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High-Energy Foods Revealed by Hair Stable Isotope Analysis**

(ヒグマの採食生態：体毛安定同位体比分析で明らかとなった
高エネルギー食物の利用における地域・年齢・性別による差異)

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JIMBO Mina

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List of abbreviations

Chapter no.	Abbreviation	Meaning
2	BH-type bulb	Hair bulbs that appear black hooked shape. This indicates that the hairs are growing (i.e., anagen stage).
2	WH-type bulb	Hair bulbs that appear white hooked shape. This indicates that the hairs are almost stop growing (i.e., catagen stage).
2	WS-type bulb	Hair bulbs that appear white sphere shape. This indicates that the hairs are stop growing (i.e., telogen stage).
3	SNP	Shiretoko National Park.

Notes

Publications related to the dissertation

1. Jimbo M, Matsumoto N, Sakamoto H, Yanagawa Y, Torii Y, Yamanaka M, Ishinazaka T, Shirane Y, Sashika M, Tsubota T, and Shimozuru M. Hair growth in brown bears and its application to ecological studies on wild bears. *Mamm Study*, 45(4), 337-345, 2020.
2. Jimbo M, Ishinazaka T, Shirane Y, Umemura Y, Yamanaka M, Uno H, Sashika M, Tsubota T, Shimozuru M. Diet selection and asocial learning: Natal habitat influence on lifelong foraging strategies in solitary large mammals. *Ecosphere*, in press.

The contents of Chapter 2 have been published as Publication 1, and the contents of Chapter 3 have been published as Publication 2.

Chapter 1 | General introduction

Intraspecific differences in feeding habits

Intraspecific differences in feeding habits have been reported in a wide range of animal species. In many cases, species thought to be dietary generalists actually contain a variety of individuals with different foraging niches (e.g., Bolnick et al. 2007). Understanding this process of niche differentiation is ecologically important because large interindividual variation in diet can have profound effects on reproductive success, population dynamics, and food web dynamics (e.g., Clutton-Brock 1990, Bolnick et al. 2007).

Interindividual variation in the diet of a population can be divided into variation due to external factors such as habitat environment and food resources, variation due to internal factors such as sex and age, and variation that cannot be explained by either set of factors. In mammals, pregnant and lactating females generally have increased nutritional requirements; thus, they generally forage for longer periods of time and consume higher-quality foods. Sex differences are also found in macronutrient requirements. Males require sugars and fats to keep up with the energetic demands of their activities, whereas females require more protein to maintain their body for reproduction. Social interactions can also influence food consumption occur when food resources are spatially concentrated or limited in quantity, and in the presence of a social dominance hierarchy.

Feeding habits of brown bears

Brown bears (*Ursus arctos*) are large mammals that are widely distributed throughout northern North America, Europe, and Asia. They are generalist omnivores whose diets are most influenced by environmental conditions that relate to the spatial distribution of available food resources (Bojarska and Selva 2012, Coogan et al. 2018). Bears consume a wide range of foods, from plant matter such as grasses, fruits, and berries to animal matter such as insects, ungulates, and salmonids (e.g., Bojarska and Selva 2012, Shirane et al. 2021). In particular, salmonids are an important food item for the survival and reproductive success of bears inhabiting areas where they are abundantly available (Fig. 1-1) (e.g., Hilderbrand et al. 1999, Belant et al. 2006, Shirane et al. 2021). In areas where salmonids are not available, pine nuts, berries, and hard masts are important foods for bear survival and reproductive success (Schwartz et al. 2006, Costello et al. 2016, Hertel et al. 2018). Ungulates are also an energy-rich foods

for bears, but they are more incidental (Bastille-Rousseau et al. 2011).

