

Body Length, Stable Carbon, and Nitrogen Isotope Ratios and Mercury Levels in Common Minke Whales Stranded Along the Coast of Hokkaido, Japan

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Abstract

We quantified the stable isotope ratios of carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) as well as the total mercury (Hg) concentration in muscle samples from 20 common minke whales (*Balaenoptera acutorostrata*) stranded along the coast of Hokkaido, Japan. We investigated the relationship between the quantified values and the body length (BL) of the whales to examine the BL at which ontogenetic changes, such as nursing and weaning, occur. The whales investigated consisted of 12 calves (BL \leq 5 m), including newborns, and eight sexually mature whales (7.2 to 10.2 m BL). The $\delta^{15}\text{N}$ values in the calves were in the range of 11.2 to 13.8‰ and were fitted to a quadratic function ($F_{10} = 4.588$, $R^2 = 0.505$, $p = 0.0423$). The $\delta^{15}\text{N}$ -enriched peak was calculated to be 4.0 m BL, which may correspond with the onset of weaning. In contrast, the $\delta^{15}\text{N}$ values of the mature whales showed no particular change regardless of growth (10.8 to 12.4‰). The $\delta^{13}\text{C}$ values in the calves showed no particular pattern (-18.3 to -20.0‰), probably due to the large variation in the lipid concentration in milk, while those in the mature whales tended to decrease with growth ($F_6 = 3.400$, $R^2 = 0.362$, $p = 0.115$). The Hg concentrations in the muscle samples were trace but increased linearly with an increase in BL ($F_{18} = 87.68$, $R^2 = 0.830$, $p < 0.0001$). The $\delta^{15}\text{N}$ and Hg levels of mature whales were lower than respective levels observed in odontocetes stranded along the coast of Hokkaido, suggesting that mature whales feed mainly on zooplankton and small fish containing low Hg concentrations at low trophic positions.

Key Words: calf, nursing, weaning, ontogeny, milk, baleen whale, curve fitting

Introduction

Japan consists of four large islands, with Hokkaido being the northernmost (Figure 1). Hokkaido is surrounded by the North Pacific Ocean, the Sea of Japan, and the Sea of Okhotsk. Although more than 50 cetaceans are stranded annually along the coast of Hokkaido, most of the stranded cetaceans are odontocetes, with only a limited number of stranded mysticetes. For example, we previously investigated the contamination of ^{137}Cs in stranded cetaceans around Hokkaido in 2011 and 2012; the sampled whales included 41 individuals from nine odontocete species and eight individuals from two mysticete species (Nakamura et al., 2015).

According to the stranding records kept by the Stranding Network of Hokkaido (SNH), most stranded baleen whales in Hokkaido are common minke whales (*Balaenoptera acutorostrata*), although small numbers of humpback whales (*Megaptera novaeangliae*), North Pacific right whales (*Eubalaena japonica*), and fin whales (*Balaenoptera physalus*) also strand. Most of



Figure 1. Map of Hokkaido, Japan

the baleen whales that stranded in Hokkaido are calves or immature animals. Of the 10 common minke whales stranded in 2016, four were calves, one was an immature animal, three were mature animals, and the ages of two could not be estimated. Furthermore, two humpback whales, a North Pacific right whale, and a fin whale, all stranded in 2016, were calves or immature animals. These baleen whales may migrate annually from north to south, but detailed information regarding their migration routes and their calving and feeding grounds is unknown. The reason why many calves and immature whales tend to strand along the coast of Hokkaido is also unknown.

Common minke whales inhabiting the waters around Japan can be morphologically and genetically categorized into at least two types: (1) the “O-type,” found primarily offshore in the Pacific Ocean, and (2) the “J-type,” found primarily in the Sea of Japan and nearshore waters along Japan’s Pacific coast (Endo et al., 2003; Wade et al., 2010). Both types of whales are believed to inhabit the Sea of Okhotsk (Wade et al., 2010). Common minke whales are opportunistic and polyphagous feeders that change their prey temporally and regionally. They feed on zooplankton such as krill (*Euphausia pacifica*) and copepods, in addition to Pacific saury (*Cololabis saira*) and Japanese anchovy (*Engraulis japonicus*), and some feed on Pacific pomfret (*Brama japonica*) (Yasunaga & Fujise, 2009). The total mercury (Hg) concentrations in these fish are higher than those in zooplankton (Yasunaga & Fujise, 2009), and Hg is a typical contaminant accumulated in marine mammals in an age and/or body length (BL) dependent manner (Honda et al., 1983; Itano et al., 1984; Endo et al., 2007a, 2008). The Hg accumulation in the common minke whale should, therefore, be dependent on the Hg concentrations in their prey species and their age (BL) (Yasunaga & Fujise, 2009).

