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1 **Nitrogen stable isotopes reveal age-dependent dietary shift in the Japanese scallop**

2 ***Mizuhopecten yessoensis* (Jay, 1857)**

3

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18 Running Head: Dietary shift in Japanese scallop

19 **Abstract**

20 Ontogenetic niche shifts in diet are a consequence of changes in body size or resource  
21 partitioning between age classes. To better resolve the feeding patterns of the Japanese  
22 scallop *Mizuhopecten yessoensis*, we examined the relative importance of age and size  
23 in the diet of this species using stable isotope ratios of carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ )  
24 from 2006 to 2009. Contribution of food sources was quantified using an isotope  
25 mixing model by comparing the muscle tissue isotope ratios to those of suspended  
26 particulate organic matter (SPOM) and their zooplankton prey (e.g. micro- and meso-  
27 zooplankton). Unlike the  $\delta^{13}\text{C}$  values which remained constant with age and size,  
28 muscle  $\delta^{15}\text{N}$  values were more positively correlated with age accounting for 69% of  
29 variations than size with only 46%. Increasing  $^{15}\text{N}$  values with age suggested that shifts  
30 in diet from SPOM to micro- and meso-zooplankton occurred during ontogeny in *M.*  
31 *yessoensis*. Results of the isotope mixing model indicated that SPOM contribution to  
32 scallop's diet decreased from 68% to 8% while those of zooplankton increased from 15  
33 to 50% with increasing age. This study concludes that age-related dietary shift explains  
34 the enrichment of  $^{15}\text{N}$ , as a result of predation on zooplankton by *M. yessoensis*.

35 **Keywords:** Age;  $^{15}\text{N}$  enrichment; *Mizuhopecten yessoensis*; Diet shift; **Size**;  
36 Zooplankton; Isotope mixing

## 37 **Introduction**

38 Ontogenetic diet shifts are common among invertebrates, as a direct consequence of  
39 increase in body size [1] or resource partitioning between age classes. Body size is one  
40 of the most fundamental traits that affect individual characteristics determining food  
41 acquisition [2]. The age of the individuals however, is related with resource use  
42 capability [3]. Age-related shifts in diet composition lead to decreased potential intra-  
43 specific competition for resources [4,5]. As a predator ages, it will feed on larger and  
44 high quality food sources to meet the required energy for growth and reproductive  
45 functions [6] and an ontogenetic change in diet is expected to occur [7,8].

46 For a long time, scallops feed mainly on phytoplankton [9] and sinking particles  
47 [10,11], even though other larger zooplankton has been found in the guts of marine  
48 bivalves [9]. It has been shown that bivalves are not only herbivores, but omnivores that  
49 feed on bacteria, nano-zooplankton and micro-zooplankton [12–14]. **In fact, mussels**  
50 **are reported to have assimilated about 37 to 54% of meso-zooplankton, an**  
51 **evidence of a strong trophic link between mussels and zooplankton [14].** Scallops  
52 reared at Lake Saroma, Hokkaido, Japan feed not only on suspended particulate organic  
53 matter (SPOM) but also on zooplankton [15].

54 Stable isotope analysis of carbon ( $\delta^{13}\text{C}$ ) and nitrogen ( $\delta^{15}\text{N}$ ) are now  
55 increasingly used to estimate trophic position and the food sources of marine organisms  
56 [16–18]. The  $\delta^{15}\text{N}$  of an animal tissue is enriched by 3-4‰ relative to its diet [19] while  
57  $\delta^{13}\text{C}$  is less, about 1‰ [20]. Several studies have shown that factors such as age or size  
58 are known to influence the isotopic composition of bivalve tissues. For instance, large  
59 differences in  $\delta^{15}\text{N}$  occur with size, particularly in mussels and fishes than with age  
60 [19,21] but not in walleye [22]. Hence, it is necessary to re-examine the importance of  
61 these factors as determinants of ontogenetic niche shifts in diet of many bivalve species.

62 The Japanese scallop *Mizuhopecten yessoensis* (Jay, 1857) is a commercially  
63 important temperate scallop reared in the coastal waters of northern Japan. It is  
64 cultivated using the “rotational” harvesting system, where a particular culture zone is  
65 seeded with juveniles after the adult-sized individuals are harvested [23]. Previous  
66 studies have analyzed some aspects of *Mizuhopecten yessoensis* biology and ecology,  
67 including distribution and abundance [24], recruitment [23], and feeding rates [25,26].  
68 However, information concerning the ontogenetic dietary changes of this species is not  
69 known.

70 Therefore, this study aimed (1) to examine age and size-specific shifts in the diet  
71 of Japanese scallop; and (2) to estimate the contribution of suspended particulate

72 organic matter (SPOM) and zooplankton prey in the scallop's diet using stable isotopes  
73 of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ . We hypothesized that if the ingestion of  $^{15}\text{N}$ -rich larger zooplankton  
74 is responsible for the heavy nitrogen found in adult scallops, then it follows that juvenile  
75 individuals should have a lighter nitrogen isotope ratio, similar to that of phytoplankton-  
76 derived organic matter.

77

## 78 **Materials and Methods**

### 79 *Study area*

80 The study was carried out in Tokoro, Sea of Okhotsk ( $44^{\circ}13'\text{N}$ ,  $143^{\circ}55'\text{E}$ ), where there  
81 is an extensive culture of Japanese scallops on the seabed (**Figure 1**). It measures a  
82 maximum depth of 40–65 m. The sediments are mainly coarse sand or pebbles [27]. In  
83 the spring and autumn 2007, surface salinity ranges from 32.3 to 34.1. The fishing  
84 ground is divided into four culture zones (Zone A to D), where each zone contains  
85 different age class at a given time [24].

86

### 87 *Sampling and analyses*

88 Suspended particulate organic matter (SPOM) was sampled at Station S-O ( $44^{\circ}10'\text{N}$   
89 and  $143^{\circ}58.9'\text{E}$ ) between May and November 2007, July 2008, and July 2009. Depth-

90 integrated water samples for SPOM were collected with a 10-L Niskin bottle attached  
91 to a Kevlar wire from 0, 10, 20, 30 and 40 m depths. SPOM (1 to 2 L) samples were  
92 filtered onto pre-combusted (450°C for 5 h) Whatman GF/F (25 mm) glass fiber filters  
93 immediately after collection.

