.10
05 May	125	8.09	4.5	0.22
19 May	139	5.55	6.6	0.15
06 Jun	157	3.43	38.7	0.05
20 Jun	171	5.01	29.3	0.04
05 Jul	186	5.08	17.1	0.07
19 Jul	200	5.37	17.6	0.04
03 Aug	215	6.09	20.5	0.06
17 Aug	229	2.77	8.0	0.13

and April) than in 2011. Ranges of daily P/B varied from 0.01 to 0.42 in 2010 and from 0.04 to 0.22 in 2011 (Table 1). In general, estimates of crustacean productivity were higher in 2010 than in 2011 (Fig. 8). One notable exception is the extremely low productivity measured in May 2010 (Days 130 and 137; Fig. 8a) compared to the high productivity values in May 2011 (Fig. 8b). In 2011, production rates did not decrease until June (Day 157; Fig. 8b).

Linking primary productivity and crustacean productivity

In 2010, primary productivity was positively related to chl *a* ($adj\ R^2 = 0.60$, $p = 0.001$), whereas in 2011, primary productivity was positively related to chl *a* ($p < 0.05$) and negatively related to crustacean abundance ($p < 0.05$) (Table 2). Crustacean productivity in 2010 was best predicted by a linear com-

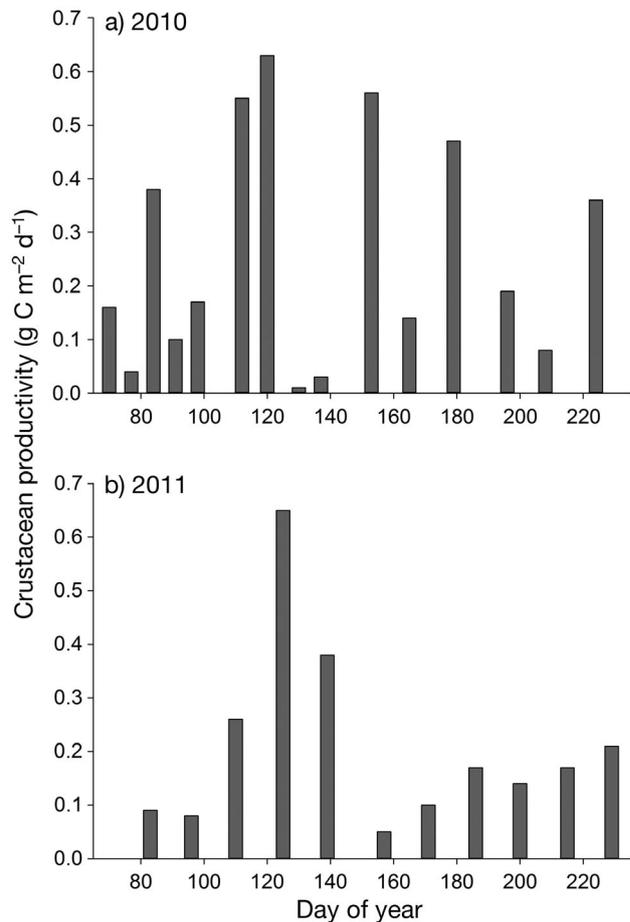


Fig. 8. Depth-integrated crustacean productivity from the surface to 100 m depth from March to August in (a) 2010 and (b) 2011 in Saanich Inlet

bin of salinity (significant, $p < 0.05$), chl *a* (non-significant, $p = 0.097$), and primary productivity (non-significant, $p = 0.064$) in 2010 ($adj\ R^2 = 0.259$) (Table 2). In 2011, the variables best predicting crustacean productivity were crustacean biomass, copepod predator abundance, temperature, and primary productivity ($adj\ R^2 = 0.858$). The range of TTEs was slightly higher in 2010 (from 3 to 32%) compared to 2011 (2 to 19%, Table 3). Mean time-averaged TTE across the sampling season was 14 and 8% for 2010 and 2011, respectively (Table 3).