While food abundance is the main factor underlying consumption, it has been suggested that interindividual variation in bear foraging behavior, rather than annual change in food abundance, has a greater impact on the feeding habits of individuals in resource-rich habitats (Deacy et al. 2018). A number of studies on brown bear feeding habits have reported interindividual variation in the consumption of energy-rich foods such as salmonids; older males consume more salmonids than females and young bears (Felicetti et al. 2004, Mowat and Heard 2006, Adams et al. 2017, Mangipane et al. 2020). Some females with cubs tend to avoid the consumption of salmon due to the high risk of encountering males or other individuals in these areas, but this strategy results in poor body condition and reduces the survival of both cubs and mothers (Ben-David et al. 2004).

Understanding individual variations and influencing factors in bear feeding habits is also important in the context of human-bear conflicts worldwide. Although human-bear conflicts vary, most are thought to be related to bear foraging behavior (e.g., Elfström et al. 2012, Artelle et al. 2016). For example, bears learn to use anthropogenic foods, such as agricultural crops, either from their mothers or incidentally after independence (e.g., Hopkins 2013, Morehouse et al. 2016). Because anthropogenic foods are easily obtained and highly nutritious, bears who have learned to consume these foods are likely to become problematic individuals who repeatedly intrude into human areas. Even if they do not consume anthropogenic foods, human-habituated bears have been proposed to utilize the presence of humans as a barrier to avoid food competition with other bears and to acquire high-energy foods (Elfström et al. 2012). By examining the dietary records of individual bears, we can know their dependency on anthropogenic foods and foraging strategies and thus determine which bears are more likely to intrude into human areas.

Stable isotope analysis

Analysis of stable isotope ratios is gaining popularity among researchers as a tool for investigating the individual-level ecology of wildlife, such as migration and foraging, as direct observation or tracking of wildlife is often difficult (e.g., Hobson 1999, Hopkins and Ferguson 2012). The stable isotope approach is based on the fact that the stable isotope composition in biological tissues varies depending on the diet, habitat, and trophic level of the organism (DeNiro and Epstein 1978, Hobson 1999). Carbon stable isotope ratios ($\delta^{13}\text{C}$) are used to distinguish whether the carbon source originated from the C_3 food chain, the C_4 food chain, or the marine food chain (e.g., Teeri and Schoeller 1979, Schoeninger and DeNiro 1984,

Hobson 1987), and nitrogen stable isotope ratios ($\delta^{15}\text{N}$) are used to estimate the trophic level (e.g., Peterson and Fry 1987, Hobson and Welch 1992). Sulfur stable isotope ratios ($\delta^{34}\text{S}$) are useful for distinguishing between marine- and terrestrial-derived diets (Felicetti et al. 2003), and for estimating the consumption of whitebark pine nuts by Yellowstone grizzly bears (e.g., Felicetti et al. 2003, Hopkins et al. 2017). Dietary analysis using the stable isotope technique quantifies the stable isotope ratios of prey assimilated into the predator's tissues, therefore allowing the calculation of the contribution, including digestion and metabolism, of specific prey items to the diet.

Stable isotope analysis of bear hair has become an increasingly popular method for studying bear feeding ecology (e.g., Felicetti et al. 2003, Hopkins et al. 2017, Adams et al. 2017, Careddu et al. 2021). Dietary analysis using stable isotope ratios on predator tissues has advantages over fecal analysis or direct observation as it provides the dietary history of an individual. The predator tissues used for stable isotope analysis include muscle, bone, blood, hair, and liver, and each tissue reflects the biological status of the animal at different time scales, depending on metabolic turnover. Generally, in mammals, isotopic turnover rates are ranked (from shortest to longest turnover) as follows: liver > muscle > hair > bone collagen (e.g., Tieszen et al. 1983, Lee-Thorp 2002, Ayliffe et al. 2004, Sponheimer et al. 2006). The use of hair for these analyses is preferable because the collection of hair is less invasive, and does not require capturing or handling the animal (Cook 2012, Kersey and Dehnhard 2014). Bear hairs can be efficiently collected from devices such as barbed-wire snares, and therefore large sample sizes can be obtained relatively easily. Another benefit of hair is that it provides biological information over a relatively long time scale (of weeks to months), as it accumulates biomarkers throughout its growth. In addition, individual information such as sex and age (maturity stage) can be determined at the same time by combining techniques such as genetic analysis, age assessment, and direct observation.

