CM 1998/M11 <Not to be cited without prior reference to the author>

MIGRATORY ROUTES OF THE JAPANESE COMMON SQUID (*Todarodes pacificus*) INFERRED FROM ANALYSES OF STATOLITH TRACE ELEMENTS, AND NITROGEN AND CARBON STABLE ISOTOPES

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Summary: The Sea of Japan is divided by the Subarctic Front into two distinct regions: the Subarctic and the Tsushima current. Japanese common squid (*Todarodes pacificus*) that inhabit these two regions may form distinct groups. Squid from both regions are similar in age, but have different growth patterns. Analyses of statolith trace elements, and nitrogen ($\delta^{15}N$) and carbon ($\delta^{13}C$) stable isotopes in muscle tissue can give information on the environmental history of marine organisms. We conducted these analyses on squid from both regions. $\delta^{13}C$ amounts significantly differed between the Subarctic (-18.4±0.3‰) and the Tsushima (-18.8±0.5‰) groups, although $\delta^{15}N$ amounts were similar, suggesting that in their northern migration, these groups are geographically distinct but feed at same trophic level. Strontium-calcium ratios (Sr/Ca ratios) at the margin of the statoliths (corresponding with the phase at 220-250 days old), were similar (ca 0.017) in both groups. A large range in Sr/Ca ratios (0.017-0.024) occurred at the phase from 70 to 140 days old. Sr/Ca ratios were high (0.019-0.021) at hatching and decreased to ca 0.015 at 10 days old in both groups.

Keywords: Japanese common squid, $\delta^{15}N$, $\delta^{13}C$, statolith, Sr/Ca ratios, Sea of Japan

Purpose: The Japanese common squid (*Todarodes pacificus*) migrates around the Japan Islands to the extent between the East China Sea and the Okhotsk Sea within one-year life span (Okutani, 1983). The Sea of Japan, a main habit of *T. pacificus*, is divided by the Subarctic Front into two distinct regions: the Subarctic and the Tsushima current. *T. pacificus* that inhabit these two regions may form distinct groups, *i.e.*, squid from both regions are similar in age, but different growth patterns (Kidokoro & Hiyama, 1996) and they feed different kind of diets (Okiyama, 1965). In order to obtain fundamental data for migratory route of *T. pacificus*, we conducted the analyses of statolith trace elements, and nitrogen ($\delta^{15}N$) and carbon ($\delta^{13}C$) stable isotopes in muscle tissue that can give information on the environmental history of marine organisms (Smith *et al.*, 1979; Minami *et al.*, 1995) on squid from both regions.

Materials and Methods: Total number of seventy sub-adult *T. pacificus* (DML, 185-226mm, 35 males and 35 females) were collected between 24 June 1997 and 14 July 1997 from the Subarctic, the Tsushima current, and the Subarctic Front (Table 1). Carbon and nitrogen isotope ratios of muscle tissue from these squid were analyzed according to the methods described in Minami *et al.* (1995) using by Delta-S mass spectrometers. Isotope ratios were expressed as ‰ deviations from standards as defined by the following equation:

$$\delta^{13}C,\ \delta^{15}N=(R_{sample}/R_{standard}-1)\cdot 1000(‰)$$

where $R={}^{13}C/{}^{12}C$ or ${}^{15}N/{}^{14}N$. Peedee belemnite and atmospheric nitrogen were used as the carbon and nitrogen isotope standards, respectively. $\delta^{13}C$ and $\delta^{15}N$ of crustaceans extracted from the stomach contents of squid from the Subarctic, and mesopelagic fish *Maurolicus muelleri japonica* collected from the Tsushima current, which are assumed as main diet of squid at the Subarctic and the Tsushima current (Okiyama, 1965), respectively, were also measured.

Statoliths of squid from those three regions were extracted for the trace elements analyses. Statoliths were mounted in polyester resin and ground and polished on the anterior side to the nucleus and margin of the statoliths. Calcium (Ca) and strontium (Sr) which are the elements as thermoindicater in the marine organism hard tissue, *e.g.*,

TABLE 1. Collection date, location of *Todarodes pacificus* (Stations A-G), *Maurolicus muelleri japonica* (St. H), and crustaceans (St. I) and 50m depth temperature at each location.

