

Stable isotopes in subtidal food webs: Have enriched carbon ratios in benthic consumers been misinterpreted?

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Abstract

We examined the hypothesis that the $\delta^{13}\text{C}$ of benthic consumers is depleted in offshore, deeper waters where kelp and benthic microalgae are absent by conducting three analyses: (1) we analyzed $\delta^{13}\text{C}$ (and $\delta^{15}\text{N}$) of consumers along a nearshore–offshore gradient in the field, (2) we analyzed $\delta^{13}\text{C}$ (and $\delta^{15}\text{N}$) of consumers placed in cages at various depths and distances from shore, and (3) we reexamined published stable isotope values for consumers taken at various depths from studies around the world. In all three situations, we did not observe the predicted change in $\delta^{13}\text{C}$ values of benthic consumers. In both the literature and in our field measurements, the $\delta^{13}\text{C}$ values for consumers were on average separated from the values for suspended particulate organic matter by 4‰, irrespective of depth, indicating that factors other than the feeding on ^{13}C -rich benthic primary producers may explain the enriched carbon ratios in benthic consumers (e.g., selective feeding on enriched particles). The hypothesis that the high $\delta^{13}\text{C}$ ratio in suspension and deposit feeders reflects feeding on benthic primary producers is thus tenuous and requires further investigation.

In coastal temperate and polar oceans, there exist three potentially important sources of primary production: phytoplankton, kelp, and benthic microalgae (microphyto-benthos). Although it is generally acknowledged that phytoplankton are the main food source available to benthic consumers, productivity measurements suggest that kelp and benthic microalgae may also be an important food source. Kelps are among the most productive plants on the planet, and up to 90% of their net production may end up as detritus (Mann 1988), which can be ingested by suspension and deposit feeders (Duggins and Eckman 1997). In a review of benthic microalgae productivity measurements in coastal ecosystems, Cahoon (1999) showed that they also can markedly contribute to overall production in temperate regions (around $60 \text{ g C m}^{-2} \text{ yr}^{-1}$), even at depths of $>20 \text{ m}$. For example, Glud et al. (2002) found that the net photosynthetic rate of benthic microalgae at depths $<30 \text{ m}$ in a Greenland fjord was twofold greater than that of phytoplankton in overlying waters. Although certain studies have shown an increased growth rate of suspension feeders in the presence of kelp both in the field (Duggins et al. 1989) and in controlled feeding

experiments (Duggins and Eckman 1997), direct evidence for the use of benthic algae by suspension and deposit feeders comes from the analysis of stable isotopes. Observations from more straightforward approaches, such as stomach content analyses, are difficult to obtain for these types of consumers.

Marine primary producers typically have well-separated carbon isotope ratios, which are conserved in their respective consumers with an enrichment (i.e., an increase in heavy isotopes) ranging from 0.5‰ to 1.2‰ between trophic levels (Vander Zanden and Rasmussen 2001; McCutchan et al. 2003). In contrast, nitrogen isotope ratios are often similar in primary producers but are enriched by $\sim 2.5\%$ in herbivores and by 3.5‰ to 4.0‰ with further increases in trophic level (McCutchan et al. 2003). Consequently, carbon isotope ratios are mainly used to determine the sources of primary production, whereas nitrogen isotope ratios are used to identify the trophic level of organisms (Michener and Schell 1994). Kelp and benthic microalgae have a much heavier carbon isotopic ratio than phytoplankton because of the depletion of light carbon in the relatively thick boundary layer that develops around them, since they are attached to the bottom. Many studies have made use of this difference to evaluate the relative importance of the different benthic primary producers and phytoplankton in coastal food webs. Although most studies in subtidal systems focus on the importance of kelp (Duggins et al. 1989; Kaehler et al. 2000; Fredriksen 2003), a number also consider the importance of benthic microalgae (Takai et al. 2002; Kang et al. 2003). All of these studies show that most benthic consumers are rich in ^{13}C , suggesting that benthic primary producers are major contributors to benthic food webs. They often appear to account for $>50\%$ of the carbon in benthic consumers. However, a few studies conducted in offshore waters, far removed from sources of kelp or benthic microalgae, also report enriched carbon ratios in benthic consumers (Fry 1988; Hobson et al. 1995). These observations suggest that

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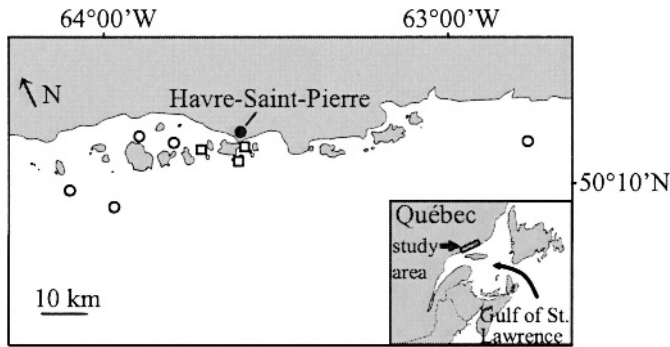


Fig. 1. Location of sampling sites on the north shore of the Gulf of St. Lawrence, Canada. Squares represent the nearshore sites where we dived to collect benthic animals and plants. Circles are deeper, offshore sites where organisms were collected by dredging.

the heavy carbon isotope ratios in benthic consumers are not entirely due to primary production from benthic algae.