DISCUSSION

Timing of spring bloom

Due to the transition from El Niño to La Niña conditions over our sampling program, slightly warmer waters occurred in 2010 compared to 2011 in Saanich

Table 2. 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 from March to August in 2010 and 2011 in Saanich Inlet. Significant values are indicated in **bold**

	Coefficient	SE	p-value
2010			
Primary productivity (N = 15, R ² = 0.711, Adj R ² = 0.596)			
Constant	30.575	22.115	
NO ₃ ⁻	0.002	0.001	0.197
1/Temp	-20.041	11.705	0.118
Salinity	-0.977	0.73	0.195
Log ₁₀ Chl <i>a</i>	1.216	0.277	0.001
Crustacean productivity (N = 15, R ² = 0.418, Adj R ² = 0.259)			
Constant	-57.954	26.252	
Salinity	1.964	0.887	0.049
Log ₁₀ Chl <i>a</i>	-0.898	0.495	0.097
Log ₁₀ Primary productivity	0.849	0.412	0.064
2011			
Primary productivity (N = 11, R ² = 0.789, Adj R ² = 0.648)			
Constant	3.622	1.965	
NO ₃ ⁻	-0.005	0.002	0.051
Si(OH) ₄	0.002	0.001	0.081
Log ₁₀ Crustacean abundance	-1.572	0.584	0.036
Log ₁₀ Chl <i>a</i>	0.874	0.291	0.024
Crustacean productivity (N = 11, R ² = 0.915, Adj R ² = 0.858)			
Constant	0.187	0.553	
Log ₁₀ Crustacean biomass	0.164	0.062	0.038
Predator abundance	-0.003	0.001	0.009
1/Temp	-12.709	3.959	0.018
Log ₁₀ Primary productivity	-0.369	0.123	0.024

Inlet and in the nearby Strait of Georgia (Irvine & Crawford 2012). La Niña conditions (colder, more precipitation) during the winter of 2010–2011 resulted in increased stratification in early spring 2011, which may have prevented high concentrations of dissolved nutrients from being replenished into the upper water column via vertical mixing, as was observed during the previous year. However, despite the marked 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 previously reported concentrations for Saanich Inlet surface waters (Grundle et al. 2009).

Although nutrient drawdown occurred at the same time during both years, the peak in chl *a* biomass was delayed by approximately 2 wk in 2011. Previous studies have found that chl *a* biomass peaks in Saanich Inlet in both spring (April, Grundle et al. 2009; mid-May, Takahashi et al. 1977) and summer (July and August, Grundle et al. 2009). The maxi-

mum chl *a* concentrations in the present study occurred in April and during the summer months (June and July) in both years, in agreement with these previous studies. Satellite images and ship-based surveys in the Strait of Georgia determined that the 2011 spring bloom was delayed and was more intense than the spring bloom in 2010, due primarily to strong wind events at the end of March/beginning of April in 2011 (Irvine & Crawford 2012). In contrast, we found that the magnitude of phytoplankton biomass in Saanich Inlet was similar in both years. This discrepancy in results from 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 & Wolfe 2013), whereas winds play a relatively weak role in Saanich Inlet due to its north/south axis, which is orthogonal to the dominant wind direction (Gargett et al. 2003).

Phytoplankton assemblage and primary productivity

The seasonal succession of major phytoplankton taxa was similar for both years, with the exception of diatoms. Diatoms were more abundant during the early spring of 2010 compared to 2011, which was also shown by high concentrations of bSiO₂ over the entire growing season in 2010. The higher abundance of diatoms was most likely a result of higher nutrient concentrations, particularly Si(OH)₄, in the upper water column prior to the onset of the spring bloom in 2010. That said, the diatoms in our early spring 2010 samples were comprised of mainly cells in the 5–20 μm size range, in contrast to the greater proportion of >20 μm diatoms observed in 2011. 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% of total phytoplankton abundance) in 2010, the highest relative abundance of diatoms was not observed until June in 2011 (68%). These trends were also reflected in the 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 rather than dinoflagellates (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 pro-