Station	Region	Collection date	Location	50m depth temperature (C°)
A	Tsushima current	24 June 1997	38°42'N: 138°25'E	13.2
B	Tsushima current	11 July 1997	39°40'N: 139°00'E	13.5
C	Tsushima current	13 July 1997	38°10'N: 138°00'E	16.0
D	Subarctic Front	25 June 1997	40°00'N: 137°00'E	8.9
E	Subarctic	26 June 1997	40°40'N: 135°00'E	2.1
F	Subarctic	27 June 1997	40°40'N: 134°00'E	3.6
G	Subarctic	30 June 1997	42°00'N: 135°40'E	1.8
H	Tsushima current	26 May 1997	37°19'N: 137°55'E	12.9
I	Subarctic	2 July 1997	42°40'N: 136°50'E	4.2

coral skeleton (Smith *et al.*, 1979) were measured for the ground statolith surface using by a wavelength dispersive spectrometer (EPMA-C1, SHIMADZU). Conditions for the measurement were as follows: an acceleration voltage of 15 kV, beam current 10 nA and a 5 µm diameter focused beam. $CaSiO_3$ and $SrTiO_3$ were used as standards for Ca and Sr calibration, respectively. Total of twenty points from nucleus to the margin of each statolith were measured for the amounts of Sr and Ca with equal interval. After the elements measurement, number of daily microincrements (Nakamura & Sakurai, 1991) on the bomber marks were counted for each statolith.

Results and Discussions: Figure 2 shows $\delta^{13}C$ and $\delta^{15}N$ amounts of *T. pacificus*, crustaceans (species not identified) and *Maurolicus muelleri japonica*. Although $\delta^{15}N$ amounts of crustaceans (7.5±0.1‰) and *Maurolicus muelleri japonica* (8.2±0.2‰) differed, those amounts of squid from the Subarctic (10.5±0.5‰), the Tsushima current (10.4±0.3‰), and the Subarctic Front (10.6±0.3‰) were not significantly different. On the other hand, $\delta^{13}C$ amounts of squid from the Subarctic (-18.8±0.5‰) and the Tsushima current (-18.4±0.3‰) were significantly different (Kruskal-Wallis test, $p<0.005$) (the Subarctic Front, -18.7±0.3‰). Those amounts of crustaceans (-20.6±0.1 ‰) and *Maurolicus muelleri japonica* (-19.2±0.2‰) also differed with ca 1.4‰. These facts suggest that squid from the Subarctic and the Tsushima current geographically distinct but feed at same trophic level, probably feed crustaceans since $\delta^{15}N$ amounts of squid are higher than crustaceans with ca 3‰ ($\delta^{15}N$ enriches for 3‰ with one trophic level). This situation is comparative with the previous report, based on squid stomach contents, for the diet of *T. pacificus* from these regions that the Subarctic and the Tsushima groups mainly feed crustaceans and *Maurolicus muelleri japonica*, respectively (Okiyama, 1965).