If the ingestion of ^{13}C -rich benthic primary producers is responsible for the heavy carbon found in nearshore consumers, then a testable prediction is that consumers located away from these sources should have a lighter carbon ratio, similar to that of phytoplankton-derived organic matter, since kelp and benthic microalgae only occur in nearshore waters. The present study examines this hypothesis using both field experiments and previously published data. We analyzed the $\delta^{13}\text{C}$ ratios of benthic consumers taken from natural habitats across a nearshore-offshore gradient. Further, we analyzed the tissues of scallops that had been suspended in cages at various depths and distances from shore for a period of 2 months. Finally, we reviewed published $\delta^{13}\text{C}$ values of suspension and deposit feeders sampled at various depths in temperate and polar oceans.

Materials and methods

Study area—Our study was made along a 100-km stretch of coastline in the region of the Mingan Islands on the northern shore of the Gulf of St. Lawrence (Fig. 1). This region is characterized by cold waters (summer temperatures averaging 7°C) and by a predominant current from the east. Kelp beds, mainly of *Alaria esculenta* and to a lesser extent *Laminaria digitata*, form a dense macroalgal fringe that extends to at most 5 m in depth at exposed sites and to lesser depths in sheltered locations. The kelps are limited to shallow depths because of intensive grazing of the abundant green sea urchin *Strongylocentrotus droebachiensis*. The movement of the kelp fronds by wave action in shallow water limits the shoreward movement of the urchins and their attachment on the kelp fronds (Himmelman 1991). The grazer-resistant macroalga *Agarum cribrosum* occurs in patches down to a depth of 30 m (Himmelman 1991). Annual phytoplankton productivity in the region is about $200\text{ g C m}^{-2}\text{ yr}^{-1}$ (Steven 1975), which is nine times less than the annual productivity of North Atlantic kelp beds ($1,750\text{ g C m}^{-2}\text{ yr}^{-1}$) reported by Mann (1972). Since kelp beds in the Mingan Islands extend to

about 0.3 km offshore on average (Nadon unpubl. data), productivity from kelp and phytoplankton should be similar in the first 3 km from shore ($0.3\text{ km} \times 9$). Although these productivity measurements are fairly dated and may have changed because of ocean warming, they should provide a good indication of the difference between kelp bed and pelagic productivity in the region. More recent annual measurements are not available for the region. Another potentially important source of primary production is from benthic microalgae, which form a thin but visible brown layer on soft bottoms. They can cover up to 80% of sandy bottoms in shallow water but are absent below 30 m (Nadon unpubl. data).

Sampling—Samples for isotopic analysis were collected from June to August in 2003 and 2004. Phytoplankton samples, taken as suspended particulate organic matter (POM), were obtained by prefiltering up to 40 liters of water through a $64\text{-}\mu\text{m}$ screen and then onto a 5- or $10\text{-}\mu\text{m}$ Nitex mesh. The material remaining on the 5- or $10\text{-}\mu\text{m}$ screens, representing 5– $64\text{-}\mu\text{m}$ and 10– $64\text{-}\mu\text{m}$ size fractions, respectively, was collected and dried. On some dates, GF/F filters were used to obtain a smaller size fraction (0.7– $10\text{ }\mu\text{m}$). In 2003, six 10– $64\text{-}\mu\text{m}$ and three 0.7– $10\text{-}\mu\text{m}$ POM samples were collected from the surface at nearshore locations. In 2004, only the 5– $64\text{-}\mu\text{m}$ size fraction was collected. We first collected three samples from nearshore surface waters during a delayed spring bloom in June. These samples were of interest since POM $\delta^{13}\text{C}$ may be enriched during periods of high productivity (Fry and Wainright 1991). We also collected samples after the bloom (July and August), both from nearshore surface waters (three samples) and 3, 6, and 10 km offshore (one surface sample at each distance) and at 20-m depth nearshore (three samples collected with a Niskin bottle).

We dived at three nearshore sites to collect samples of three common benthic consumers, the scallop *Chlamys islandica*, the sea cucumber *Cucumaria frondosa*, and the soft coral *Gersemia rubiformis*. Individuals were also collected with a dredge at up to five deeper, offshore sites (Fig. 1). At least three individuals per species were collected at each site. We took samples of the muscle tissue for all animals. We sampled kelp from both exposed and protected sites, given that water turbulence is reported to affect carbon ratios in kelp (Simenstad et al. 1993). We ground up whole kelp fronds and then took a subsample to minimize error due to possible variation in isotopic ratios in different portions of fronds (Stephenson et al. 1984). We assumed that kelp detritus had the same isotopic ratios as fresh kelp plants (Stephenson et al. 1986). Benthic microalgae were sampled by collecting the top layer of conspicuous brown algal mats with syringes, prefiltering this material with a $64\text{-}\mu\text{m}$ mesh filter, and then collecting the filtrate retained on a $5\text{-}\mu\text{m}$ mesh filter (5– $64\text{-}\mu\text{m}$ size fraction). The $64\text{-}\mu\text{m}$ filter was used to limit the amount of inorganic particles in our samples. All animal and plant samples were dried at 60°C for a minimum of 24 h immediately following collection.