94 Zooplankton samples were collected with vertical net tows from bottom to  
95 surface at Station S-0 with NORPAC nets of **100 and 330 µm mesh sizes** during the  
96 same collection dates with SPOM and stored either in a 125 or 250 mL wide-mouth  
97 polyethylene (PE) bottle. Zooplankton fractions (100–330 µm and >330 µm) were  
98 filtered onto pre-weighed, pre-combusted (450°C for 5 h) Whatman GF/F glass fiber (25  
99 mm) filters.

100 Scallop individuals in four age classes (**1<sup>+</sup> to 4<sup>+</sup>-year-old**) from six **cohorts**  
101 were obtained monthly either from April or July to December in three seasons (**2006–**  
102 **2008**) and May 2009, except during the winter months. Shell height was measured to  
103 the nearest 0.1 mm using a vernier caliper. Soft tissues were dissected and muscle  
104 samples were removed, washed with Milli-Q water and weighed.

105 All samples were stored at -30°C until analysis. Small portion of muscle samples  
106 were freeze-dried and ground with mortar and pestle. Lipids were extracted from  
107 muscle tissues using the modified “Folch” extraction method [28]. SPOM and

108 zooplankton filters were freeze-dried and exposed to HCl fumes for 4 h to eliminate  
109 inorganic carbon.

110 All samples were measured for elemental and stable isotopes. About 1 mg of  
111 two to three muscle samples were weighed with a microbalance and packed in tin  
112 capsules for analysis. SPOM filters were put in tin capsules and fashioned into a tablet  
113 using a hand compressor. Zooplankton that was retained on the filters (1 to 1.5 mg for  
114 100–330  $\mu\text{m}$  and 1.5 to 2 mg for  $>330 \mu\text{m}$  fractions) were scraped carefully, prepared in  
115 duplicate and placed in tin capsules and folded. Stable isotope ratios of carbon and  
116 nitrogen were measured with a Thermo Electron Delta V Plus Continuous Flow IRMS  
117 (Bremen, Germany) attached to a Thermo Electron Flash Elemental Analyzer 1112  
118 Series (Milan, Italy). Precision was better than 0.2‰ for both  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  estimated  
119 with alanine and tyrosine as internal standards (Kyoto University, Kyoto, Japan). Stable  
120 isotope ratios are expressed in delta notation ( $\delta^{13}\text{C}$  or  $\delta^{15}\text{N}$ ) and are reported in per mil  
121 (‰) using the equation:  $\delta^{13}\text{C}_{\text{sample}}$  or  $\delta^{15}\text{N}_{\text{sample}} = (R_{\text{sample}}/R_{\text{standard}} - 1)$ , where  $R$   
122 =  $^{13}\text{C}/^{12}\text{C}$  or  $^{15}\text{N}/^{14}\text{N}$  with Vienna Pee Dee Belemnite (VPDB) and atmospheric nitrogen  
123 as **international reference standards** for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively. **Delta  $\delta^{13}\text{C}$**   
124 **values are normalized using VPDB-LSVEC calibration curve.**

125

126 *Data analyses*

127 All statistical analyses were done using the Number Cruncher Statistical System (NCSS,  
128 07.1.4 version) 2007 software [29]. We applied one-way analyses of variance  
129 (ANOVA) to test significant differences in  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values among scallop  
130 populations and post hoc Tukey's-Kramer test was used when significant differences  
131 were detected. Linear regression analysis was performed to examine relationships  
132 between muscle tissue  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values, and their age and size. Prior to regression  
133 analysis, the actual age of scallops was recalculated from the difference between  
134 sampling dates and seeding time (3<sup>rd</sup> week of May until 1<sup>st</sup> week of June) and expressed  
135 as fractions of a year.

136 Scallop's trophic position (TP) was estimated based on a 3-year average  $\delta^{15}\text{N}$   
137 isotope ratio of SPOM ( $5.3 \pm 0.8\text{‰}$ ; F. Aya, unpubl. data) and using the average trophic  
138 fractionation of  $1.7\text{‰}$  [16, F. Aya, unpubl. data] where the average trophic level of the  
139 Japanese scallop can be calculated as [30]:

140

141 
$$\text{TP} = 1 + (\text{scallop } \delta^{15}\text{N} - \text{SPOM } \delta^{15}\text{N}) / 1.7$$
 using SPOM as the basal level of the  
142 food web

143

144 To account for the trophic increases among the populations, the percentage  $\delta^{13}\text{C}$   
 145 and  $\delta^{15}\text{N}$  contributions of micro- (**PL100–330**), mesozooplankton (**PL>330**) and SPOM  
 146 to scallops' diet were estimated with an isotope mixing model [31]. The isotopic values  
 147 of SPOM during 2007 and 2008 at Station S-0 were reported elsewhere [11].  
 148 **Application of the mixing model required correction of consumer  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$**   
 149 **values using tissue-diet isotopic fractionation values. The difference between**  
 150 **isotopic ratios of gut content and that of muscle tissue was used as representation**  
 151 **of tissue-diet isotopic fractionation [11, F. Aya, unpubl. data]. Muscle  $\delta^{13}\text{C}$  and**  
 152  **$\delta^{15}\text{N}$  values were corrected for 3.4‰ and 1.7‰ isotopic fractionation, respectively,**  
 153 prior to analysis [16, F. Aya, unpubl. data]. The contribution is calculated as follows:

154

$$155 \quad \text{Mix } \delta^{13}\text{C} = f_{\text{SPOM}} * \delta^{13}\text{C}_{\text{SPOM}} + f_{\text{PL100-330}} * \delta^{13}\text{C}_{\text{PL100-330}} + f_{\text{PL>330}} * \delta^{13}\text{C}_{\text{PL>330}};$$

156 **and**

$$157 \quad \text{Mix } \delta^{15}\text{N} = f_{\text{SPOM}} * \delta^{15}\text{N}_{\text{SPOM}} + f_{\text{PL100-330}} * \delta^{15}\text{N}_{\text{PL100-330}} + f_{\text{PL>330}} * \delta^{15}\text{N}_{\text{PL>330}};$$

158 **with**

$$159 \quad f_{\text{SPOM}} + f_{\text{PL100-330}} + f_{\text{PL>330}} = 1$$

160

161 where the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values of the consumer is the combination of the  
162  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values of potential food sources (SPOM, micro- and meso-  
163 zooplankton) and  $f$  is the fractional contribution of a source [31]. The last equation  
164 specifies that the fractions must equal to 1.

165

## 166 **Results**

167 Suspended particulate organic matter (SPOM) and zooplankton showed no significant  
168 differences in  $\delta^{13}\text{C}$  ( $P < 0.05$ ) (Table 1). **C/N molar ratios in SPOM and zooplankton**  
169 **were between 6.1 and 6.8**, indicating phytoplankton-derived organic material.  
170 However, **zooplankton, composed of micro- and meso-zooplankton, was 1.2 to 1.5‰**  
171 more enriched in  $^{15}\text{N}$  than SPOM, suggesting different trophic position between primary  
172 producers and zooplankton.