Japan stopped scientific whaling of baleen whales from the western North Pacific Ocean in June 2019 and resumed commercial whaling in waters around Japan. The common minke whale is one of the whale species caught in scientific and commercial whaling, and the red meat products are available from Japanese markets (Endo et al., 2012). The red meat products from stranded or incidentally caught common minke whales are also available from Korean markets (Baker et al., 2006; Endo et al., 2007b).

Stable isotope analysis is a useful tool for obtaining information on feeding ecology (reviewed by Kelly [2000] and O’Brien [2016]). The stable isotope ratio of nitrogen ($\delta^{15}\text{N}$) shows a stepwise increase with increases in the trophic level within a food chain, while the stable isotope ratio of

carbon ($\delta^{13}\text{C}$) is used to estimate the relative contribution to the diet of potential primary sources. Increases in the $\delta^{15}\text{N}$ value of 2 to 4‰ have been shown to occur between predators and their prey at each trophic level, usually varying among tissues and species (Caut et al., 2009; Borrell et al., 2012). Furthermore, these isotopic ratios are used to study the habitat preference such as pelagic vs benthic, and nearshore vs offshore vs estuarine (Newsome et al., 2010), as well as the geographical differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values due to latitude (Kelly, 2000; Popp et al., 2007; Endo et al., 2010, 2016; Bird et al., 2018).

Stable isotope ratios have been used extensively in the study of the diet, habitat, movement, and physiology of marine mammals (reviewed by Newsome et al., 2010). The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values are particularly useful in evaluating mother-to-offspring nutrient transfer as (1) lactating mothers catabolize their tissues to produce milk, which means the nursing offspring consumes its mother’s tissues and, thus, their trophic level is higher than their mothers (Kelly, 2000; Newsome et al., 2010; Borrell et al., 2016); and (2) milk contains high concentrations of ^{13}C -depleted lipids (Polischuk et al., 2001; Newsome et al., 2010; Borrell et al., 2016); therefore, nursing offspring should have enriched $\delta^{15}\text{N}$ values and $\delta^{13}\text{C}$ values either lower or similar to their mothers, depending on the milk lipid content and nursing period (Newsome et al., 2010).

Newsome et al. (2010) compared the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ profiles in the teeth of California sea lions (*Zalophus californianus*), killer whales (*Orcinus orca*), and sperm whales (*Physeter macrocephalus*) and suggested that not only the shift from milk to solid prey but also the length of the weaning period could affect the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ profiles—that is, a longer lactation period may result in gradual and complicated changes in those profiles, with $\delta^{15}\text{N}$ values possibly stabilizing and remaining relatively constant once animals are fully weaned. In general, odontocetes have much longer lactation periods, typically lasting 1 to 3 y, while that of baleen whales is only 5 to 6 mo (Oftedal, 1997; Newsome et al., 2010). Age and BL-related changes in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in the muscle have been previously investigated in bowhead whales (*Balaena mysticetus*; Lee et al., 2005) and fin whales (Borrell et al., 2012), both baleen species. However, neither study quantified the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in calf samples.

As mentioned above, common minke whales inhabiting the North Pacific Ocean are polyphagous feeders, and their calves have often been stranded along the coast around Hokkaido, Japan. To our knowledge, however, no detailed study of stable isotope profiles or Hg levels in calves

during lactation has yet been undertaken. In the present study, we quantified $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and Hg concentrations in muscle samples from common minke whales, including many calves, stranded along the coast of the Sea of Japan and the Sea of Okhotsk around Hokkaido, and investigated the ontogenetic changes in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values as well as Hg concentrations in muscle tissue. Next, we compared the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and Hg concentrations in the mature common minke whales with those in odontocetes stranded along the coast of Hokkaido. Finally, we compared the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and Hg concentrations in the muscle samples from the present study with those in the red meat products purchased from Japanese and Korean markets as reported previously (Endo et al., 2007b, 2012).