We used the carbon ratio of fast-growing juvenile scallops suspended in cages to evaluate the vertical and

horizontal distribution of material from benthic primary producers in the water column. We predicted that the contribution of different primary producers, and thus the isotope carbon ratios, would differ among the scallops in the various cages, given the varying distances from the nearshore kelp beds and from microalgae on the sea floor. The scallops were initially collected from the shallow (15–20 m) portion of a scallop bed at Île du Fantôme (~100 m from the nearest kelp bed) and then transplanted in cages to other locations that varied in depth and distance to shore. At two offshore sites (~2.3 km from land in 30-m depths), cages were suspended from anchored lines (1) near the surface, (2) in midwater, and (3) a few centimeters off the bottom. It was impossible to place lines beyond 2.3 km because of boat traffic and for safety reasons. As previously stated, the overall kelp and phytoplankton production is likely about the same for the first 3 km from shore. However, the availability of kelp detritus for scallops placed in cages ~2 km from kelp beds should be sufficiently reduced to cause a reduction in the $\delta^{13}\text{C}$ in scallop tissues. Similarly, although we cannot rule out input from benthic microalgae to the scallops located in the offshore surface cages, the reduced photosynthetic activities of microalgae at these depths coupled with the distance from the bottom (30 m) should greatly reduce potential inputs from this source. To provide a point of comparison for the scallops suspended in offshore cages, we also suspended cages near the surface and a few centimeters off the bottom at two nearshore sites (<100 m from land in 10-m depths). After 2 months, we collected the muscle tissue from at least three scallops from each of these cages to determine isotopic ratios. Although we could not leave the scallops for a longer period of time for logistical reasons, 2 months appears sufficient for a change in the isotopic ratio to occur in oysters (Riera and Richard 1997) and the half-life of carbon in mammalian muscle tissues has been estimated at just 28 days (Tieszen et al. 1983). In August 2004 we also sampled 40-mm scallops that had been suspended 3 m above the bottom in aquaculture cages for 2 yr (initial size = 15 mm) at a site 16-m deep located about 100 m from shore (about 700 m from kelp beds). Although these scallops were only moderately removed from benthic algae (700 m from kelp and 3 m from microalgae), they had been suspended in the water column for an extended period and thus could have registered some change in $\delta^{13}\text{C}$ if a change in diet had occurred.

Sample analysis—The POM and benthic microalgae samples were exposed to HCl fumes for 24 h to remove traces of carbonates prior to analysis. Samples were not defatted since only muscle tissue containing little fat was analyzed. Stable isotope samples were analyzed in an isotope mass ratio spectrometer (Fisons Instruments, model VG Prism Isotech) at the Commission Géologique du Canada Laboratory in Quebec City. Stable isotope ratios were expressed in δ notation as parts per thousand (‰) using the standard definition: $\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1,000$, where X is ^{13}C or ^{15}N and R is the corresponding ratio $^{13}\text{C}:^{12}\text{C}$ or $^{15}\text{N}:^{14}\text{N}$. Pee Dee Belemnite was the standard for $\delta^{13}\text{C}$ and atmospheric N_2 for

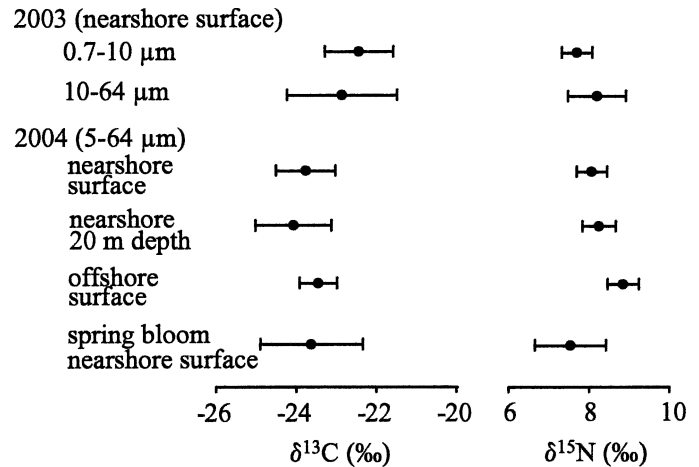


Fig. 2. Mean isotopic ratios of carbon and nitrogen (\pm SD) for different size fractions of particulate organic matter (POM) sampled at various locations in 2003 and 2004.

$\delta^{15}\text{N}$. Replicates of internal standards showed analytical errors to be on the order of $\pm 0.2\text{‰}$ for both carbon and nitrogen. If carbon ratios for kelp and benthic microalgae are similar, it should be possible to estimate the percentage contribution of benthic primary producers (BPP) compared to POM in benthic consumers using the following mixing model equation:

$$\%_{\text{BPP}} = (\delta^{13}\text{C}_{\text{consumer}} - I - \delta^{13}\text{C}_{\text{POM}}) / (\delta^{13}\text{C}_{\text{BPP}} - \delta^{13}\text{C}_{\text{POM}}) \times 100 \quad (1)$$

where I is the average postphotosynthetic fractionation of $\delta^{13}\text{C}$ per trophic level.

Literature review—We reviewed 20 studies encompassing 29 different locations (see Web Appendix 1 at http://www.aslo.org/lo/toc/vol_51/issue_6/2828al.pdf for the complete list), which provided 88 measurements of $\delta^{13}\text{C}$ and 59 measurements of $\delta^{15}\text{N}$ for suspension and deposit feeders. For each consumer at each site we calculated the difference between the isotope ratio of the consumer and the background POM ratio ($\delta^{13}\text{C}_{\text{consumer}} - \delta^{13}\text{C}_{\text{POM}}$) and then plotted these values against depth. We chose depth instead of shore distance as a measure of the presence of benthic algae since shore distance can lead to some confusion, as when values are on offshore reefs colonized by kelp and benthic algae. We assumed, in accordance with the authors in these studies, that the POM samples were composed mostly of phytoplankton-derived material. Linear regression analyses were applied to determine whether the relationship changed with depth, as would be predicted if materials from primary benthic producers became less important further from shore.