173 Muscle  $\delta^{15}\text{N}$  values significantly increased from 7.2 to 8.3‰ with age ( $P < 0.01$ ;  
174 Table 2), indicating differences in food preferences and trophic position. The  $\delta^{15}\text{N}$   
175 values of age-3<sup>+</sup> and 4<sup>+</sup> scallops were similar and significantly higher than those of age-  
176 1<sup>+</sup> and 2<sup>+</sup> individuals ( $P < 0.05$ ). Muscle  $\delta^{15}\text{N}$  were independent of location or site with  
177 values ranging from 7.6 to 8.1‰ (mean = 7.9‰). Ontogenetic changes in stable isotope  
178 ratios were found for all the cohorts examined, showing significant positive trends of

179 increasing  $\delta^{15}\text{N}$  values with age ( $n = 83$ ,  $r^2 = 0.69$ ,  $P < 0.001$ ):  $\delta^{15}\text{N} = 6.7 + 0.35$  (age)  
180 (in years) and size ( $n = 83$ ,  $r^2 = 0.46$ ,  $P < 0.001$ ):  $\delta^{15}\text{N} = 5.3 + 0.02$  ( $S_H$ ) (in mm)  
181 (**Figures 2a,b**). However, muscle  $\delta^{15}\text{N}$  values were highly correlated with age than with  
182 size among cohorts, indicating greater influence of age on  $\delta^{15}\text{N}$ . On the contrary, the  
183  $\delta^{13}\text{C}$  values did not vary significantly among the four age classes (Table 2), although  
184 age-1<sup>+</sup> individuals had less depleted  $^{13}\text{C}$ . Moreover,  $\delta^{13}\text{C}$  values remained constant on  
185 average among parameters age and size, in which the  $\delta^{13}\text{C}$  values were neither  
186 correlated with age nor size (**Figures 2c,d**). On the average, *M. yessoensis* occupied a  
187 trophic position between 2.3 to 2.7 (Table 3).

188 Application of the mixing model to muscle  $\delta^{15}\text{N}$  values indicated that **SPOM**  
189 **contribution to scallop's diet decreased from 68% to 8%, being the highest** in age-  
190 1<sup>+</sup> and 2<sup>+</sup>, and lowest for age-3<sup>+</sup> and 4<sup>+</sup> individuals (**Figure 3, Table 4**). **However,**  
191 **consumption of zooplankton increased from 15 to 50% as scallop aged (Figure 3,**  
192 **Table 4)**. These results indicated that *M. yessoensis* to some degree shift their diet from  
193 SPOM to zooplankton, potentially influencing the  $\delta^{15}\text{N}$  values of this species. **However,**  
194 **application of the mixing model to muscle  $\delta^{13}\text{C}$  values did not agree well with the**  
195 **isotope values of their presumed potential food sources (Figure 3), indicating that**

196 scallops were preferentially assimilating highly enriched component (i.e. sinking  
197 particles [11]) in the suspended organic matter pool.

198

## 199 Discussion

200 Our study provides information on the relative importance of age as an indicative of  
201 dietary change in *Mizuhopecten yessoensis*. It also uncovers an ontogenetic shift in diet  
202 wherein juvenile scallops mainly depend on suspended particulate organic matter  
203 (SPOM), whereas adult individuals forage on zooplankton as the most important dietary  
204 component. We indeed observed with our isotopic dataset that juveniles and adult  
205 scallops, independent of age and size, exhibited similar  $\delta^{13}\text{C}$  values indicative of similar  
206 quality of food sources. This is also evident from the lack of significant relationship  
207 between the  $\delta^{13}\text{C}$  values for muscle and age or size because of the small variability in  
208 the  $\delta^{13}\text{C}$  isotopic values. These results however are not supported by previous dietary  
209 studies using bivalves [31,32], where carbon isotope ratios showed greater differences  
210 than nitrogen isotope ratios. The large differences in  $\delta^{15}\text{N}$  between young and adult  
211 scallops may suggest that they preferred different food sources but to some extent,  
212 scallops still occupied the same trophic position (2.3–2.7). Our estimates of scallop's  
213 trophic position supported the results of [33] confirming the herbivorous and

214 detritivorous nature of *M. yessoensis*. Bivalve species, scallops in particular, are feeding  
215 on a mixed diet sources but mainly on phytoplankton [9] or sinking particles in their  
216 natural habitats [10,11]. The increase in  $^{15}\text{N}$  values with age may result from foraging  
217 on zooplankton, which supports the observed enrichment in  $^{15}\text{N}$ . The  $\delta^{15}\text{N}$  isotope ratios  
218 could be expected to increase with age if nitrogen undergoes fractionation during the  
219 transamination or deamination process where the lighter  $\delta^{14}\text{N}$  is excreted at a faster rate  
220 than  $\delta^{15}\text{N}$  from the body of some animals [19]. The isotope fractionation, with the  
221 lighter isotope reacting faster in each direction of the reversible reactions has been  
222 determined to occur during transamination reactions [34]. We may interpret that age is a  
223 stronger determinant of the diet of *M. yessoensis* and the increase of  $^{15}\text{N}$  values with age  
224 as indicative of the ontogenetic foraging shift from SPOM to zooplankton.

225 *Mizuhopecten yessoensis*, as they age and grow, gradually shift their diet from  
226 SPOM to zooplankton, and through assimilation, the isotopic ratios of their food  
227 sources were reflected in the scallop muscle tissues. Our study indicated significant  
228 discrepancy in the isotopic composition between SPOM and zooplankton which may  
229 have a large impact on the  $\delta^{15}\text{N}$  values observed in the different age classes of *M.*  
230 *yessoensis*. In fact, a significant increase in  $\delta^{15}\text{N}$  values with age in the present study  
231 suggests ontogenetic shifts similar to that of freshwater mussels [35] and walleye [22]

232 and shows that age is a stronger determinant of dietary change than size. However,  
233 previous studies on mussels [19] and rainbow smelts [21] failed to identify any  
234 significant change of  $\delta^{15}\text{N}$  with age. The percentage of the total variation in  $\delta^{15}\text{N}$  as  
235 explained by size (46%) was lower than those accounted for by age (69%), possibly due  
236 to constant growth among adult scallops. Indeed, *M. yessoensis* reached an asymptote  
237 size of 126 mm between age-3<sup>+</sup> and 4<sup>+</sup>, the difference in growth rate between these age  
238 classes was not significant [F. Aya, unpubl. data].