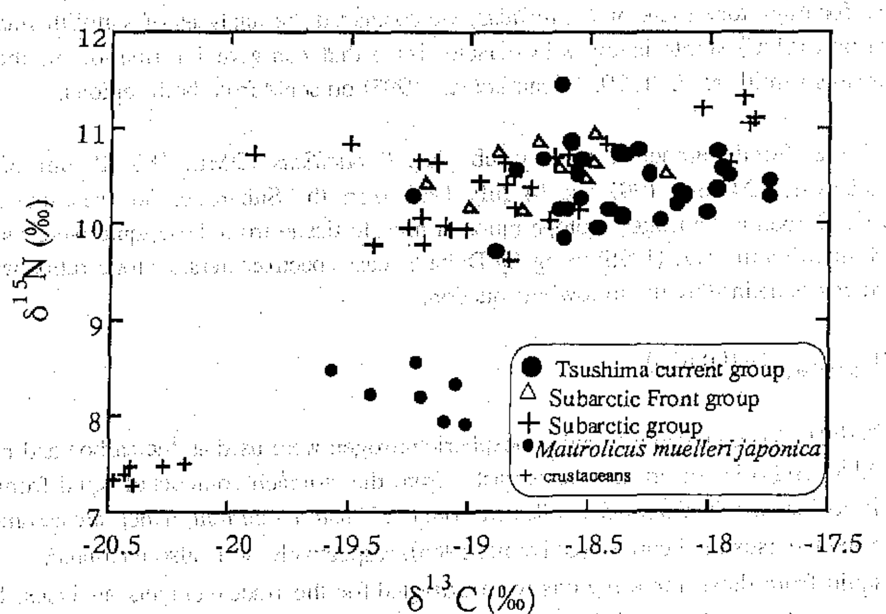

FIGURE 2. $\delta^{13}C$ and $\delta^{15}N$ amounts of *Todarodes pacificus*, crustaceans and *Maurolicus muelleri japonica* from the Sea of Japan.

Figure 3 shows age versus the variation of Sr/Ca ratios in statoliths of *T. pacificus* from the Subarctic, the Tsushima current, and the Subarctic Front. Sr/Ca ratios at the margin of the statoliths (corresponding with the phase at 220-250 days old), were similar (ca 0.017) in three groups. A large range in Sr/Ca ratios (0.017-0.024) occurred at the phase from 70 to 140 days old. Sr/Ca ratios were high (0.019-0.021) at hatching and decreased to ca 0.015 at 10 days old in both groups. The variation did not have any particular relationships with three groups.

Although the main factor affecting the deposition of trace elements in the statoliths is not clear so far (Ikeda *et al.*, 1998; Yatsu *et al.*, 1998), a hypothesis on migratory routes of *T. pacificus* can be made based on the present observations (Fig. 4). On their northern migration, squid forms geographically distinct groups around the Subarctic Front, namely the Subarctic and the Tsushima groups. Before reaching these regions, squids experienced similar migratory routes since the profile of Sr/Ca ratios are almost similar in both groups from hatching to sub-adult (220-250 days old) except for 70 to 140 days old phase. Based on the idea that Sr/Ca ratios in hard tissue of marine organisms have negative relationship with temperature (Smith *et al.*, 1979), the high amounts of Sr/Ca ratios at hatching followed by radical decrease at 10 days old suggest that squid hatch at middle layer and hatchlings move to surface layer afterward. *T. pacificus* at young age (118 to154 days old) inhabit at Wakasa Bay (Kidokoro & Wada, 1997) and also the offshore water in the Sea of Japan (Murata, 1983). Thus, a large variation of Sr/Ca ratios during 70 to 140 days old tentatively suggests that young squids inhabit at various regions with different water temperature as a result of transportation by the Tsushima current.