Table 3. Calculations of trophic transfer efficiency (TTE) 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)	TTE (%)	TTE artefact samples removed (%)
2010				
74	0.18	0.10	53	–
81	0.35	0.21	60	–
88	1.43	0.24	17	17
95	2.67	0.13	5	5
105	1.94	0.36	19	19
116	5.02	0.59	12	12
125	5.04	0.32	6	6
134	0.70	0.02	3	3
145	1.09	0.29	27	27
159	1.56	0.35	22	22
172	0.96	0.31	32	32
188	0.59	0.33	56	–
202	2.51	0.13	5	5
216	2.71	0.22	8	8
			Average	14
2011				
90	2.44	0.17	7	7
103	1.85	0.34	19	19
118	1.08	0.91	84	–
132	0.81	1.03	128	–
148	5.52	0.43	8	8
164	8.51	0.15	2	2
179	5.53	0.28	5	5
193	6.12	0.31	5	5
208	4.65	0.31	7	7
222	2.82	0.38	13	13
			Average	8

ductivity, which was comparable to that observed during previous El Niño conditions in Saanich Inlet (Timothy & Soon 2001). The >20 µm chl *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 significant correlations between >20 µm chl *a* and productivity were observed in 2011. In 2011, the highest primary productivity values were observed during the summer (July–August), in agreement with observations by Grundle et al. (2009). Despite these differences, average primary productivity in Saanich Inlet 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 average primary productivity of 1.6 g C m⁻² d⁻¹ observed by Timothy & Soon (2001) and the 1.6 to 1.9 g C m⁻² d⁻¹ average productivity observed at various sampling stations in Saanich Inlet by Grundle et al. (2009).

Zooplankton community and crustacean productivity

The most notable difference in the zooplankton community between years was the earlier initial peak in 2010 and substantially higher abundance of the larger calanoid copepods (e.g. *Calanus*, *Metridia*) in 2010 than in 2011. Zooplankton phenology is strongly correlated with early spring temperatures (Edwards & Richardson 2004, Greve et al. 2004, Mackas et al. 2007, 2012). For example, the development of the dominant Northeast Pacific copepod *Neocalanus plumchrus* shifted by approximately 1 mo in response to only a 1°C warming (Mackas et al. 2007). It is therefore possible that the higher average water column temperatures in spring 2010 may have triggered earlier developmental cues for crustacean zooplankton. In addition, while all of our daily *P/B* estimates (assumed to be equivalent to daily growth rate) for the planktonic crustacean community fit well with previous estimates for the region (Peterson et al. 2002, Sastri & 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 1980, Kimmerer & McKinnon 1987, Uye 1988), it is unlikely that the slightly higher temperatures experienced by copepods in early spring of 2010 contributed to the faster growth rates that we observed. A more plausible explanation for the higher values of daily *P/B* in 2010 is a higher abundance of nauplii and early-stage copepodites, which have faster growth rates than late-stage copepods and comparatively low biomass per individual (Peterson et al. 1991).

The resulting biomass of the copepod community in 2010, alone, was also much higher than previous estimates for other parts of the region (Sastri & Dower 2009, Tommasi et al. 2013a) due to the high abundance of larger-bodied *Calanus* observed in this study. However, overall crustacean biomass was even higher in 2011 due to 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, probably due to the fact that variations in seasonal biomass are closely linked to the life cycles of the dominant crustaceans (*Calanus* and euphausiids) (Mackas & Tsuda 1999).

It is important to note that the low crustacean productivity observed on any given day in this study did not necessarily correspond to low crustacean bio-

Table 4. Comparison of chitobiase-based planktonic crustacean productivity estimates with those based on more traditional methods. n/a: data not available

Region	Method	Productivity		Source
		Range	Mean	
Saanich Inlet	Chitobiase	0.05–15.61 mg C m ⁻³ d ⁻¹	4.71 mg C m ⁻³ d ⁻¹	Present study
		or 0.01–0.65 g C m ⁻² d ⁻¹	or 0.24 g C m ⁻² d ⁻¹	
Skagerrak, between Norway and Denmark	Artificial cohort	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	Kjørboe & Nielsen (1994)
Sagami Bay, Japan	Hirst & Lampitt (1998) model	0.09–7.77 mg C m ⁻³ d ⁻¹	0.94 mg C m ⁻³ d ⁻¹	Ara & Hiromi (2007)