239         Our analysis of the isotope mixing model supports the hypothesis that juveniles  
240 (age-1<sup>+</sup> and 2<sup>+</sup>) exploit higher proportion of SPOM than adults (age-3<sup>+</sup> and 4<sup>+</sup>), possibly  
241 due to difference in foraging capabilities. Juveniles have small mouth gape which limits  
242 their consumption to SPOM, whereas adult individuals, with larger mouth gape size,  
243 can feed on a wider range of prey including SPOM to large-sized food sources such as  
244 zooplankton. Previous work on the same species showed that for juveniles, sinking  
245 particles, composed of aggregated fresh microalgae, are considered more important  
246 energy source although they also consumed SPOM [11]. This explains the higher  
247 contributions of SPOM for the age-1<sup>+</sup> and 2<sup>+</sup> scallops, while zooplankton was  
248 significantly important for the age-3<sup>+</sup> and 4<sup>+</sup> individuals. Feeding studies analyzing  
249 stomach contents of other bivalve species have observed shifts from planktivorous to

250 piscivorous diets in different size classes [31]. In other studies, zooplankton is  
251 considered a potential food source for zebra mussels [14], while larger zebra mussels  
252 have been reported to suppress zooplankton abundance [36]. Also, [35] reported that the  
253 stomach contents of fan shell *Pinna nobilis* consist of water column calanoid copepods,  
254 an indication of exploitation on zooplankton by this species. Older and larger mussel  
255 individuals, with larger inhalant siphons, may eat drifting invertebrates or other  
256 organisms that feed higher on the food chain [35]. These age-related dietary resource  
257 partitioning is consistent with physiological differences arising with age [37]. **However,**  
258 **the large discrepancy between the diet  $\delta^{13}\text{C}$  values and those of scallop muscle**  
259 **tissues could be attributed to the low assimilation of low amount of carbon from**  
260 **ingested material [11].**

261 In summary, the present study demonstrates the greater influence of age than  
262 size on  $\delta^{15}\text{N}$  isotope ratios of *M. yessoensis*. Results of the isotope mixing model  
263 suggest unequal contributions of SPOM and zooplankton to scallops' diet, indicating  
264 gradual shift in prey consumption. These findings explain the potential accumulation  
265 of  $^{15}\text{N}$  with age, as suggested by a strong nutritional dependence of *M. yessoensis* on  
266 zooplankton.

267

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274

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## Figure legends

Figure 1. Sampling stations and culture zones for scallops in the Sea of Okhotsk.

**Figure 2. Relationship between muscle  $\delta^{15}\text{N}$  and (a) age and (b) size, and between muscle  $\delta^{13}\text{C}$  and (c) age and (d) size (as shell height,  $S_H$ ) from different cohorts of *Mizuhopecten yessoensis*.**

Figure 3. Dual isotope plots showing the natural  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values (mean  $\pm$  SD) of scallop muscle tissues in different age classes and potential food sources (PL100–330, micro-zooplankton; PL>330, meso-zooplankton; SPOM, suspended particulate organic matter). **Scallop age classes are denoted by open symbols, with expected diet values after trophic correction of 3.4‰ and 1.7‰ for muscle  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  isotope values, respectively, indicated by solid symbols.**