Aim of this study

The purpose of this study was to understand the feeding ecology of brown bears inhabiting the Shiretoko Peninsula, eastern Hokkaido, Japan, by applying hair stable isotope ratio analysis. In Chapter 2, I clarified the growth period of bear hairs, which is essential for correctly interpreting the results of the stable isotope analysis on whole hairs and for distinguishing the growth stages of hairs. In Chapter 3, I examined regional, sex, and age differences in the diet of wild brown bears captured on the Shiretoko Peninsula. In particular, I focused on the role of asocial learning in the use of high-energy foods (salmons

and pine nuts). In Chapter 4, I investigated the triggers of human-bear conflicts according to the regional characteristics of the diet of female bears and the context of the conflict. I attempted to distinguish the dietary characteristics of bears that intrude into human areas by comparing the diets of bears that were captured and killed in human areas with those of bears that remained in the forest.



Figure 1-1. Brown bears hunt for salmon at the Rusha River, Shiretoko, Hokkaido, August 24, 2018 (photo by Jimbo M.).

Chapter 2 | Hair growth pattern in brown bears

2.1 Introduction

In bears (*Ursus*), hair stable isotope analyses are widely used to examine their foraging ecology (e.g., Hobson et al. 2000, Felicetti et al. 2003, Mowat et al. 2017, Deacy et al. 2018, Naganuma et al. 2020). It is known that whole hairs collected from shoulder, rump, and abdomen do not differ in values of hair biomarkers (Macbeth et al. 2010, Ueda and Bell 2019), therefore, non-invasive hair sampling, such as the hair traps resulting in the hair being collected from various regions of the body, is effective to collect bear hairs. Analyzing stable isotope ratios ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$) in whole hairs is the most popular method to determine the average annual assimilated diet especially meat consumption (e.g., Hobson et al. 2000, Mowat et al. 2017, Adams et al. 2017, Deacy et al. 2018). Segmental analysis of stable isotope values of guard hair, an advanced method to estimate an individual's feeding history on a more detailed time scale has also been developed (Mizukami et al. 2005b, Hata et al. 2017). For such analyses, detailed information is needed on the factors influencing the values of each biomarker, including turnover rate, discrimination factor, and hair growth pattern. Among these, hair growth pattern is the most basic factor; however, few studies have been published describing hair growth in brown bears (*Ursus arctos*).

Bear hair grows throughout the year, except during hibernation, and sheds once per year; however, such common knowledge has not been scientifically verified via observation test. The pelage of bears, consists of three hair types: guard hair is long and coarse, underfur is short and fine, and intermediate hair is intermediate in structure (Perrin and Campbell 1980). And classification of bear hair based on visual observation of the hair shaft and hair bulb thickness was previously published (Yamauchi et al. 2014). Although the growing period of each hair type is likely to differ (Jones et al. 2006), the growth periods of each hair type have not yet been documented in brown bears.

Another difficulty with using hair samples in empirical studies is the issue of determining, whether sampled hair has grown during the current or previous year especially during periods of shedding. Noninvasive hair traps are a common method for sampling wild bears, and some studies distinguish hairs on the basis of length in relation to sampling season (e.g., Mowat et al. 2017); however, this method is not absolute because hair length varies among individuals and body parts. A more appropriate method for distinguishing the onset of hair growth is needed.

In this study, I described the hair growth pattern of brown bears including (1) the growth periods of

each hair type, (2) shedding, and (3) the growth rate of guard hairs. I also investigated a method to discriminate between the previous year's hair and the current year's hair via hair follicle structure.