mass. For example, in 2010 the lowest productivity values (in May) were associated with high crustacean biomass, particularly of large, non-moulting adults. Therefore, periods of lowest productivity could have simply been a result of the low growth rates (daily *P/B* between 0.01 and 0.04) of the non-moulting adults dominating the biomass. In addition to the low growth rates, the lowest productivity in 2010 also coincided with the highest abundances of >20 µm diatoms. We suspect a link between the observed low productivity and what the copepods were feeding on at the time, in agreement with a previous laboratory study that showed a link between low to 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 & Dower 2009). Nutrient limitation in diatoms may also cause a decrease in growth rate of copepods and halt copepod development (Klein Breteler et al. 2005), which could have negatively influenced crustacean productivity in the Strait of Georgia (Sastri & Dower 2009). Interestingly, our observations of low crustacean productivity 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. Given that consumption of diatoms with faster growth rates has been shown to result in increased reproductive success of copepods (Jónasdóttir & Kjørboe 1996), it is possible that productivity in Saanich Inlet was negatively affected when crustaceans were feeding on phytoplankton with slower 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 chaetognaths and cnidarians.

The lack of a consistent method for estimating crustacean productivity makes it difficult to meaningfully compare these results to previous studies. The range of crustacean productivity observed in this study is similar to estimates calculated for the Strait of Georgia (Sastri & Dower 2009). However, the field estimates of chitobiase-based productivity obtained in this study were consistently higher than previously reported estimates for copepod communities. For example, chitobiase-based crustacean productivity in Saanich Inlet 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 4). In comparison, Peterson et al. (1991) found that the range of community-level productivity obtained using artificial cohort incubation methods in the Skagerrak region between Norway and Denmark was slightly lower (3.0 to –8.0 mg C m⁻³ d⁻¹) than productivity in Saanich Inlet. Maximum depth-integrated copepod production 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) (Kjørboe & Nielsen 1994). Using yet another method, Ara & Hiromi (2007) estimated production based on the Hirst & 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.

The discrepancy between these studies likely stems from the fact that the chitobiase method captures 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 (as done in our study) may result in under-sampling and even damaging of

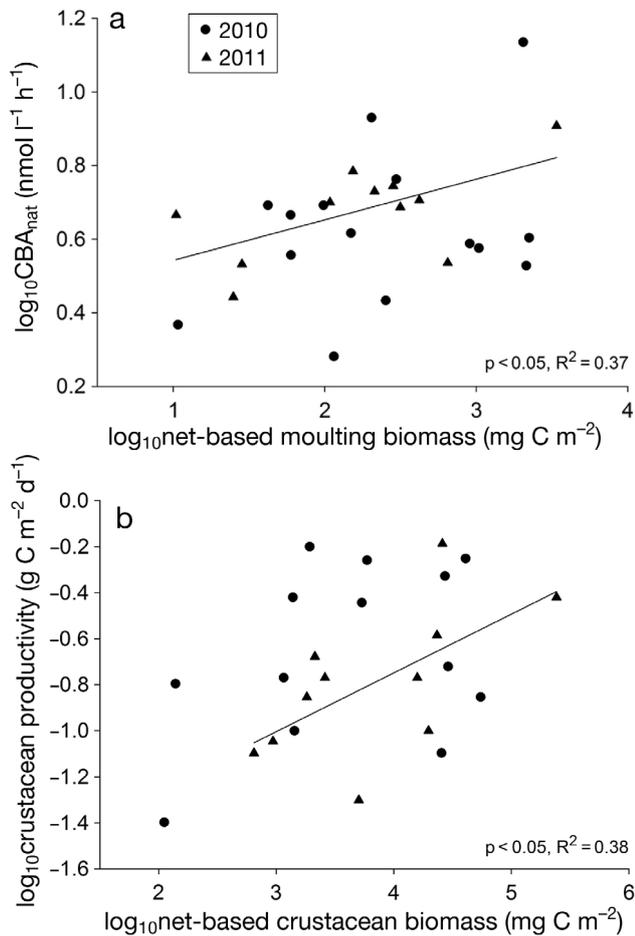


Fig. 9. Relationships between (a) native *in situ* chitobiase activity (CBA_{nat}) and net-based moulting biomass and (b) crustacean productivity and net-based adult crustacean biomass from March to August 2010 and 2011 in Saanich Inlet. Solid lines are linear regressions