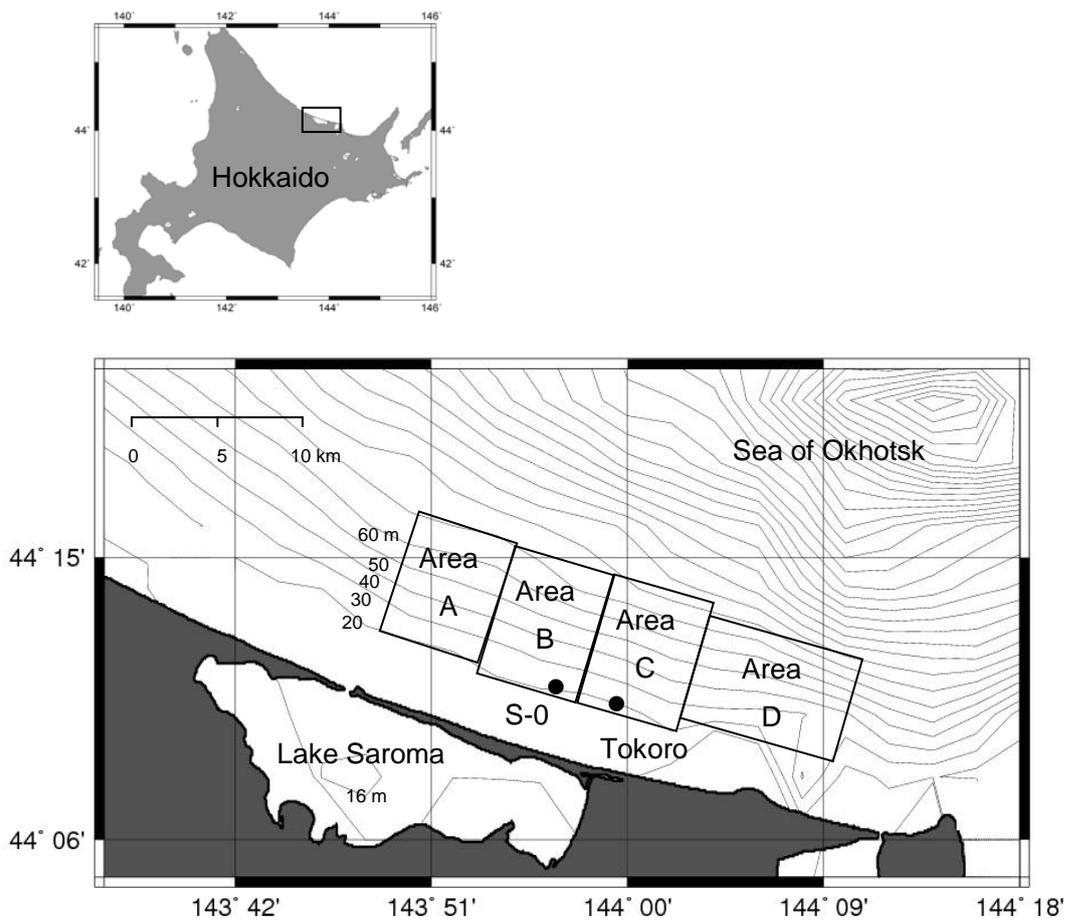
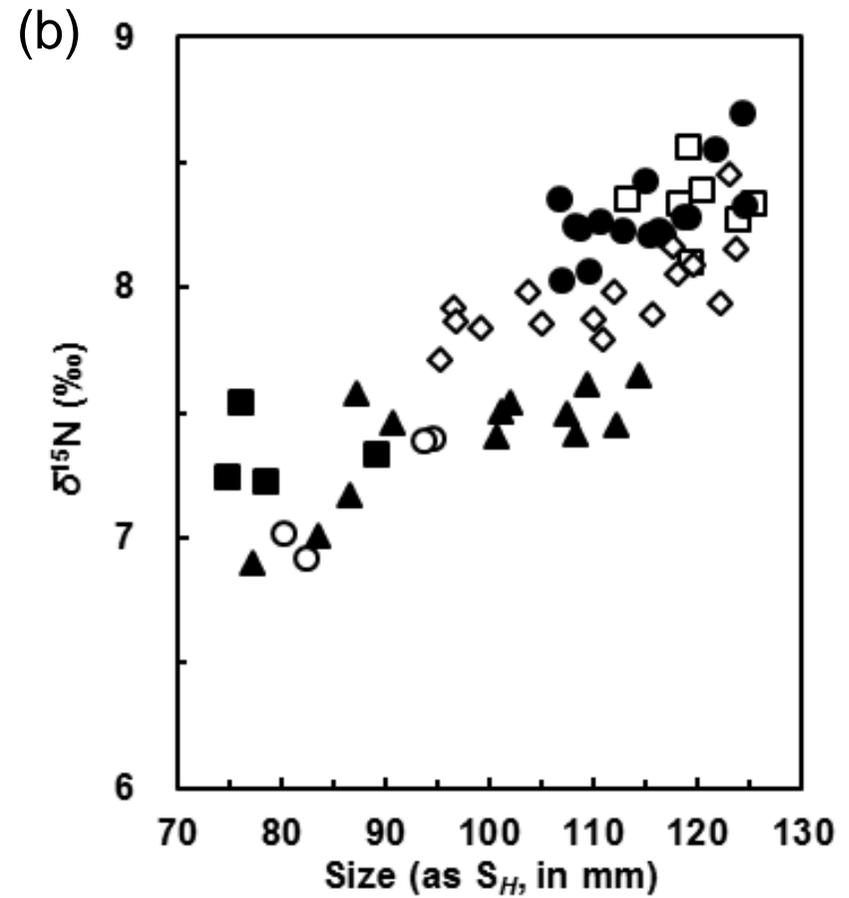
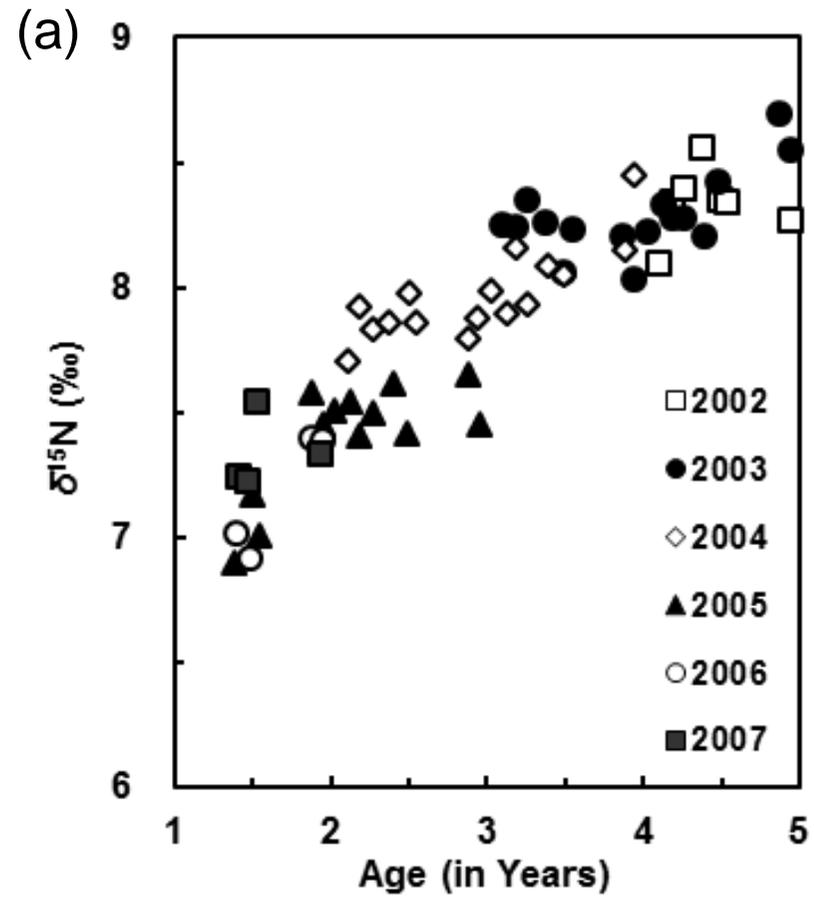