Results

Primary producers—For the samples of POM, $\delta^{13}\text{C}$ values ranged from -25‰ to -22‰ and $\delta^{15}\text{N}$ values ranged from 6‰ to 8‰ (Fig. 2). These values are similar to previously reported values in the Gulf of St. Lawrence (Tan and Strain 1979). The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values did not differ

significantly between the two size fractions (0.7–10 μm and 10–64 μm) sampled in 2003 (Fig. 2; $t = -0.30$, $\text{df} = 7$, $p = 0.78$ for $\delta^{13}\text{C}$; $t = 1.18$, $\text{df} = 7$, $p = 0.28$ for $\delta^{15}\text{N}$). Also, an analysis of variance (ANOVA) applied to samples taken at different locations in 2004 did not indicate differences among the samples from different sites and dates for either carbon or nitrogen (Fig. 2; $F_{2,5} = 0.42$, $p = 0.68$ for $\delta^{13}\text{C}$; $F_{2,5} = 2.16$, $p = 0.21$ for $\delta^{15}\text{N}$), although the $\delta^{15}\text{N}$ of the offshore POM tended to be slightly more enriched (+1‰). The average POM carbon ratios were slightly more enriched (+1‰) in 2003 than in 2004 ($t = 2.52$, $\text{df} = 18$, $p = 0.02$). *SeaWiFS* satellite images of chlorophyll *a* concentration in the northern Gulf of St. Lawrence showed that the spring phytoplankton bloom occurred during May in 2003 and thus prior to when we took samples (June through August). However, the phytoplankton bloom of 2004 was late and mainly occurred during June to early July. No differences were detected between samples taken during the bloom (19 June, 01 July, and 10 July) and those taken after the bloom (27–29 July and 03–16 August) for either $\delta^{13}\text{C}$ ($t = -0.46$, $\text{df} = 6$, $p = 0.66$) or $\delta^{15}\text{N}$ ($t = 1.56$, $\text{df} = 6$, $p = 0.17$). Since we did not detect significant variations related to size fraction, depth, and distance from shore for either $\delta^{13}\text{C}$ or $\delta^{15}\text{N}$, and because the range of isotopic signals was relatively small, we pooled all POM measurements ($n = 20$), giving an average of -23.3‰ for $\delta^{13}\text{C}$ and 8.1‰ for $\delta^{15}\text{N}$. The small degree of spatial variability in the POM isotopic ratios, even between nearshore and offshore samples, suggests that the POM was mostly composed of phytoplankton-derived material, rather than detritus from terrestrial sources or nearshore benthic algae.

Of the three kelp species analyzed, the two fast-growing kelps, *A. esculenta* and *L. digitata*, and the grazer-resistant kelp, *A. cribrosum*, had well-separated carbon ratios (-18.9‰ , -15.9‰ , and -22.7‰ , respectively), but similar nitrogen ratios (6.7‰ , 6.8‰ , and 6.0‰ , respectively). The kelp carbon ratios showed high variability both within (mean $\text{SD} = 1.3$) and among sites (mean $\text{SD} = 1.5$). This was expected because of the uneven storage of compounds with variable isotope ratios within a plant (Stephenson et al. 1984) and marked differences in water turbulence among the sites sampled (Simenstad et al. 1993). Despite this variability, *A. esculenta* and *L. digitata*, the highly productive nearshore kelp species that likely contribute most to overall benthic primary production, had carbon and nitrogen ratios well separated from those of POM (Fig. 3).

Finally, the carbon ratio for benthic microalgae was -19.4‰ ($\text{SD} = 0.2$) and the nitrogen ratio 6.0‰ (0.2). The $\delta^{13}\text{C}$ values were low compared to those reported in the literature: -16.7‰ (Corbisier et al. 2004), -14.1‰ (Kang et al. 2003), and -14.3‰ (Machas et al. 2003). This may be because our samples included some POM that had settled on the bottom, in which case pure benthic microalgae would have had an even more enriched $\delta^{13}\text{C}$.

Consumers—The benthic consumers at nearshore sites were generally highly enriched in $\delta^{13}\text{C}$ (Fig. 3), with carbon ratios closer to those of benthic microalgae (-19.4‰) and

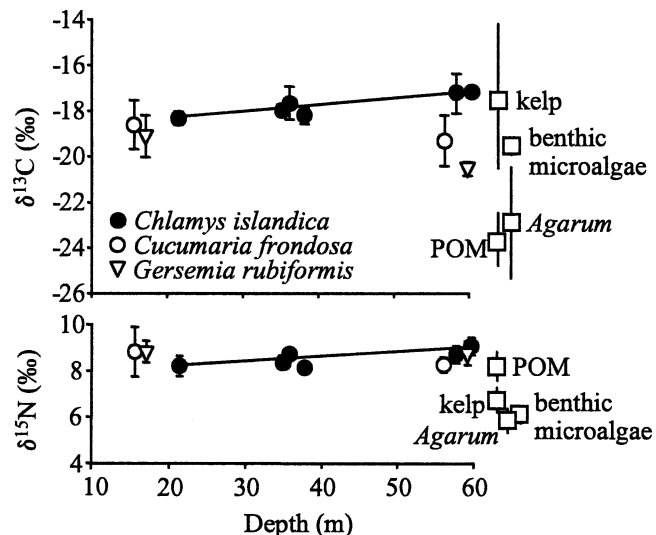


Fig. 3. Mean carbon and nitrogen isotope ratios for scallops (*C. islandica*), sea cucumbers (*C. frondosa*), and soft corals (*G. rubiformis*) collected at various depths (and distances from shore) using a dredge. The mean isotope ratios ($\pm\text{SD}$) of the various primary producers are shown as reference points (the value for kelp is the pooled mean for the two fast-growing kelp species, *A. esculenta* and *L. digitata*).