2.2 Materials and Methods

Hair growth observation in captive brown bears

Captive adult female brown bears (n = 4) housed in the Noboribetsu Bear Park (Noboribetsu, Hokkaido, Japan) were observed for hair growth patterns (Table 2-1). Observations were performed monthly from late April 2019 to late November 2019. Bears in the Noboribetsu Bear Park were fed bear pellets (ZOOFOOD bear; Nosan Co., Kanagawa, Japan), concentrated feed formulated for cows (Soyokazenokaori MG; Nippon Formula Food Manufacturing Co., Ltd., Kanagawa, Japan), vegetables, and fruits once or twice a day with water ad libitum. The bears were immobilized using blow darts to administer intramuscular xylazine HCl (1 mg / kg; Selactar; Bayer, Tokyo, Japan) and a 1:1 mixture of zolazepam HCl and tiletamine HCl (2.0–4.0 mg / kg; Zoletil 100; Virbac, Carros, France). After all of the experiments were finished, atipamezole HCl (1 mg / kg, Atipame; Kyoritsu, Tokyo, Japan) was injected intramuscularly to aid in recovery. All experimental procedures were conducted in accordance with the Guidelines for Animal Care and Use of Hokkaido University, and were approved by the Hokkaido University Animal Care and Use Committee (permit number: 19-0021).

All hair samples were collected along the center of the upper back, where hair would be most likely be taken from wild bears that rubbed their backs against trees. On the first day of the experiment at the end of April, two 5 cm × 5 cm patches along the back were shaved ("patch A" and "patch B"). On another 5 cm × 5 cm patch in the same area ("patch C"), a commercial hair bleaching agent was applied and incubated at room temperature for 30 minutes to mark the previous year's hair. Newly grown hair was shaved and collected from patch A once per month. All hair samples were classified into one of the three types (guard hair, intermediate hair, or underfur) through visual observation in accordance with the criteria of Yamauchi et al. (2014): guard hairs are thick and straight, underfur is thin and downy, and all others are classified as intermediate hair. The length of guard hair was measured for ten randomly selected guard hairs, and the measured values were averaged. All subsequent measurements of guard hair length were performed using the same method. Because shaving once a month may change the rate of hair growth, the total hair length was measured at patch B, where no procedures were performed from May to October. Fully grown guard hairs were collected and measured at the end of November. In each month, the presence

of bleached hair in patch C was checked to identify the shedding period.

Fur observation in wild brown bears

Fur samples were collected from the same area on the center of upper back (as described above) of wild brown bears that were killed for hunting and nuisance control from April 2019 to November 2019 ($n = 45$) in the Shiretoko Peninsula, eastern area of Hokkaido, Japan (Fig. 2-1). Before sampling, body weight and sex were recorded. In accordance with the growth curve of brown bears in Hokkaido (Moriwaki et al. 2018), mean body mass at age five years (148 kg for males and 91 kg for females) was used as a threshold for discrimination between adult and sub-adult bears. Each bear was classified based on body weight: newborn cubs (< 10 kg), adult females (> 91 kg), adult males (> 148 kg); all others were considered as sub-adults. Hairs were pulled out from the fur and classified as either guard hair, underfur, or intermediate hair. Then, the lengths of guard hairs were measured and the structures of hair follicles were observed.

Statistical analysis

To account for the effect of shaving on hair growth, a correction value was calculated by dividing the length of fully-grown guard hair measured in patch B by the total length of guard hair measured during the experimental period in patch A. Then, the monthly-grown hair length in patch A was multiplied by the correction value, which was used to calculate the growth rate (mm / day) for each month. Turkey's multiple comparison test was used to compare monthly growth rates among individuals. These statistical analyses were performed in the multcomp package (Hothorn et al. 2008) in R ver.3.6.2 (R Core Team 2019).

The current year's guard hair was determined based on the shape of hair bulbs (details regarding hair discrimination are described below), and its lengths were used to calculate the hair growth rate of wild bears (Table 2-2). First, a multiple regression analysis was conducted to examine the effects of maturity class and sex on hair growth rates of wild bears. Since there were no significant effects of maturity class or sex on the hair growth rate of wild bears (all $P > 0.05$), I pooled all samples for wild bears irrespective of maturity class and sex for the next analysis. The approximate hair growth rate was calculated; guard hair length over time was plotted independently for wild bears to form a linear regression.