Figure 1



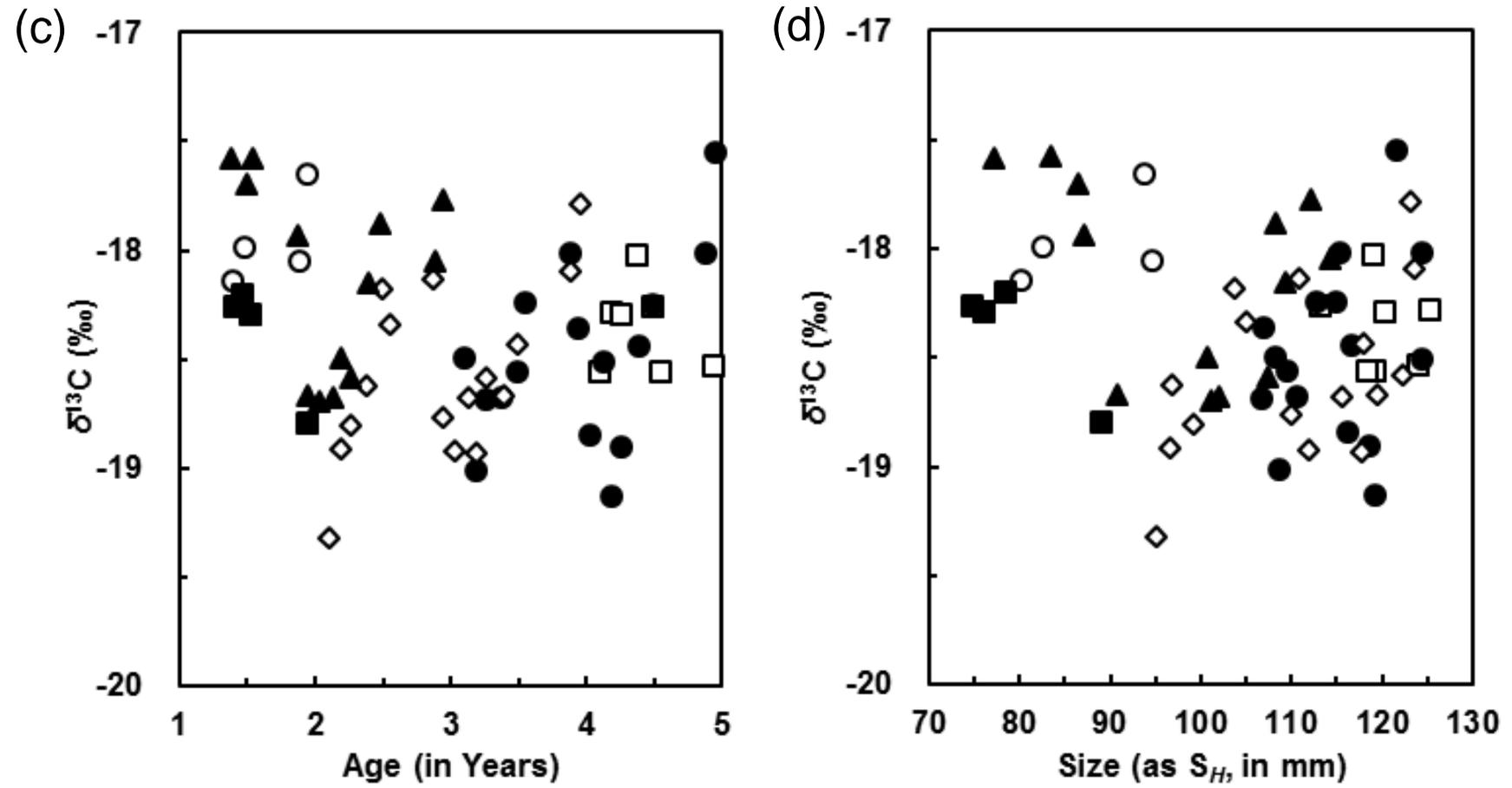


Figure 2

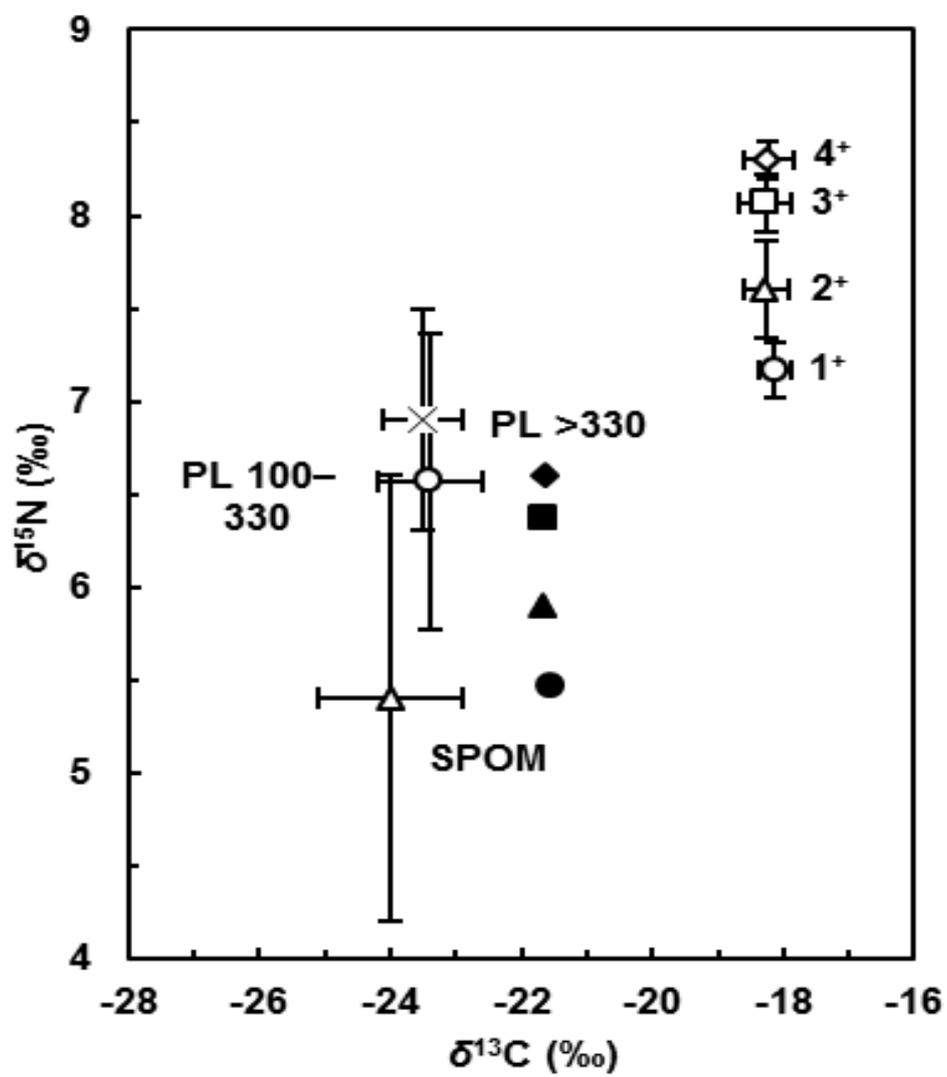


Fig. 3

Table 1

Stable isotopes ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) and carbon to nitrogen (C/N) molar ratios of potential food sources collected at Station S-0, Sea of Okhotsk

Food source	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	C/N molar ratios
SPOM	$-24.0 \pm 1.1$ (25) <sup>a</sup>	$5.4 \pm 1.2$ (24) <sup>a</sup>	$6.1 \pm 1.4$ <sup>a</sup>
Zooplankton			
100–330 $\mu\text{m}$	$-23.4 \pm 0.8$ (8) <sup>a</sup>	$6.6 \pm 0.8$ (8) <sup>b</sup>	$6.7 \pm 1.4$ <sup>a</sup>
>330 $\mu\text{m}$	$-23.5 \pm 0.6$ (8) <sup>a</sup>	$6.9 \pm 0.6$ (8) <sup>b</sup>	$6.8 \pm 0.9$ <sup>a</sup>