the two fast-growing kelp species (mean $\sim 18\text{‰}$) than to that of POM (-23.3‰). Surprisingly, individuals taken with a dredge at greater depths did not have $\delta^{13}\text{C}$ values closer to POM (Fig. 3). In fact, the $\delta^{13}\text{C}$ values for scallops increased with increasing depth (linear regression, $F_{1,4} = 21.06$, $r^2 = 0.84$, $p = 0.01$), although the change was only by 0.9‰ between the shallowest and deepest sites. Scallop $\delta^{15}\text{N}$ also slightly increased with increasing depth, but not significantly (regression, $F_{1,4} = 5.63$, $r^2 = 0.25$, $p = 0.07$). These slight enrichments may be related to the slightly higher isotope values noted for offshore POM (Fig. 2). This pattern was not apparent for *G. rubiformis* and *C. frondosa*, since their $\delta^{13}\text{C}$ decreased slightly ($\sim 1\text{‰}$) at greater depth, although they were still far removed from POM $\delta^{13}\text{C}$, by 3‰ and 4‰ , respectively. Similarly, we did not detect variations in the $\delta^{13}\text{C}$ ratio for juvenile scallops maintained for 2 months in cages suspended at various depths in nearshore and offshore areas (Table 1; ANOVA, $F_{5,13} = 2.01$, $p = 0.15$). We also did not detect a difference in the $\delta^{13}\text{C}$ ratio between the juvenile scallops from the aquaculture cages, which had been suspended for 2 yr at 3 m above the bottom at a nearshore site, and those from the scallop bed at Île du Fantôme (Table 1; $t = 0.71$, $\text{df} = 4$, $p = 0.51$). We did observe a slight decrease (about 1‰) in the $\delta^{13}\text{C}$ of juvenile scallops in 2004, compared to the ratio of the adult scallops collected in 2003. Although this difference could be related to the change in scallop size, it is more likely because of a change in the $\delta^{13}\text{C}$ signal for POM. It was slightly lower in 2004 (-24.7‰) than in 2003 (-23.7‰).

Literature review—POM carbon isotope ratios averaged -23.1‰ across the 20 studies reviewed and did not change significantly with the general depth of the sampled area (linear regression, $F_{1,20} = 2.81$, $r^2 = 0.09$, $p = 0.11$).

Table 1. Mean carbon ratios of juvenile scallops (\pm SD) from the collection site, a scallop bed at Île du Fantôme, from aquaculture cages and from cages suspended at various depths at nearshore and offshore sites. n is the number of situations sampled. A least three scallops were analyzed in each situation.

Locations	Distance from kelp (km)	Distance from bottom (m)	Potential input of benthic algae	n	Mean $\delta^{13}\text{C}$ (‰)
Scallop collection site	0.1	0	High	1	-19.2
Aquaculture cages	0.7	3	Moderate	1	-19.5
Nearshore					
Bottom cages	0	0.3	Very high	2	-19.4 \pm 0.2
Surface cages	0	10	High	2	-18.9 \pm 0.8
Offshore					
Bottom cages	2.0	0.3	Moderate	2	-19.9 \pm 1.0
Middle cages	2.0	10	Low	2	-19.2 \pm 0.5
Surface cages	2.0	30	Very low	1	-20.3

Although the difference between consumers and POM carbon ratios varied from 2.4‰ to 4.7‰ among the eight taxonomic groups examined in our literature review (Table 2), this variation was not significant (ANOVA, $F_{6,46} = 1.36$, $p = 0.24$). Similarly, no effect of taxonomic group was detected for nitrogen ratios (ANOVA, $F_{6,46} = 0.89$, $p = 0.51$). Thus, we pooled the various consumers at each location in the subsequent analysis. Further, given the large number of bivalves sampled in these studies, we also examined them separately. The data from several studies show that the difference between consumer and POM $\delta^{13}\text{C}$ values (all consumers together or bivalves analyzed separately) either increased or decreased as collection sites increased in depth (lines connecting points in Fig. 4). The slope was almost zero for the overall relationship at the 28 sites, both for all consumers together and bivalves analyzed separately (Fig. 4; linear regressions, $F_{1,24} = 0.01$, $r^2 = 0.001$, $p = 0.91$ for all consumers; $F_{1,26} = 0.09$, $r^2 = 0.003$, $p = 0.77$ for bivalves only). Almost all consumers, irrespective of depth, were highly enriched in $\delta^{13}\text{C}$ compared to POM, and the variation in values reflected a bell-shaped curve with a mean of 4.1‰ (Fig. 5). The only notable exception was a study in St. Margaret Bay, Nova Scotia, where two bivalve species had $\delta^{13}\text{C}$ values 0.5‰ lower than that of POM, probably because POM was particularly enriched (-19‰). The enrichment in nitrogen between POM and benthic consumers closely agreed with

the expected value of 3‰ (Vander Zanden and Rasmussen 2001), but the distribution of values was strongly skewed toward low values (Fig. 5). In short, benthic consumers in temperate and polar ecosystems generally have a background $\delta^{13}\text{C}$ enrichment of about 4.1‰ compared to POM, and the degree of enrichment does not appear to be related to the proximity of benthic primary producers.