Methods

Muscle Samples of Common Minke Whales

Muscle samples were obtained from 20 common minke whales (Table 1) that were stranded along the coast of the Sea of Okhotsk (O) and the Sea of Japan (J) around Hokkaido, Japan, during the

period 2011 to 2018. Samples were analyzed for $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and Hg (Table 1). Most samples were obtained from SNH and from whales that showed relatively little sign of decomposition. However, one sample was collected from a decomposed whale for which sex could not be determined based on appearance.

Table 1 shows the sex (M/F), area of stranding (J/O), date (month/year), BL (m), $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values, and Hg concentrations in the muscle samples from the 20 minke whales. The 20 muscle samples were comprised of two calves at birth (neonates), 10 nursing calves, and eight mature animals. The measured BLs of 2.6 to 2.8 m and approximately 5 m correspond to those of calves at birth and at the cessation of weaning, respectively, and the BLs and ages at sexual maturity (6 to 7 y; Perrin et al., 2017) correspond to 6.3 m for males and 7.1 m for females, respectively (Omura & Sakiura, 1956; Kato, 1992). The largest and next largest mature animals at 10.2 m BL (female) and at 9.5 m BL (male) correspond nearly to the maximum BLs of females at 10.7 m and males at 9.8 m in common minke whales, respectively (Reeves et al., 2002).

Table 1. Stable isotope ratios of carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$), and mercury (Hg) concentrations in the muscle samples from common minke whales (*Balaenoptera acutorostrata*) stranded on the coast of the Sea of Japan (J) or the Sea of Okhotsk (O) in Hokkaido, Japan; UK = sex unknown.

Muscle sample	Sex	Area	Date of stranding	BL (m)	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Hg ($\mu\text{g}/\text{wet g}$)
1	F	J	July 2016	2.6	-19.1	11.2	0.03
2	UK	J	June 2019	3.0	-19.1	12.1	0.01
3	F	O	Oct 2013	3.9	-18.8	13.7	0.04
4	F	O	July 2015	4.1	-18.5	13.0	0.01
5	F	O	Oct 2012	4.1	-19.0	13.4	0.02
6	M	O	Nov 2016	4.4	-19.6	12.0	0.06
7	M	O	Nov 2018	4.5	-19.7	13.8	0.02
8	F	O	Nov 2016	4.6	-20.0	12.7	0.01
9	M	O	Nov 2011	4.7	-19.6	12.4	0.07
10	M	O	Nov 2012	4.8	-19.0	12.8	0.02
11	F	O	Oct 2015	4.9	-19.2	11.4	0.06
12	F	J	Dec 2012	5.0	-18.3	12.9	0.03
13	M	J	Jan 2015	7.2	-19.0	11.5	0.12
14	M	J	Jan 2018	7.4	-18.4	10.8	0.11
15	F	O	June 2017	7.6	-18.7	12.3	0.14
16	M	O	July 2018	7.7	-18.6	11.8	0.13
17	F	O	May 2018	7.7	-18.9	12.2	0.15
18	F	O	May 2017	7.9	-18.9	12.4	0.07
19	M	O	July 2014	9.5	-18.9	11.5	0.17
20	F	O	July 2011	10.2	-19.2	12.4	0.18

Chemical Analyses

Dried subsamples of the muscle were analyzed for the stable isotope ratios of ^{13}C and ^{15}N after the removal of lipids by chloroform/methanol extraction (Logan & Lutcavage, 2008). The extraction was repeated three times or more until the color of the extraction solvent became clear. The ^{13}C and ^{15}N analyses of the muscle samples were performed using an IRMS (Delta S, Finnigan MAT, Bremen, Germany, and EA1108, Fisons, Roano, Milan, Italy) as described previously (Ogasawara et al., 2018).

Isotope ratios are reported in the standard delta (δ) notation relative to the internal standard of Vienna Pee Dee Belemnite ($\delta^{13}\text{C}$) and atmospheric nitrogen ($\delta^{15}\text{N}$) based on the following equation:

$$\delta(\text{‰}) = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1,000$$

The $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were corrected by calibration curves using working standards of CERKU-1, -2, and -5, certified by Kyoto University and the Institute of Biogeosciences in Japan (Tayasu et al., 2011). Coefficient determinations (R^2) of both calibration curves were 0.999. The replicate errors were within 0.2 and 0.3% for the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values, respectively.