SPOM, suspended particulate organic matter. Values in parenthesis represent the number of samples analyzed. Column means followed by a different letter superscript are significantly different at  $P < 0.05$

Table 2

Stable isotope ratios ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) of muscle tissues in different age classes and sizes

Age class (in Years)	Shell height (mm)	<i>N</i>	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
1 <sup>+</sup>	65-95	13	-18.1 ± 0.3 <sup>a</sup>	7.2 ± 0.2 <sup>a</sup>
2 <sup>+</sup>	95-116	24	-18.3 ± 0.4 <sup>a</sup>	7.6 ± 0.3 <sup>a</sup>
3 <sup>+</sup>	108-125	24	-18.3 ± 0.4 <sup>a</sup>	8.1 ± 0.2 <sup>b</sup>
4 <sup>+</sup>	116-127	22	-18.2 ± 0.4 <sup>a</sup>	8.3 ± 0.1 <sup>b</sup>

Column means followed by a different letter superscript are significantly different at  $P < 0.05$

Table 3  
 Mean trophic position of scallops grouped by age

Age class (in Years)	<i>N</i>	Mean $\pm$ SD	Range
1 <sup>+</sup>	<b>13</b>	2.5 $\pm$ 0.3	2.1–3.2
2 <sup>+</sup>	24	2.3 $\pm$ 0.1	2.1–2.4
3 <sup>+</sup>	24	2.5 $\pm$ 0.2	2.2–2.9
4 <sup>+</sup>	22	2.7 $\pm$ 0.2	2.4–3.0

Table 4

Percentage  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  contributions of micro- (100–330  $\mu\text{m}$ ), meso-zooplankton (>330  $\mu\text{m}$ ) and suspended particulate organic matter (SPOM) to scallops' diet in different age classes. Values represent mean (range).

Age class (in Years)	Food sources		
	SPOM	Zooplankton	
		100–330 $\mu\text{m}$	>330 $\mu\text{m}$
1 <sup>+</sup>	68 (65-70)	18 (0-35)	15 (0-30)
2 <sup>+</sup>	46 (41-50)	30 (0-59)	25 (0-50)
3 <sup>+</sup>	19 (12-25)	44 (0-88)	38 (0-75)
4 <sup>+</sup>	8 (0-15)	50 (0-100)	43 (0-85)

Table S1

Summary table showing the mean  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  (‰) values of suspended particulate organic matter (SPOM) sampled from 2007 to 2009 at Tokoro seabed, Okhotsk Sea

Date	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
2007 ( $n = 18$ )	$-24.7 \pm 0.8$	$5.4 \pm 1.7$
2008 ( $n = 24$ )	$-24.0 \pm 0.9$	$4.4 \pm 1.3$
2009 ( $n = 12$ )	$-22.7 \pm 0.5$	$6.0 \pm 1.0$
Mean	-23.8	5.3
SD	1.0	0.8

Table S2

Summary table showing the tissue-diet isotopic fractionation values ( $\Delta\delta^{13}\text{C}$  and  $\Delta\delta^{15}\text{N}$ ) among ages of *Mizuhopecten yessoensis* grown at Tokoro seabed, Okhotsk Sea. The difference between isotopic ratios of gut content and that of muscle tissue was used as representation of tissue-diet isotopic fractionation.

Age (in Years)	$\Delta\delta^{13}\text{C}$	$\Delta\delta^{15}\text{N}$
1 <sup>+</sup> ( <i>n</i> = 12)	3.3 ± 0.7	1.6 ± 1.0
2 <sup>+</sup> ( <i>n</i> = 12)	3.3 ± 0.9	1.3 ± 0.8
3 <sup>+</sup> ( <i>n</i> = 12)	3.4 ± 0.7	1.6 ± 1.3
4 <sup>+</sup> ( <i>n</i> = 12)	3.7 ± 0.6	2.3 ± 1.2
Mean	3.4	1.7
SD	0.2	0.4

Table S3  
 Mean  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  (‰) values of muscle tissues from different cohorts of *Mizuhopecten yessoensis* grown at Tokoro seabed, Okhotsk Sea

Cohorts	Area	Date (mo/yr)	Recalculated Age (in years)	Shell Height (mm)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
2007	B	10/2008	1.42	74.94	$-18.3 \pm 0.0$	$7.2 \pm 0.1$
		11/2008	1.47	78.56	$-18.2 \pm 0.0$	$7.2 \pm 0.3$
		12/2008	1.53	76.20	$-18.3 \pm 0.1$	$7.5 \pm 0.2$
		5/2009	1.95	89.20	$-18.8 \pm 0.1$	$7.3 \pm 0.1$
2006	C	10/2007	1.40	80.30	$-18.2 \pm 0.4$	$7.0 \pm 0.3$
		11/2007	1.49	82.60	$-18.0 \pm 0.1$	$6.9 \pm 0.2$
		4/2008	1.88	94.66	$-18.1 \pm 0.2$	$7.4 \pm 0.2$
		5/2008	1.95	93.81	$-17.7 \pm 0.0$	$7.4 \pm 0.3$
		6/2008	2.06	98.10	$-17.8 \pm 0.1$	$7.7 \pm 0.2$
		7/2008	2.15	103.54	$-18.1 \pm 0.1$	$7.5 \pm 0.1$
		8/2008	2.21	101.39	$-17.9 \pm 0.3$	$7.7 \pm 0.2$
		9/2008	2.28	106.38	$-18.0 \pm 0.3$	$7.6 \pm 0.1$
		10/2008	2.42	108.12	$-17.9 \pm 0.3$	$7.2 \pm 0.3$
		11/2008	2.47	112.02	$-17.6 \pm 0.1$	$7.2 \pm 0.0$
		12/2008	2.53	108.86	$-17.6 \pm 0.1$	$7.4 \pm 0.2$
		5/2009	2.95	115.60	$-18.5 \pm 0.2$	$7.1 \pm 0.1$
		2005	A	10/2006	1.38	77.20
11/2006	1.50			86.60	$-18.0 \pm 0.1$	$7.2 \pm 0.1$
12/2006	1.55			83.60	$-18.1 \pm 0.2$	$7.0 \pm 0.1$
4/2007	1.88			87.25	$-17.7 \pm 0.0$	$7.6 \pm 0.3$
5/2007	1.95			90.75	$-17.8 \pm 0.1$	$7.5 \pm 0.2$
6/2007	2.03			101.25	$-18.1 \pm 0.1$	$7.5 \pm 0.1$
7/2007	2.13			102.00	$-17.9 \pm 0.3$	$7.5 \pm 0.1$
8/2007	2.19			100.75	$-18.0 \pm 0.3$	$7.4 \pm 0.1$
9/2007	2.27			107.50	$-17.9 \pm 0.3$	$7.5 \pm 0.2$
10/2007	2.40			109.50	$-17.6 \pm 0.1$	$7.6 \pm 0.1$
11/2007	2.49			108.38	$-17.6 \pm 0.1$	$7.4 \pm 0.3$
12/2008	2.88			114.46	$-18.5 \pm 0.2$	$7.7 \pm 0.2$
5/2008	2.95			112.21	$-18.2 \pm 0.4$	$7.5 \pm 0.2$
6/2008	3.06			118.77	$-18.0 \pm 0.1$	$8.0 \pm 0.2$
7/2008	3.15			121.94	$-18.1 \pm 0.2$	$8.0 \pm 0.1$
8/2008	3.21			117.89	$-17.7 \pm 0.0$	$8.0 \pm 0.0$
9/2008	3.28			118.87	$-17.8 \pm 0.1$	$7.9 \pm 0.2$
10/2008	3.42			123.39	$-18.1 \pm 0.1$	$7.7 \pm 0.4$
11/2008	3.47			121.34	$-17.9 \pm 0.3$	$7.6 \pm 0.1$
12/2008	3.53			121.55	$-18.0 \pm 0.3$	$8.3 \pm 0.1$
5/2009	3.95	124.58	$-17.9 \pm 0.3$	$7.5 \pm 0.1$		
2004	D	7/2006	2.11	95.20	$-19.3 \pm 0.0$	$7.7 \pm 0.3$
		8/2006	2.19	96.60	$-18.9 \pm 0.0$	$7.9 \pm 0.2$
		9/2006	2.27	99.20	$-18.8 \pm 0.1$	$7.8 \pm 0.4$
		10/2006	2.38	96.90	$-18.6 \pm 0.3$	$7.9 \pm 0.1$
		11/2006	2.50	103.80	$-18.2 \pm 0.4$	$8.0 \pm 0.2$