Discussion

Enriched $\delta^{13}\text{C}$ values in nearshore benthic consumers, as observed in our study, are usually interpreted as indicating a strong dependence on benthic primary producers as a food source. Since fast-growing kelp species and benthic microalgae are limited to shallow depths (by urchin grazing and light penetration, as explained previously), we predicted that the $\delta^{13}\text{C}$ values of benthic consumers would decrease moving offshore because of an increased use of phytoplankton-derived material, and further that $\delta^{13}\text{C}$ values would decrease if consumers were maintained for a period in the upper water column. However, our comparison of nearshore and offshore consumers (an offshore gradient of up to 10 km) and the $\delta^{13}\text{C}$ values observed in scallops that were suspended for 2 months at different depths and distances from shore did not support these predictions. In fact, scallop $\delta^{13}\text{C}$ values were slightly more enriched in offshore than in nearshore scallops.

Table 2. Mean difference (\pm SD) between the $\delta^{13}\text{C}$ (‰) in the tissues of benthic consumers and the $\delta^{13}\text{C}$ (‰) values for POM as reported by 20 studies for different groups of suspension and deposit feeders in temperate and polar oceans.

Taxonomic group	n	$\delta^{13}\text{C}_{\text{benthos}} - \delta^{13}\text{C}_{\text{POM}}$	n	$\delta^{15}\text{N}_{\text{benthos}} - \delta^{15}\text{N}_{\text{POM}}$
Bryozoa	8	2.4 \pm 1.4	4	2.0 \pm 1.6
Octocorallia	3	3.3 \pm 0.8	2	3.2 \pm 3.7
Polychaeta	3	3.4 \pm 2.3	2	1.3 \pm 1.1
Porifera	8	3.6 \pm 2.1	3	2.5 \pm 2.1
Ascidacea	6	4.0 \pm 1.5	3	3.9 \pm 2.8
Holothuroidea	7	4.1 \pm 1.7	7	3.0 \pm 2.5
Bivalvia	51	4.4 \pm 2.1	37	3.2 \pm 2.2
Ophiuroidea	2	4.7 \pm 0.2	1	3.9
Total	88	4.1 \pm 2.0	59	3.0 \pm 2.2

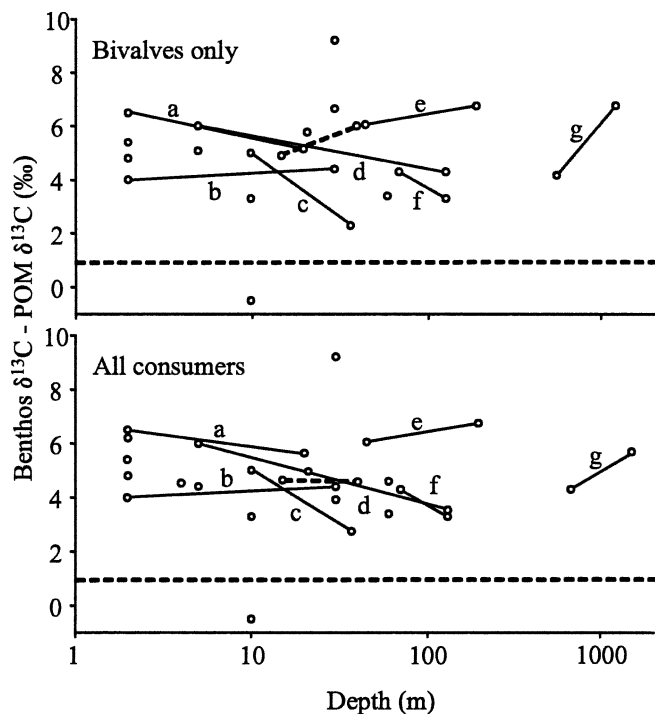


Fig. 4. Mean difference between $\delta^{13}\text{C}$ values for POM and benthic primary consumers in relation to depth (bivalves only and all consumers together) calculated from values taken in 20 studies in the literature. Points connected by solid lines are for different depths reported in single studies (a, Takai et al. 2002; b, Machas et al. 2003; c, Kang et al. 2003; d, Kaehler et al. 2000; e, Hobson et al. 1995; f, Fry 1988; g, Iken et al. 2005), and the dashed line connects the minimum and maximum depth sampled in the present study. The dashed horizontal line at the bottom is the predicted location of organisms feeding strictly on POM (with the usual 1‰ enrichment).

Furthermore, although offshore soft corals and sea cucumbers showed a slight decrease in $\delta^{13}\text{C}$, they were still far removed from the POM $\delta^{13}\text{C}$ ratio. Further, the carbon ratios of aquaculture scallops that had been suspended above the bottom for 2 yr did not differ from scallops in a natural bottom habitat at Île du Fantôme. Finally, our literature review failed to detect lighter carbon ratios for consumers with increasing depth (up to 1,200 m). Rather, benthic primary consumers almost always have a carbon ratio about 4‰ heavier than POM. The lack of a decrease in carbon ratios with increasing distance from benthic primary producers casts doubt on the interpretation that the enriched $\delta^{13}\text{C}$ in nearshore benthic consumers is attributable to feeding on kelp detritus and benthic microalgae. The above observations suggest that other enrichment processes could confound estimations of the amount of carbon originating from benthic primary producers in the coastal zone.