The total Hg concentrations in the muscle samples were quantified using a flameless atomic absorption spectrophotometer (HG-310; Hiranuma Sangyo Co. Ltd., Mito, Ibaraki, Japan). As reported previously (Endo et al., 2016), about 0.5 g of the subsample was digested by a mixture of HNO_3 , H_2SO_4 , and HClO_4 . DOLT-2 (National Research Council of Canada, Ottawa, ON) was used as an analytical quality control for Hg, and the recovery of Hg was $94 \pm 3\%$ ($n = 5$). The Hg concentrations in the muscle were expressed on a wet weight basis, and the determination limit of Hg was around $0.01 \mu\text{g}/\text{wet g}$. The Hg data shown in Table 1 represent the means of at least two measurements.

Statistical Analyses

The data were analyzed by Student's t test and Pearson's correlation coefficient test using the Statcell program (add-in software on *Excel*; OMS Co., Tokyo, Japan). As reported previously (Endo et al., 2017), we investigated whether the relationship between BL and $\delta^{13}\text{C}$ value, $\delta^{15}\text{N}$ value, or Hg concentration could be fitted by a linear, quadratic, or exponential function using *JMP*, Version 14.3 (SAS Institute Japan Ltd., Tokyo, Japan). The data were expressed as mean \pm SD.

Results

The $\delta^{15}\text{N}$ value for the calves was significantly higher than that for the mature animals ($12.6 \pm 0.8\text{‰}$ vs $11.9 \pm 0.6\text{‰}$; $t_{18} = 2.18$; $p = 0.0422$;

Table 1). Among the 12 calves, the highest and next highest $\delta^{15}\text{N}$ values (13.8 and 13.7‰) were found at BLs of 4.5 and 3.9 m, respectively, and the lowest $\delta^{15}\text{N}$ value (11.2‰) was found in a newborn animal with a BL of 2.6 m. These $\delta^{15}\text{N}$ values for the calves were fitted to a quadratic function ($F_{10} = 4.588$, $R^2 = 0.505$, $p = 0.0423$; Figure 2), and the $\delta^{15}\text{N}$ -enriched peak was calculated to be at 4.0 m BL and 13.3‰ . In comparison, the $\delta^{15}\text{N}$ values for the eight mature animals showed no particular change and were almost constant with BL ($11.9 \pm 0.6\text{‰}$).

The $\delta^{13}\text{C}$ value for the calves was similar to that for the mature animals ($-19.2 \pm 0.5\text{‰}$ vs $-18.8 \pm 0.2\text{‰}$; $t_{18} = 1.74$; $p = 0.0995$; Table 1). Variation in the $\delta^{13}\text{C}$ values for calves showed no particular pattern (from -18.3 to -20.0‰), while the $\delta^{13}\text{C}$ values for the mature animals tended to decrease with increases in BL ($F_6 = 3.400$, $R^2 = 0.362$, $p = 0.115$; Figure 2). A significant linear decrease was found when the smallest mature whale (7.2 m BL and -19.0‰) was removed ($F_5 = 7.879$, $R^2 = 0.612$, $p = 0.0377$).

Figure 3 shows the dual isotope plot of common minke whales investigated in this study (Table 1) and our previous study (Endo et al., 2012). The $\delta^{13}\text{C}$ values of O-type ($-18.6 \pm 0.8\text{‰}$) and J-type ($-18.4 \pm 0.7\text{‰}$) animals from the North Pacific Ocean were slightly higher than those of the mature whales ($-18.8 \pm 0.2\text{‰}$) and calves ($-19.2 \pm 0.5\text{‰}$) stranded along the coast of the Sea of Okhotsk and the Sea of Japan (Table 2).

There were only trace amounts in Hg concentration in the muscle of calves ($0.03 \pm 0.02 \mu\text{g}/\text{wet g}$, $n = 12$; Table 1), which was lower than that of mature animals ($0.133 \pm 0.035 \mu\text{g}/\text{wet g}$, $n = 8$) ($t_{18} = 8.15$, $p < 0.0001$). The Hg concentrations in those animals increased with increases in BL (Figure 2), which was well-fitted to a linear function ($F_{18} = 87.68$, $R^2 = 0.830$, $p < 0.0001$). The highest and next highest concentrations of Hg (0.18 and $0.17 \mu\text{g}/\text{wet g}$) were found in the largest and next largest mature animals at 10.2 m BL (female) and 9.5 m BL (male), respectively (Table 1).

Discussion

We quantified $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and Hg concentrations in muscle samples from common minke whales stranded along the coast of Hokkaido, Japan. We investigated ontogenetic changes and compared the present data with previous studies of common minke whales.