Cohorts	Area	Sampling Month	Recalculated Age (in years)	Shell Height (mm)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)
2004	D	12/2006	2.55	105.10	$-18.3 \pm 0.1$	$7.9 \pm 0.0$
		4/2007	2.88	111.00	$-18.1 \pm 0.2$	$7.8 \pm 0.2$
		5/2007	2.95	110.00	$-18.8 \pm 0.2$	$7.9 \pm 0.2$
		6/2007	3.03	112.00	$-18.9 \pm 0.2$	$7.4 \pm 0.3$
		7/2007	3.13	115.75	$-18.7 \pm 0.3$	$8.0 \pm 0.2$
		8/2007	3.19	117.75	$-18.9 \pm 0.1$	$7.9 \pm 0.2$
		9/2007	3.27	122.25	$-18.6 \pm 0.1$	$8.2 \pm 0.1$
		10/2007	3.40	119.50	$-18.7 \pm 0.2$	$7.9 \pm 0.1$
		11/2007	3.49	118.13	$-18.4 \pm 0.2$	$8.1 \pm 0.1$
		4/2008	3.88	123.74	$-18.1 \pm 0.0$	$8.1 \pm 0.1$
		5/2008	3.95	123.16	$-17.8 \pm 0.1$	$8.5 \pm 0.1$
		6/2008	4.06	121.92	$-17.7 \pm 0.1$	$8.4 \pm 0.2$
		7/2008	4.15	124.32	$-18.1 \pm 0.1$	$8.4 \pm 0.4$
		8/2008	4.21	122.80	$-18.0 \pm 0.3$	$8.3 \pm 0.2$
		9/2008	4.28	125.18	$-17.9 \pm 0.0$	$8.3 \pm 0.1$
		10/2008	4.42	125.07	$-17.7 \pm 0.1$	$8.1 \pm 0.3$
11/2008	4.47	125.85	$-17.8 \pm 0.1$	$7.9 \pm 0.4$		
12/2008	4.53	127.36	$-17.6 \pm 0.4$	$8.0 \pm 0.4$		
2003	B	7/2006	3.11	108.40	$-18.5 \pm 0.4$	$8.2 \pm 0.2$
		8/2006	3.19	108.80	$-19.0 \pm 0.1$	$8.2 \pm 0.2$
		9/2006	3.27	106.80	$-18.7 \pm 0.0$	$8.4 \pm 0.1$
		10/2006	3.38	110.60	$-18.7 \pm 0.1$	$8.3 \pm 0.3$
		11/2006	3.50	109.60	$-18.6 \pm 0.3$	$8.1 \pm 0.2$
		12/2006	3.55	112.90	$-18.3 \pm 0.1$	$8.2 \pm 0.1$
		4/2007	3.88	115.50	$-18.0 \pm 0.1$	$8.2 \pm 0.3$
		5/2007	3.95	107.00	$-18.4 \pm 0.0$	$8.0 \pm 0.3$
		6/2007	4.03	116.25	$-18.9 \pm 0.5$	$8.2 \pm 0.3$
		7/2007	4.13	124.50	$-18.5 \pm 0.2$	$8.3 \pm 0.1$
		8/2007	4.19	119.25	$-19.1 \pm 0.2$	$8.3 \pm 0.1$
		9/2007	4.27	118.75	$-18.2 \pm 0.2$	$8.3 \pm 0.0$
		10/2007	4.40	116.75	$-18.5 \pm 0.1$	$8.2 \pm 0.1$
		11/2007	4.49	115.00	$-18.3 \pm 0.1$	$8.4 \pm 0.1$
		4/2008	4.88	124.44	$-18.0 \pm 0.1$	$8.7 \pm 0.2$
		5/2008	4.95	121.75	$-17.6 \pm 0.1$	$8.5 \pm 0.2$
2002	C	7/2006	4.11	119.40	$-18.6 \pm 0.0$	$8.1 \pm 0.1$
		8/2006	4.19	125.40	$-18.3 \pm 0.5$	$8.3 \pm 0.5$
		9/2006	4.27	120.40	$-18.3 \pm 0.0$	$8.4 \pm 0.2$
		10/2006	4.38	119.20	$-18.0 \pm 0.2$	$8.6 \pm 0.5$
		11/2006	4.50	113.20	$-18.3 \pm 0.3$	$8.4 \pm 0.1$
		12/2006	4.55	118.40	$-18.6 \pm 0.1$	$8.3 \pm 0.3$
		5/2007	4.95	124.00	$-18.5 \pm 0.2$	$8.3 \pm 0.2$