Unfortunately, a simple explanation for the general enrichment of benthic consumers is not available. The trophic shift of $\delta^{13}\text{C}$ between consumers and their known diets is typically around 1‰ (Vander Zanden and Rasmussen 2001; McCutchan et al. 2003). This means that we cannot account for about 3.1‰ of the increase in $\delta^{13}\text{C}$

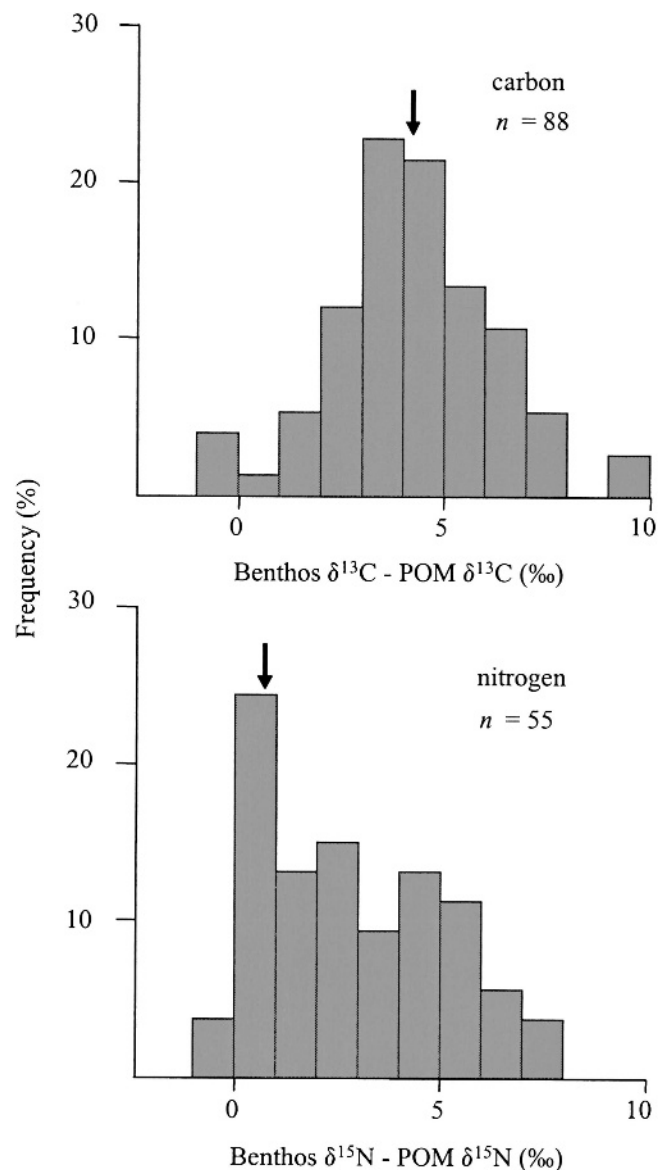


Fig. 5. Frequency distribution of the difference between $\delta^{13}\text{C}$ values for POM and that of individual benthic primary consumer species calculated from values reported by 20 studies in the literature. The number of isotopic ratios (n) is indicated in each graph. The arrows show the mean differences found in the current study (Mingan Islands).

values between POM and benthic primary consumers, especially in offshore locations where benthic algae are absent. A combination of several plausible explanations may explain this enrichment.

First, the carbon ratio of POM can become heavier as light carbon becomes depleted during periods of high productivity, thus providing a seasonal pulse of heavy carbon to benthic consumers. Fry and Wainright (1991) reported POM with particularly heavy $\delta^{13}\text{C}$ (−19‰ to −15‰) during spring bloom events over Georges Bank and in other temperate regions. These observations, however, do not form a general pattern. Our study and others

(Kaehler et al. 2000; Fredriksen 2003) found no enrichment of the carbon ratio of POM during spring bloom events. Further, although Lorrain et al. (2002) measured a heavier POM carbon ratio during high-productivity periods in the Bay of Brest, France, this enrichment in POM $\delta^{13}\text{C}$ values could only account for a small fraction of the total enrichment in the scallops they studied. Although this explanation is worth considering, it cannot by itself explain the general enrichment pattern found in temperate and polar oceans as indicated in our review of the literature.

A second potential explanation for the high $\delta^{13}\text{C}$ enrichment in benthic consumers is that the carbon ratio of POM becomes enriched as particles sink in the water column and are reworked and degraded by consumers and bacteria (e.g., fecal pellet production, the microbial loop). Subsequent degradation on the ocean floor could further enrich the $\delta^{13}\text{C}$ ratio of organic matter made available to consumers by resuspension. Furthermore, the burial and upbrining of refractory organic matter in the sediment layer by bioturbation could also cause carbon enrichment. The hypothesis that the ingestion of degraded, ^{13}C -rich, organic matter is responsible for the elevated $\delta^{13}\text{C}$ found in benthic consumers would be highly appealing, especially in offshore waters where a more complex pelagic food web typically leads to highly degraded material reaching the seafloor. However, we have yet to find any $\delta^{13}\text{C}$ measurements that clearly show that the degradation of organic matter causes significant increases in $\delta^{13}\text{C}$. Water column measurements, taken to depths of 500 m, show that POM $\delta^{13}\text{C}$ actually becomes lighter with increasing depth and thus degradation level (Tan and Strain 1979; Ostrom et al. 1997). Tan and Strain (1979) also report that superficial sediment organic matter $\delta^{13}\text{C}$ are usually similar to surface POM values down to 500-m depths in the Gulf of St. Lawrence, and Naidu et al. (2000) found no difference in sediment $\delta^{13}\text{C}$ versus settling POM $\delta^{13}\text{C}$ at depths ranging from a few meters to 3.6 km in a study that sampled 465 locations in various regions of the Arctic Ocean. Similarly, Davenport and Bax (2002) found no difference between sediment $\delta^{13}\text{C}$ and surface POM $\delta^{13}\text{C}$ in 103 measurements taken off southeastern Australia at up to 200-m depths. Furthermore, carbon isotope ratios from different sediment depth layers taken by Peters et al. (1978) did not reveal major differences in organic matter $\delta^{13}\text{C}$ between layers (down to more than 3 m in the sediments). The experimental aging of algal- and plant-derived organic matter for 2 months in field and laboratory conditions did not result in a change in their $\delta^{13}\text{C}$ (Stephenson et al. 1986). Finally, the possible influence of decomposition on $\delta^{13}\text{C}$ values would also fail to explain why benthic consumers suspended near the surface, which have access to fresh particles, still have the same highly enriched carbon ratios as observed in consumers on the bottom (current study; Machas et al. 2003; Takai et al. 2002). All these lines of argument tend to show that the diagenesis of organic matter does not cause an increase in $\delta^{13}\text{C}$ and that the ingestion of refractory organic matter does not satisfactorily explain the elevated $\delta^{13}\text{C}$ found in benthic consumers of polar and temperate oceans.