Ontogenetic Changes of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ Values in Muscle

We found a peak in the $\delta^{15}\text{N}$ values of common minke whale calves, which was estimated to occur at a BL of 4.0 m. The increase in $\delta^{15}\text{N}$ values

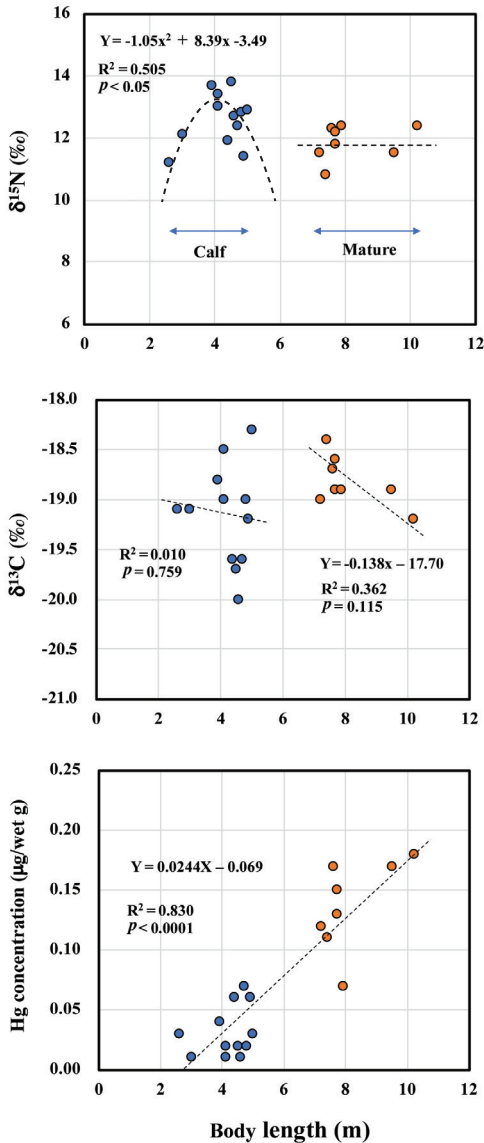


Figure 2. Relation between body length and $\delta^{15}\text{N}$ value, $\delta^{13}\text{C}$ value, or Hg concentrations in the muscle of common minke whales (*Balaenoptera acutorostrata*); calf (blue circles) and mature whale (orange circles).

across the peak may represent nursing, and the decrease in $\delta^{15}\text{N}$ values from the peak may represent weaning (i.e., a shift from milk to zooplankton and small fish at lower $\delta^{15}\text{N}$ values; Newsome et al., 2006; Vales et al., 2015). To our knowledge, we are the first to show a clear $\delta^{15}\text{N}$ -enriched peak in the calves of cetaceans and the estimated BL at the onset of weaning (4.0 m) in common

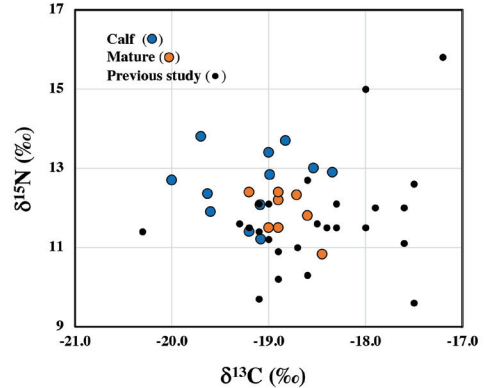


Figure 3. Relation between $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in the muscles of calves and mature common minke whales in the present study and values from the red meat products of common minke whales from a previous study (Endo et al., 2012)

minke whales. Our samples came from two newborn calves stranded in June and July and a larger group of older calves in November and December (Table 1), the timing of which may be consistent with the lactation period of 5 to 6 mo reported for common minke whales (Perrin et al., 2017).

We could not calculate the exact BL at the completion of weaning as our samples did not include juveniles. However, the weaned BL could be estimated to be 5.1 m from the quadratic equation of calves and the average $\delta^{15}\text{N}$ value of mature animals (11.9‰), which is in accordance with the weaned BL of approximately 5 m reported previously (Omura & Sakiura, 1956; Kato, 1992). We previously examined the relationship between BL and ontogenetic physiological changes in essential metals such as zinc, copper, and manganese in shark livers using quadratic curve fitting (Endo et al., 2017). Estimation of the peak position (BL and/or age) by curve fitting may be a useful tool with which to investigate ontogenetic physiological changes in animals.