2.3 Results

Hair growth and shedding stages

In patch A on captive bears, newly grown hairs could be visually distinguished by their thin and pointed shaft tips. Guard hairs began to grow first, followed by intermediate hairs, and then underfur. Newly grown hairs were not observed on the first day of the experiment except on Bear 1, whose guard hairs were about 10.9 mm by late April and continued growing into September. In Bears 2 and 3, guard hairs began growing in May and continued growing into September. In Bear 4, guard hair grew slightly in May and June, grew as long as other individuals in July and continued growing into October. Guard hair did not grow in November (Fig. 2-2).

There was variation in the onset of intermediate hair growth. The growth periods varied from three to five months: from June to November (Bear 1), June to September (Bear 2), June to October (Bear 3), and July to November (Bear 4). Underfur grew from August to late November in all captive bears, although I did not identify the period of growth termination. The amount of the bleached hairs (i.e., previous year's hair) declined beginning in May, and they were completely shed by July (Bear 1), August (Bears 2 and 3), or September (Bear 4). Hairs in this section did not regrow during the experiment.

Newly grown hairs were not observed in wild bears in April or early May. From the end of May to August, guard hairs of two different lengths were observed. Since this period was consistent with the shedding and growing periods of guard hairs in captive bears, I considered the shorter hairs to be newly grown and the longer ones to be the previous year's hairs. From September to November, guard hairs were uniform in length, indicating that shedding was complete by September (Table 2-2).

Guard hair growth rate

There were no significant differences in monthly growth rates of guard hairs between captive bears from May to September (Tukey-Kramer test; $P > 0.10$). Because guard hair growth was terminated in October in all captive bears, the approximate hair growth rate was calculated using data from May to September (i.e., data from April, October, and November were excluded). The approximate hair growth rate of captive bears was calculated as 0.5 mm / day ($r^2 = 0.75$), and that of wild bears was calculated as 0.7 mm / day ($r^2 = 0.74$) (Fig. 2-3). The average ($\pm SE$) length of fully-grown guard hairs was 79.4 ± 1.7 mm (range: 59.1–106.6 mm); this did not significantly differ between captive and wild bears (Student's t-test; $t = 0.734$, $P = 0.47$, $df = 31$).

Hair follicle structure

The shape of hair bulb was classified into three types by gross observation: white spheres (WS-type), black hooks (BH-type), and white hooks (WH-type) (Fig. 2-4). WS-type bulbs were observed from April to August, while BH-type bulbs were observed from June to October. From June to August, guard hairs with different lengths were observed together; the longer hair was WS-type bulb, while shorter hair was BH-type bulb (Table 2-2). From late-August into October, WH-type bulbs were observed alongside BH-type bulbs. Only one sample was collected in November; this sample was WS-type bulb (Table 2-2).

2.4 Discussion

The growth periods of guard hair and underfur were almost the same for all captive bears; guard hairs began growing in spring prior to shedding and continued growing to autumn, while underfur began growing in late summer. Although the termination of underfur growth was not clear, it should grow thicker towards winter, because its principal function is insulation (Leblond 1951). These results suggest that hair biomarkers can be used to compare the physiology and ecology of bears between spring to autumn (guard hair) and autumn to hibernation (underfur). Jones et al. (2006) previously showed that stable isotopic values differ between whole guard hair and underfur. In contrast to guard hair and underfur, the growth of intermediate hairs varied greatly among individuals, and thus this type of hairs should not be used for practical applications.

The regression analysis of guard hair growth indicated that the average growth from late April to early October was 0.5 mm / day for captive bears. Assuming that guard hairs grow at a constant rate until fully grown (mean length = 79.4 ± 1.7 mm), the growth period of guard hairs is approximately five months for captive bears. This pattern is similar to that of captive grizzly bears, as reported by Felicetti et al. (2004) (i.e., growth from early May to October at a rate of approximately 1.5 cm /month). Conversely, the approximate hair growth rate was 0.7 mm / day for wild bears, and the growth period spanned approximately four months from late May to late September. This implies that the hair growth rate of wild bears is higher than that of captive bears and that hair growth in wild bears begins later than that in captive bears. This difference could be attributed to differences in feeding conditions between wild and captive bears. Bears in Noboribetsu Bear Park are usually fed a mixture of commercial foods and the amount of feeding is almost constant throughout the year. In contrast, the diets of wild brown bears are restricted to