small zooplankton, which can contribute substantially to overall production given their fast growth rates (Banse 1982, Arendt et al. 2013). Furthermore, although the majority of the zooplankton community in Saanich Inlet resides at around 100 m (Devol 1981), sampling during only daylight hours may have slightly underestimated the contribution to total abundance and biomass of vertically migrating species (e.g. *Metridia* sp.) or individuals with the ability to visually detect nets (e.g. euphausiids). Regardless, planktonic crustacean biomass has been shown to vary significantly with CBA_{nat} (Sastri & Dower 2009). We also found statistically significant correlations between $\log_{10}CBA_{nat}$ and \log_{10} net-based biomass of moulting crustaceans (Fig. 9a; slope = 0.11, $R^2 = 0.37$, $p < 0.05$), and between \log_{10} crustacean productivity and \log_{10} net-based biomass of adult crustaceans (Fig. 9b; slope = 0.26, $R^2 = 0.38$, $p < 0.05$). Given that

the majority of copepod production estimates in the literature are based on egg production rather than somatic production, future studies could benefit from combining measurements of both community-level somatic crustacean productivity using the chitobiase method and productivity based on female egg production in order to assess the biases associated with using these different methods.

Trophic transfer efficiency

TTE calculations using averages of primary and crustacean productivity over 4 wk showed that energy transfer between phytoplankton and zooplankton in Saanich Inlet was 14 and 8% over the whole sampling period in 2010 and 2011, respectively. These values fall well within the range of 2 to 24% predicted for most marine ecosystems (Pauly & Christensen 1995), but were substantially lower than the 23% TTE calculated for the nearby Strait of Georgia by Sastri & Dower (2009). While the assumption of a 10 to 20% TTE is useful on an annual basis, our results showed substantial seasonal variation from early spring to summer. For instance, TTE varied by a factor of 11 (3 to 32%) in 2010, and by a factor of 10 (2 to 19%) in 2011. The high values of TTE from Saanich Inlet (excluded as artefacts of sampling) are close to the maximum values of transfer efficiency (74.6%) reported in a similar study for Sagami Bay, Japan (Ara & Hiromi 2007). Artefact values in TTE were also found by Gladyshev et al. (2011) in calculations of the transfer efficiency of essential polyunsaturated fatty acids and organic carbon from phytoplankton to secondary producers. These artefact values likely result from the time lags in metabolic processes of phytoplankton and zooplankton. Therefore, future studies are needed to directly assess these time lags between primary and crustacean productivity in order to determine the most accurate timescales over which to calculate TTE, thus avoiding high values of TTE that are thermodynamically impossible.

Our TTE estimates assume that primary productivity from phytoplankton is the only energy available to support crustacean productivity. However, given that heterotrophic protozoa (e.g. ciliates and dinoflagellates) and marine snow can also comprise a significant proportion of copepod diets (Dilling et al. 1998, 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 based only on phytoplankton productivity may be some-

what overestimated. Therefore, field studies incorporating crustacean productivity and estimates of both heterotrophic protozoa and primary productivity are necessary in order to determine the relative contributions of the 'classic' marine food chain and the microbial loop to TTE. Moreover, further 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 may be more important (Cushing 1990).

CONCLUSIONS

To our knowledge, this the first study to routinely couple community-level *in situ* production rates for phytoplankton and crustacean zooplankton. Our results highlight the importance of using field-derived crustacean production rates given that low productivity can occur even when zooplankton biomass is high. Had we used biomass estimates to calculate productivity, the temporal and interannual variability of productivity in Saanich Inlet may well have been missed. Furthermore, the fact that our chitobiase-based estimates of crustacean productivity and TTE fit with current estimates for other coastal marine ecosystems (Pauly & Christensen 1995) provides further support for the use of the chitobiase method to estimate crustacean productivity, which is less time-consuming and more practical for obtaining routine estimates in the field. However, because the chitobiase method captures all of the enzyme activity 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 led to a slight overestimation in crustacean production and resulting TTE calculations. In addition, exuviotrophic ciliates consume the fluid of exuvia after the crustacean moult (Landers et al. 2006), which could result in a slight underestimation of the total chitobiase enzyme in the water column. Finally, this method does not consider the contribution of adult production (i.e. egg production), which can represent up to 20% of the dry weight of mature females per day (Gómez-Gutiérrez & Peterson 1999).