While the fact that our estimated BL at weaning via the curve fitting is in agreement with previous morphological studies of minke whales (Omura & Sakiura, 1956; Kato, 1992) and lends support for this method, the time lag between the dietary shifts and changes in $\delta^{15}\text{N}$ values in muscle should be taken into consideration. To our knowledge, there is no information available in the literature regarding the metabolic turnover rate of calf whale muscle. The available general information indicates that the isotopic half-life ($T_{1/2}$) in mammalian muscle (1 to 3 mo) is markedly longer than that of metabolically active tissue in the mammalian liver

Table 2. Stable isotope ratios of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and Hg concentrations in the muscle (red meat) of common minke whales

	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Hg concentration ($\mu\text{g}/\text{wet g}$)	Area	Method	Source
Calf ($n = 12$)	-19.2 ± 0.5	12.6 ± 0.8	0.031 ± 0.024	Sea of Okhotsk and Sea of Japan	Stranding	This study
Mature ($n = 8$)	-18.8 ± 0.2	11.9 ± 0.6	0.133 ± 0.035			
J-type* ($n = 13$)	-18.4 ± 0.7	12.0 ± 0.7	0.091 ± 0.065	North Pacific Ocean	Japan's research whaling	Endo et al., 2012
O-type* ($n = 12$)	-18.6 ± 0.8	11.4 ± 0.7	0.099 ± 0.076			
J-type ?* ($n = 30$)			0.220 ± 0.110	Around South Korea	Bycatch	Endo et al., 2007
Mature ($n = 821$)			0.220 ± 0.070	Western North Pacific	Japan's research whaling	Yasunaga & Fujise, 2009

*No data concerning age or body length were available.

(3 to 7 d) (Boecklen et al., 2011; O'Brien, 2016), and $T_{1/2}$ decreases with a decrease in animal body mass and with increases in metabolic and growth rates (Trueman et al., 2005; Zanden et al., 2015). We believe the $T_{1/2}$ of the $\delta^{15}\text{N}$ value is shorter in calf muscle than in adult muscle (1 to 3 mo), and, thus, there is little time lag. Fortunately, decomposition of muscle tissue due to stranding appears not to affect the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values as muscle samples left unpreserved outdoors and exposed to rain and direct sunlight for 62 d did not show any alteration in those values (Payo-Paya et al., 2013).

The $\delta^{15}\text{N}$ values for the eight mature animals (< 7.1 m BL) were almost constant ($11.9 \pm 0.6\text{‰}$, $n = 8$), which would be expected for individuals that were weaned completely and fed mainly on zooplankton and small fish at a low trophic level. Similar $\delta^{15}\text{N}$ profiles were found in mature bowhead whales (Lee et al., 2005) and fin whales (Borrell et al., 2012). Thus, the $\delta^{15}\text{N}$ profile seen in our study ($\delta^{15}\text{N}$ -enriched peak in calves and constant of $\delta^{15}\text{N}$ values in mature animals) may be common among baleen whales which have short weaning periods and feed on solid prey at a low trophic level throughout their lives. In contrast, the changes in $\delta^{15}\text{N}$ values in the calves of odontocetes (Newsome et al., 2009; Jansen et al., 2012) and pinnipeds (Newsome et al., 2006; Riofrio-Lazo et al., 2012; Vales et al., 2015) which have long weaning periods are more gradual and complex, including $\delta^{15}\text{N}$ -depleted peaks. Reflecting differences in feeding habits, the average $\delta^{15}\text{N}$ value of mature common minke whales in this study ($11.9 \pm 0.6\text{‰}$, $n = 8$) is lower than that observed in the odontocete species stranded along the coast of Hokkaido such as Dall's porpoises (*Phocoenoides dalli*; $13.5 \pm 1.0\text{‰}$, $n = 45$), harbor porpoises (*Phocoena phocoena*; $13.1 \pm 1.0\text{‰}$, $n = 63$), and Pacific white-side dolphins (*Lagenorhynchus obliquidens*; $12.2 \pm 0.9\text{‰}$, $n = 34$) (Matsuda, 2017).