low-energy food, e.g., plants, in spring and shift to high energy foods, e.g., fruits and salmon, in summer and fall (Ohdachi and Aoi 1987). Because energy or protein restrictions influence hair growth (e.g., McGregor 1988, Rasmussen and Børsting 2000), it is conceivable that, in wild bears, hair growth is restricted during periods of food scarcity in spring and is initiated at a high rate in summer and autumn when food is abundant.

I confirmed that there were no differences in hair growth among maturity classes or between sexes. However, hair growth in cubs and some adult males might begin later (i.e., in June), as their hair length was only 10–17 mm in early July. Because firm conclusions could not be drawn from my dataset, further experiments will be required to investigate the relationship between age and hair growth more thoroughly.

Shedding in captive bears was completed between late July and late September, as in wild brown bears. In wild bears, long and short hairs were intermixed from June to mid-August, which suggested that shedding was ongoing. Subsequently, a uniform length was observed, which indicates that shedding is typically complete by late August; however, it can fluctuate by approximately one month, depending on the individual (Table 2-1). It is unlikely that the previous year's hair remains after October; therefore, guard hairs collected after October can be regarded as the current year's hair.

I recommend analyzing the hair bulb shape to distinguish between the previous year's hair and the current year's hair collected during shedding period. I observed that morphological changes of hair bulbs occur in conjunction with hair growth cycle. Hair growth occurs in cycle, alternating between periods of growth (anagen), quiescence (telogen), and the transition between growth and quiescence (catagen) (e.g., Parakkal 1970, Diaz et al. 2004). My observations suggest that, during shedding, telogen hair (i.e., hair grown during the previous year) has a WS-type bulb, while anagen hair (i.e., hair grown during the current year) has a BH -type bulb. After shedding, catagen hair has a WH-type bulb. WS-type bulb was observed again in November, suggesting that the growth of the current year's hair had ended. Based on these results, I can distinguish hairs during shedding on the basis of hair bulbs: the previous year's hairs have the WS-type bulbs, and the current year's hairs have BH-type or WH-type bulbs. This is a simple and reliable method that can be implemented without the use of a microscope.

To increase the practicality of hair biomarker analyses, further studies of other factors influencing the values of each biomarker are needed. The application of hair stable isotope data to dietary studies requires understanding the isotopic discrimination value and turnover rate. The isotopic discrimination value represents the stepwise isotopic changes between a consumer and its prey, and the turnover rate is the

length of time it takes for stable isotopes in tissues to be replaced by isotopes derived from the diet. When conducting experiments using a controlled diet to quantify variation in isotopes according to hair growth, information on hair growth is essential to select the length of feeding trials. The hair growth patterns of brown bears revealed in this study provide the basic knowledge for such future research.

Table 2-1. List of captive brown bears used in monthly observations.

Bear ID	Sex	Age	Shedding periods	Growth periods			Length of guard hair (mm)
				Guard	Intermediate	Underfur	
Bear 1	F	20	-Jul	Apr-Sep	Jun/Jul-Nov	Aug-	91.7
Bear 2	F	17	-Aug	May-Sep	Jun-Sep	Aug-	77.8
Bear 3	F	23	-Aug	May-Sep	Jun-Oct	Aug-	64.6
Bear 4	F	6	-Sep	May-Oct	Jul-Nov	Aug-	77.4

Table 2-2. List of samples from wild brown bears.