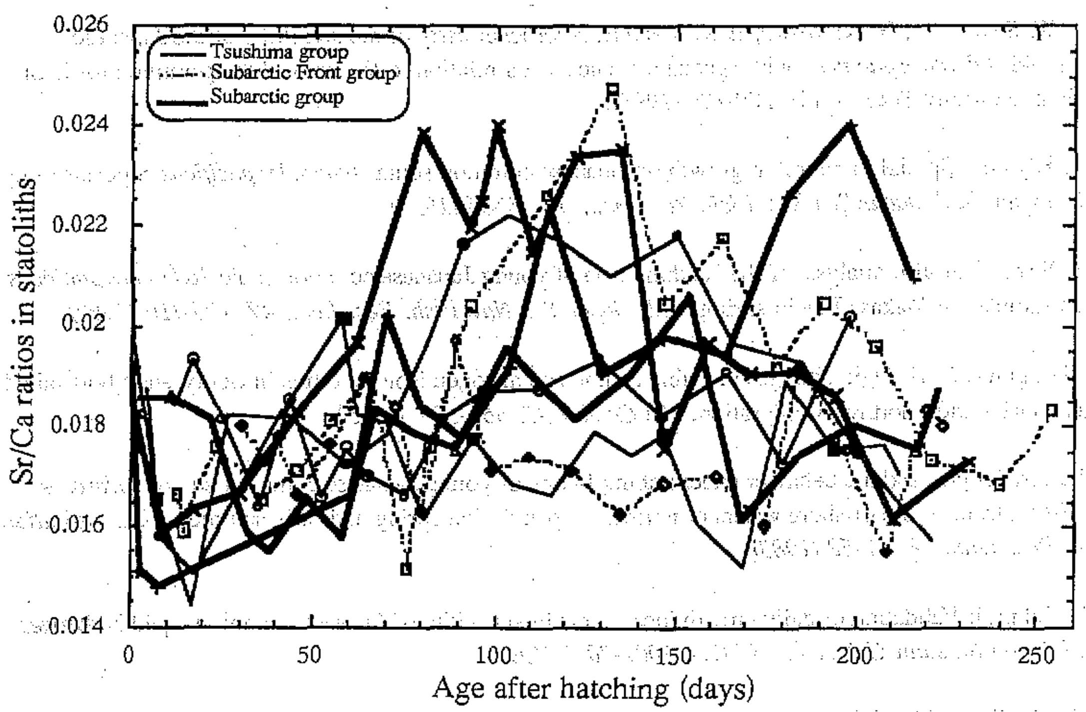

FIGURE 3. Transects of statolith Sr/Ca ratios of *Todarodes pacificus* (3 squid of the Tsushima current group, 2 squid of the Subarctic Front group, 3 squid of the Subarctic group) related to age after hatching.

Diet would be an another factor to affect a large variation of Sr/Ca ratios in young squids since $\delta^{13}C$ and $\delta^{15}N$ of young *T. pacificus* from Wakasa Bay significantly differ according to collection sites (Onaka, *unpublished data*), which indicates that this age of squids form geographically different groups and feed on different trophic level.

Conclusion: Carbon and nitrogen stable isotopes reveal that on their northern migration *Todarodes pacificus* form

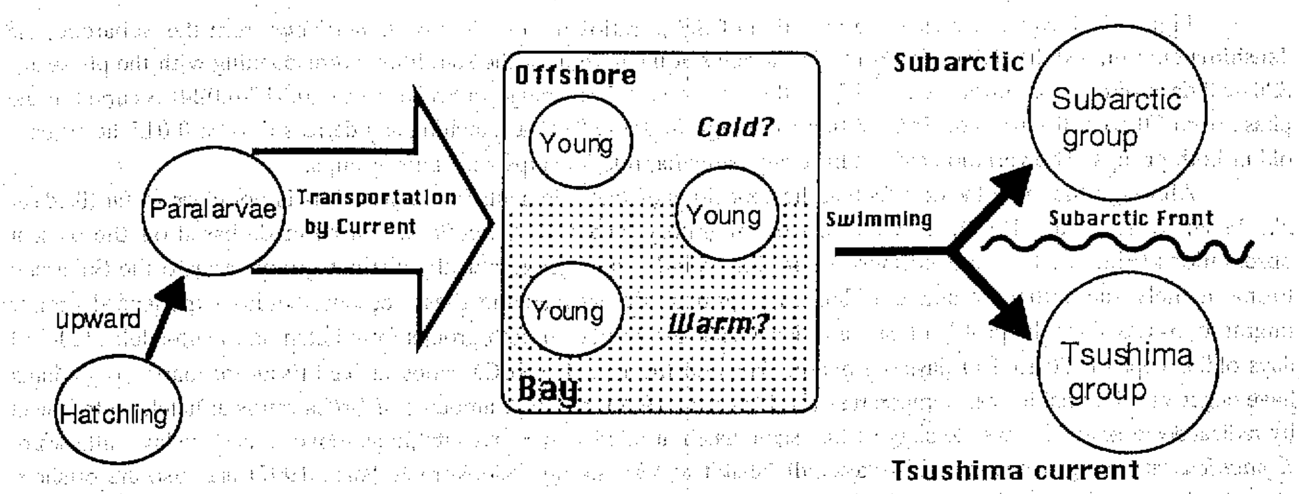

FIGURE 4. Hypothesis on migratory route of *T. pacificus*, which inferred by $\delta^{13}C$, $\delta^{15}N$, and statolith Sr/Ca ratios.

geographically distinct groups around the Subarctic Front in the Sea of Japan on summer, but they feed at same trophic level. Statolith Sr/Ca ratios suggest that *T. pacificus* in different groups have moved similar migratory route but at young age they have inhabited various locations with different temperature as a result to of transportation by the Tsushima current.

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