In contrast to the clear pattern in $\delta^{15}\text{N}$ values in calves, the $\delta^{13}\text{C}$ values in our study showed no particular pattern (-18.3 to -20.0‰). Milk of marine mammals contains a high concentration of ^{13}C -depleted lipids (Polischuk et al., 2001; Newsome et al., 2010; Borrell et al., 2016), and the lipid concentration in milk is highly variable. For instance, the lipid concentration in the milk from common minke whale ranges from 4.8 to 61.7%, and the lipid content decreases in the late stages of lactation (Lockyer, 1984). The absence of any particular pattern of change in the $\delta^{13}\text{C}$ values in the calves in our study could be ascribed to variations in the ^{13}C -depleted milk that the calves consumed. On the other hand, the $\delta^{13}\text{C}$ values for the mature animals tended to decrease with increases in BL, probably reflecting the prey-shift due to growth and/or the decrease in growth rate (Newsome et al., 2010).

Comparison of $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ Values with Previous Studies

Eight mature whales in this study were located within narrow ranges of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (Figure 3). In contrast, the 12 calves were located across wide ranges of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, which may reflect changes in $\delta^{15}\text{N}$ value due to nursing and weaning and the variations in $\delta^{13}\text{C}$ values in the milk. We believe that most of those whales are J-type according to the stranding area.

On the other hand, the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values in the common minke whales from the combined data of O- and J-types taken from the North Pacific Ocean (Endo et al., 2012) represented wider ranges in isotope values in comparison to the mature animals and calves in the present study. The $\delta^{13}\text{C}$ values of O-type ($-18.6 \pm 0.8\text{‰}$) and J-type ($-18.4 \pm 0.7\text{‰}$) animals from the North Pacific Ocean were also slightly higher than those of the mature whales ($-18.8 \pm 0.2\text{‰}$) and calves ($-19.2 \pm 0.5\text{‰}$) stranded along the coast of the Sea

of Okhotsk and the Sea of Japan (Table 2). Small but apparent geographical differences in $\delta^{13}\text{C}$ values were previously reported in Baird's beaked whales (*Berardius bairdii*; Endo et al., 2010) and Pacific cod (*Gadus macrocephalus*; Nakamura et al., 2018), which were taken around Hokkaido, such that values were highest in the North Pacific Ocean, followed by the Sea of Okhotsk, and lowest in the Sea of Japan. The lower $\delta^{13}\text{C}$ values found in the common minke whales stranded along the coast of the Sea of Okhotsk and the Sea of Japan compared to those taken from the North Pacific Ocean could be ascribed to geographical differences in $\delta^{13}\text{C}$ values.

Comparison of Hg Concentration with Previous Studies

The Hg concentration in the milk of cetaceans is trace—for instance, 0.002 ± 0.002 $\mu\text{g}/\text{wet g}$ for striped dolphins (Itano et al., 1984) and 0.22 ng/mL for franciscana dolphins (*Pontoporia blainvillei*; Caon et al., 2008). In our study, the Hg concentration in the calf muscle of the minke whale was markedly higher than that reported in the milk but significantly lower than that in the mature muscle.

Overall, the Hg level of common minke whales is markedly lower than that of odontocetes such as Dall's porpoises (0.880 ± 0.766 $\mu\text{g}/\text{wet g}$, $n = 56$), harbor porpoises (0.619 ± 0.451 $\mu\text{g}/\text{wet g}$, $n = 47$), and Pacific white-side dolphins (1.120 ± 0.613 $\mu\text{g}/\text{wet g}$, $n = 36$). Dall's porpoises, harbor porpoises, and Pacific white-sided dolphins stranded along the coast of Hokkaido (Endo, unpub. data), reflecting their lower trophic level.

According to Yasunaga & Fujise (2009), common minke whales feed on zooplankton (krill and copepods), Pacific saury, and Japanese anchovy, and some whales additionally feed on Pacific pomfret. The Hg concentrations in those species from the western North Pacific are below 0.05 $\mu\text{g}/\text{wet g}$, except for Pacific pomfret (0.232 ± 0.027 $\mu\text{g}/\text{wet g}$) (Table 3). As the average Hg concentration in the muscle samples from the mature

common minke whales in this study (0.133 ± 0.035 $\mu\text{g}/\text{wet g}$, $n = 8$) was apparently lower than that in Pacific pomfret (0.232 ± 0.027 $\mu\text{g}/\text{wet g}$), the common minke whales investigated in this study appear not to consume Pacific pomfret to a great degree. In addition, the $\delta^{15}\text{N}$ values of the mature whales in this study ($11.9 \pm 0.6\text{‰}$, $n = 8$) were similar to that of Pacific pomfret ($10.9 \pm 2.1\text{‰}$) and markedly higher than those of krill, Pacific saury, and Japanese anchovy (Table 3). These findings are further evidence that common minke whales investigated in this study do not feed to any great extent on Pacific pomfret.