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 bio-

mass 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). Ultimately, accurate estimates of both crustacean productivity and TTE will be particularly critical in terms of investigating the impact of (1) a potential increase in the occurrence of mismatches between lower and higher trophic levels, and (2) potential shifts in the size-structure of zooplankton communities under future warming scenarios.

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LITERATURE CITED

- 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. *Mar Biol* 159:2441–2453
- Allen SE, Wolfe MA (2013) Hindcast of the timing of the spring phytoplankton bloom in the Strait of Georgia, 1968–2010. *Prog Oceanogr* 115:6–13
- Ara K, Hiromi J (2007) Temporal variability in primary and copepod production in Sagami Bay, Japan. *J Plankton Res* 29:i85–i96
- 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). *J Plankton Res* 35:105–120
- Banse K (1982) Mass-scaled rates of respiration and intrinsic growth in very small invertebrates. *Mar Ecol Prog Ser* 9: 281–297
- Barwell-Clarke J, Whitney F (1996) Institute of Ocean Sciences nutrient methods and analysis. Canadian Technical Report of Hydrography and Ocean Sciences, No. 182
- 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. *Mar Coast Fish* 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
- Brzezinski MA, Nelson DM (1986) A solvent extraction method for the colorimetric determination of nanomolar

- concentrations of silicic acid in seawater. *Mar Chem* 19: 139–151
- Brzezinski MA, Nelson DM (1989) Seasonal changes in the silicon cycle within a Gulf Stream warm-core ring. *Deep Sea Res A* 36:1009–1030
- Castonguay M, Plourde S, Robert D, Runge JA, Fortier L (2008) Copepod production drives recruitment in a marine fish. *Can J Fish Aquat Sci* 65:1528–1531
- Collins AK, Allen SE, Pawlowicz R (2009) The role of wind in determining the timing of the spring bloom in the Strait of Georgia. *Can J Fish Aquat Sci* 66:1597–1616
- Cushing DH (1990) Plankton production and year-class strength in fish populations: an update of the match/mismatch hypothesis. *Adv Mar Biol* 26:249–293
- Dalsgaard J, St John MA, Kattner G, Muller-Navarra D, Hagen W (2003) Fatty acid trophic markers in the pelagic marine environment. *Adv Mar Biol* 46:225–340
- Devol AH (1981) Vertical distribution of zooplankton respiration in relation to the intense oxygen minimum zones in two British Columbia fjords. *J Plankton Res* 3:593–602
- Dilling L, Wilson J, Steinberg D, Alldredge AL (1998) Feeding by the euphausiid, *Euphausia pacifica*, and the copepod, *Calanus pacificus*, on marine snow. *Mar Ecol Prog Ser* 170:189–201
- Drinkwater KF, Jones EP (1987) Density stratification, nutrient and chlorophyll distributions in the Hudson Strait region during summer and their relation to tidal mixing. *Cont Shelf Res* 7:599–607
- Dugdale RC, Wilkerson F (1986) The use of ^{15}N to measure nitrogen uptake in eutrophic oceans: experimental considerations. *Limnol Oceanogr* 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. *Mar Ecol Prog Ser* 382:151–161
- Fulton J (1968) A laboratory manual for the identification of British Columbia marine zooplankton. No. 55