A third hypothesis is that the enrichment of benthic primary consumers is caused by their selective feeding on enriched POM (either before or after ingestion). Possibly, the trend toward more enriched $\delta^{13}\text{C}$ in consumers such as bivalves and sea cucumbers compared to sponges and bryozoans (Table 2) reflects their greater capacity to select particles, given their more complex filtering and digestive systems. Many studies, reviewed by Ward and Shumway (2004), demonstrate the capacity of bivalves to select particles according to size and even chemical properties. Also, holothurians are thought to select particles according to size and quality in function of tentacle morphology (Billett 1991), polychaetes seem to select particles according to size (Bock and Miller 1999), and sponges are known to be able to discriminate between types of bacteria (Wilkinson et al. 1984). The selection of organic particles by benthic feeders is a widespread mechanism that could account for the elevated $\delta^{13}\text{C}$ of benthic consumers in both the coastal zone (e.g., preferential selection of kelp or benthic microalgae particle) and offshore areas (e.g., selection of ^{13}C -rich diatoms).

It is important to note that the enriched carbon isotope values found in offshore consumers could still be caused by unknown trophic processes not present in coastal areas. In such a case, the $\delta^{13}\text{C}$ enrichment of nearshore consumers could very well be entirely attributed to feeding on benthic algae, as was done in past studies. However, our two tentative explanations for the enrichment of offshore consumers (feeding on enriched POM produced during high-productivity periods and ^{13}C -rich particle selection by consumers) should also apply to coastal food webs. This suggests that the widespread occurrence of highly enriched $\delta^{13}\text{C}$ in nearshore benthic consumers cannot entirely or largely be attributed to materials from benthic primary producers (i.e., kelp or benthic microalgae). Workers that do attribute this enrichment to benthic primary producers are likely to overestimate the importance of benthic primary producers in coastal ecosystems.

Recommendations for future studies—Based on our results, it appears that factors other than the ingestion of benthic primary producers can cause a high $\delta^{13}\text{C}$ enrichment in benthic consumers in temperate and polar waters. It is thus necessary to control for this enrichment when studying carbon isotopes in benthic organisms. One way to do this would be to use the $\delta^{13}\text{C}$ values of organisms feeding strictly on phytoplankton-derived material (e.g., offshore or suspended at the surface) as a reference point instead of the POM $\delta^{13}\text{C}$ in a mixing model (see Eq. 1):

$$\%_{\text{BPP}} = \frac{[(\delta^{13}\text{C}_{\text{nearshore consumer}} - I) - (\delta^{13}\text{C}_{\text{offshore consumer}} - I)]}{[\delta^{13}\text{C}_{\text{BPP}} - (\delta^{13}\text{C}_{\text{offshore consumer}} - I)]} \times 100 \quad (2)$$

where BPP represents benthic primary producers and I is the average postphotosynthetic fractionation of $\delta^{13}\text{C}$ per trophic level. Note that the I used to adjust the $\delta^{13}\text{C}$ of the nearshore consumer may be different from the one used to adjust the offshore consumer if both organisms are not at the same trophic level. If no difference is found between nearshore and offshore organisms, as in our study, no

conclusion can be made about the importance of benthic primary producers. If collecting offshore or suspended benthic organisms is not possible, another solution may be to compare the isotopic ratio of benthic consumers with pelagic animals, as done by Hobson et al. (1995). However, the enrichment in pelagic consumers can be variable, since some consumers have values close to POM (Kaehler et al. 2000) and others are highly enriched (Martineau et al. 2004). If none of these options are possible, one should expect a basic enrichment of about 4‰ before interpreting the enrichment due to heavy carbon coming from benthic primary producers, although this value varies between studies and consumer taxa. Ignoring the widespread pattern of enrichment in consumers can lead to erroneous conclusions about the importance of benthic primary producers, especially in studies where there is no information on the isotopic ratios of benthic consumers feeding exclusively on phytoplankton-derived material. We suggest that primary consumers (e.g., bivalves) be used as a baseline in calculating trophic levels of benthic predators, since the trophic fractionation between primary producers and consumers in benthic communities is highly variable and unpredictable. Clearly, there is a need for studies on the causes of the high $\delta^{13}\text{C}$ enrichment in benthic consumers. One approach would be to examine particle selection in benthic consumers (suspension and deposit feeders) and stable isotopes simultaneously.

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