The Hg concentration in red meat products of common minke whales from O- and J-types sold in Japan, which were taken from the North Pacific Ocean, were 0.099 ± 0.076 $\mu\text{g}/\text{wet g}$ and 0.091 ± 0.065 $\mu\text{g}/\text{wet g}$, respectively (Endo et al., 2003). On the other hand, the Hg concentration in red meat products of common minke whales sold in South Korea was 0.22 ± 0.11 $\mu\text{g}/\text{wet g}$. The Hg concentration in the muscle samples of common minke whales taken by Japan's scientific whaling program from the western North Pacific (7.46 ± 0.31 m BL, $n = 821$) was 0.22 ± 0.07 $\mu\text{g}/\text{wet g}$ (Yasunaga & Fujise, 2009), and that of stranded whales in this study (7.2 to 10.2 m BL) was 0.133 ± 0.035 $\mu\text{g}/\text{wet g}$. Such variations in Hg concentration may reflect the opportunistic and polyphagous feeding of common minke whales and the variations in the BL (age) of the animals analyzed, although the Hg level of common minke whales is markedly lower than that of odontocetes. The Hg concentrations in the muscle products originating from the common minke whale rarely exceed 0.4 $\mu\text{g}/\text{wet g}$, which is the Japanese permitted level of Hg for fish and shellfish, while most of those originating from odontocetes exceeds 1.0 $\mu\text{g}/\text{wet g}$ (Endo et al., 2003).

Conclusion

The $\delta^{15}\text{N}$ values for the calves, including newborns, were fitted to a quadratic function, and the $\delta^{15}\text{N}$ -enriched peak may be associated with

Table 3. Stable isotope ratios of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and Hg concentrations in the prey species of common minke whales

	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	Hg ($\mu\text{g}/\text{wet g}$)
Krill (<i>Euphausia pacifica</i>)	$-20.7 \pm 0.1^{\text{a}}$ (-19.7 ± 0.2) ^{a*}	$7.2 \pm 0.5^{\text{a}}$ (8.0 ± 0.2) ^{a*}	$0.001\text{--}0.013^{\text{b}}$
Pacific saury (<i>Cololabis saira</i>)	$-18.9 \pm 0.5^{\text{a}}$	$8.8 \pm 0.9^{\text{a}}$	$0.039 \pm 0.016^{\text{b}}$
Japanese anchovy (<i>Engraulis japonicus</i>)	$-19.6 \pm 0.6^{\text{a}}$	$9.3 \pm 1.4^{\text{a}}$	$0.037 \pm 0.025^{\text{b}}$
Pacific pomfret (<i>Brama japonica</i>)	$-19.2 \pm 0.8^{\text{c}}$	$10.9 \pm 2.1^{\text{c}}$	$0.232 \pm 0.027^{\text{b}}$

^aMitani et al. (2006); ^bYasunaga & Fujise (2009); and ^cCarlisle et al. (2012)

^{*}Sampled in June and September 1996 (parentheses); and [#]Hg concentrations in the combined samples of krill and copepods

nursing and weaning. The $\delta^{15}\text{N}$ value for the mature whales was almost constant despite their growth (11.9‰), and the Hg concentrations in their muscle samples were low (below 0.2 $\mu\text{g}/\text{wet g}$), a result of their feeding on zooplankton and small fish containing low Hg concentrations at a low trophic position. The $\delta^{13}\text{C}$ values for the calves showed no particular pattern, probably due to the large variations in the composition of their mothers' milk, while those for the mature whales tended to decrease. Most minke whales sampled for this study were stranded for unknown reasons; thus, it is likely that some or all of them were in poor health prior to stranding. It is possible that prolonged starvation due to stranding may have slightly increased $\delta^{15}\text{N}$ values and decreased $\delta^{13}\text{C}$ values (Petzke et al., 2010). Further study is needed to confirm the present results using biopsy samples or hunted whale samples.

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T. Endo contributed to the conception and design of this research and drafted the manuscript; O. Kimura and M. Terasaki contributed to the analysis and interpretation of the data; and M. Kobayashi collected the muscle samples from the stranded whales.

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