- Gargett AE, Stucchi D, Whitney F (2003) Physical processes associated with high primary production in Saanich Inlet, British Columbia. *Estuar Coast Shelf Sci* 56:1141–1156
- Gladyshev MI, Sushchik NN, Anishchenko OV, Makhutova ON and others (2011) Efficiency of transfer of essential polyunsaturated fatty acids versus organic carbon from producers to consumers in a eutrophic reservoir. *Oecologia* 165:521–531
- Gómez-Gutiérrez J, Peterson WT (1999) Egg production rates of eight calanoid copepod species during summer 1997 off Newport, Oregon, USA. *J Plankton Res* 21: 637–657
- 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. *Helgol Mar Res* 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. *Cont Shelf Res* 29:2257–2269
- 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. *Mar Biol* 73:31–36
- Hirst AG, Bunker AJ (2003) Growth of marine planktonic copepods: global rates and patterns in relation to chlorophyll *a*, temperature, and body weight. *Limnol Oceanogr* 48:1988–2010
- Hirst AG, Lampitt RS (1998) Towards a global model of *in situ* weight-specific growth in marine planktonic copepods. *Mar Biol* 132:247–257
- Hopcroft RR, Roff JC (1998) Zooplankton growth rates: the influence of size in nauplii of tropical marine copepods. *Mar Biol* 132:87–96
- Huntley ME, Lopez MDG (1992) Temperature-dependent production of marine copepods: a global synthesis. *Am Nat* 140:201–242
- Irvine JR, Crawford WR (2012) State of the physical, biological, and selected fishery resources of Pacific Canadian marine ecosystems in 2011. DFO Can Sci Advis Sec Res Doc 2012/072
- Jónasdóttir SH, Kiørboe T (1996) Copepod recruitment and food composition: do diatoms affect hatching success? *Mar Biol* 125:743–750
- Kainz M, Arts MT, Mazumder A (2004) Essential fatty acids in the planktonic food web and their ecological role for higher trophic levels. *Limnol Oceanogr* 49:1784–1793
- Kimmerer WJ (1987) The theory of secondary production calculations for continuously reproducing populations. *Limnol Oceanogr* 32:1–13
- Kimmerer WJ, McKinnon AD (1987) Growth, mortality, and secondary production of the copepod *Acartia tranteri* in Westernport Bay, Australia. *Limnol Oceanogr* 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. *Limnol Oceanogr* 39:493–507
- Klein Breteler WCM, Schogt N, Rampen S (2005) Effect of diatom nutrient limitation on copepod development: role of essential lipids. *Mar Ecol Prog Ser* 291:125–133
- Knotz S, Boersma M, Saborowski R (2006) Microassays for a set of enzymes in individual small marine copepods. *Comp Biochem Physiol A* 145:406–411
- Landers SC, Gómez-Gutiérrez J, Peterson WT (2006) *Gymnodinioides pacifica*, n. sp., an exuviotrophic ciliated protozoan (Ciliophora, Apostomatida) from euphausiids of the Northeastern Pacific. *Eur J Protistol* 42:97–106
- 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. *J Plankton Res* 27:647–662
- Mackas DL, Tsuda A (1999) Mesoplankton in the eastern and western subarctic Pacific: community structure, seasonal life histories, and interannual variability. *Prog Oceanogr* 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. *Can J Fish Aquat Sci* 58: 685–702
- Mackas DL, Batten S, Trudel M (2007) Effects on zooplankton of a warmer ocean: recent evidence from the Northeast Pacific. *Prog Oceanogr* 75:223–252
- Mackas DL, Greve W, Edwards M, Chiba S and others (2012) Changing zooplankton seasonality in a changing

- ocean: comparing time series of zooplankton phenology. *Prog Oceanogr* 97–100:31–62
- Mackas D, Galbraith M, Faust D, Masson D and others (2013) Zooplankton time series from the Strait of Georgia: results from year-round sampling at deep water locations, 1990–2010. *Prog Oceanogr* 115:129–159
- Miller AJ (1990) *Subset selection in regression* Chapman & Hall, London
- Muzzarelli RAA (1977) Chitinases and related enzymes. In: Muzzarelli RA (ed) *Chitin*. Pergamon Press, Oxford p 155–177
- Oosterhuis SS, Baars MA, Klein Breteler WCM (2000) Release of the enzyme chitinase by the copepod *Temora longicornis*: characteristics and potential tool for estimating crustacean biomass production in the sea. *Mar Ecol Prog Ser* 196:195–206
- Paffenhöfer GA, Harris RP (1976) Feeding, growth and reproduction of the marine planktonic copepod *Pseudocalanus elongatus* Boeck. *J Mar Biol Assoc UK* 56: 327–344
- Parrish C (1999) Determination of total lipids, lipid classes and fatty acids in aquatic samples. In: Arts M, Wainmann BC (eds) *Lipids in freshwater ecosystems*. Springer, New York, NY, p 4–17
- 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
- Peterson WT, Tiselius P, Kiørboe T (1991) Copepod egg production, moulting and growth rates, and secondary production, in the Skagerrak in August 1988. *J Plankton Res* 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. *Mar Biol* 141: 353–365
- Poulet SA, Ianora A, Laabir M, Klein Breteler WCM (1995) Towards the measurement of secondary production and recruitment in copepods. *ICES J Mar Sci* 52:359–368
- R Development Core Team (2013) *R: a language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna
- Runge JA, Roff JC (2000) The measurement of growth and reproductive rates. In: Harris RP, Weibe PH, Lenz J, Skjoldal HR, Huntley M (eds) *ICES zooplankton methodology manual*. Academic Press, San Diego, CA, p 401–454
- Sastri AR, Dower JF (2006) Field validation of an instantaneous estimate of in situ development and growth for marine copepod communities. *Can J Fish Aquat Sci* 63: 2639–2647
- Sastri A, Dower JF (2009) Interannual variability in chitinase-based production rates of the crustacean zooplankton community in the Strait of Georgia, British Columbia, Canada. *Mar Ecol Prog Ser* 388:147–157
- Sastri AR, Roff JC (2000) Rate of chitinase degradation as a measure of development rate in planktonic Crustacea. *Can J Fish Aquat Sci* 57:1965–1968
- Sastri AR, Nelson RJ, Varela DE, Young KV, Wrohan I, Williams WJ (2012) Variation of chitinase-based estimates of crustacean production rates in high latitude waters. *J Exp Mar Biol Ecol* 414–415:54–61
- Sommer U, Aberle N, Engel A, Hansen T and others (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. *J Cons Int Explor Mer* 18:117–140
- Suchy KD, Dower JF, Sastri AR, Neil MC (2013) Influence of diet on chitinase-based production rates for the harpacticoid copepod *Tigriopus californicus*. *J Plankton Res* 35:657–667
- Takahashi M, Seibert DL, Thomas WH (1977) Occasional blooms of phytoplankton during summer in Saanich Inlet, BC, Canada. *Deep-Sea Res* 24:775–780
- 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. *Mar Coast Fish* 4: 415–437
- Timothy DA, Soon MYS (2001) Primary production and deep-water oxygen content of two British Columbia fjords. *Mar Chem* 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. *Mar Biol Res* 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. *J Mar Syst* 115–116:10–32
- Utermöhl H (1958) Zur Vervollkommnung der quantitativen Phytoplankton-Methodik. *Mitt Int Ver Theor Angew Limnol* 9:1–38
- Uye S (1988) Temperature-dependent development and growth of *Calanus sinicus* (Copepoda: Calanoida) in the laboratory. *Hydrobiologia* 167–168:285–293
- Vargas CA, Martinez RA, Cuevas LA, Pavez MA and others (2007) The relative importance of microbial and classical food webs in a highly productive coastal upwelling area. *Limnol Oceanogr* 52:1495–1510
- Vidal J (1980) Physioecology of zooplankton. II. Effects of phytoplankton concentration, temperature, and body size on the development and molting rates of *Calanus pacificus* and *Pseudocalanus* sp. *Mar Biol* 56:135–146
- Viso AC, Marty JC (1993) Fatty acids from 28 marine microalgae. *Phytochemistry* 34:1525–1533
- Yang EJ, Kang HK, Yoo S, Hyun JH (2009) Contribution of auto- and heterotrophic protozoa to the diet of copepods in the Ulleung Basin, East Sea/Japan Sea. *J Plankton Res* 31:647–659

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