Capture date	Bear ID	Sex	Maturity	Weight (kg)	Previous guard hair		Present Guard hair	
					Length (mm)	Bulb type	Length (mm)	Bulb type
2019-04-08	19B01	M	Sub adult	53	80.6	WS	-	-
2019-04-12	SB19B01	M	Sub adult	146	75 <	WS	-	-
2019-04-16	19B02	M	Sub adult	25	73.1	WS	-	-
2019-04-19	R19B01	M	Sub adult	27	106.6	WS	-	-
2019-05-02	R19B02	F	Adult	103	80.0	WS	-	-
2019-05-02	R19B03	M	Sub adult	48	84.3	WS	-	-
2019-05-02	R19B04	M	Sub adult	39	92.2	WS	-	-
2019-05-24	R19B05	M	Sub adult	52	84.5	WS	11.5	BH
2019-06-11	19B07	F	Sub adult	61	82.1	WS	20.8	BH
2019-06-16	19B08	M	Sub adult	45	66.9	WS	12.2	BH
2019-06-27	R19B06	M	Sub adult	45	81.9	WS	19.7	BH
2019-06-28	19B09	F	Sub adult	85	85 <	WS	11.3	BH
2019-06-28	19B10	M	Sub adult	27	79.3	WS	14.3	BH
2019-06-28	19B11	M	Sub adult	21	77.7	WS	14.3	BH
2019-06-28	19B12	M	Adult	187	75 <	WS	24.6	BH
2019-07-02	19B16	F	Cub	8	59.1	WS	13.0	BH
2019-07-02	19B17	M	Cub	7	61.1	WS	14.0	BH
2019-07-02	19B18	F	Cub	8	69.1	WS	17.7	BH
2019-07-02	R19B07	M	Adult	216	86.4	WS	10.3	BH
2019-07-08	19B20	M	Adult	161	73.4	WS	10.8	BH
2019-07-09	19B21	M	Adult	174	80 <	WS	38.4	BH
2019-07-12	19B22	M	Sub adult	32	77.8	WS	47.4	BH
2019-07-13	19B24	M	Adult	262	79.5	WS	14.6	BH
2019-07-13	19B25	F	Adult	95	82.4	WS	24.4	BH
2019-07-14	19B26	M	Adult	180	80.3	WS	20.5	BH
2019-07-14	19B27	F	Adult	119	91.3	WS	33.1	BH
2019-07-14	R19B08	M	Sub adult	60	81.8	WS	49.7	BH
2019-07-19	19B28	F	Sub adult	83	84.4	WS	57.8	BH
2019-07-24	19B29	F	Adult	103	71.8	WS	40.9	BH
2019-07-26	19B30	M	Sub adult	103	82.3	WS	39.9	BH
2019-07-28	19B32	M	Sub adult	73	63 <	WS	48.4	BH
2019-07-29	19B33	M	Sub adult	45	67.3	WS	33.9	BH
2019-08-01	19B34	M	Adult	158	-	-	60.2	BH
2019-08-05	19B35	F	Sub adult	35	66.3	WS	53.3	BH
2019-08-14	R19B09	M	Adult	189	65 <	WS	42.1	BH
2019-08-16	R19B10	M	Sub adult	107	-	-	76.6	BH
2019-08-24	R19B11	M	Sub adult	114	85.9	WS	59.0	BH
2019-08-28	19B36	M	Adult	158	-	-	50.4	BH&WH
2019-08-31	SB19B03	F	Sub adult	74	-	-	69.7	BH&WH
2019-09-04	SB19B04	M	Sub adult	71	-	-	70.7	WH
2019-09-06	19B37	M	Adult	199	-	-	72.2	WH

Capture date	Bear ID	Sex	Maturity	Weight (kg)	Previous guard hair		Present Guard hair	
					Length (mm)	Bulb type	Length (mm)	Bulb type
2019-09-06	R19B12	F	Sub adult	61	-	-	66.8	WH
2019-10-16	19B39	M	Sub adult	67	-	-	87.2	BH&WH
2019-10-25	R19B13	F	Adult	108	-	-	80.1	BH&WH
2019-11-04	19B40	M	Sub adult	75	-	-	85.1	WS

Hair bulb types are shown as WS: white sphere, BH: black hook, and WH: white hook. Lengths of guard hairs written in bold were used to calculate hair growth rate.

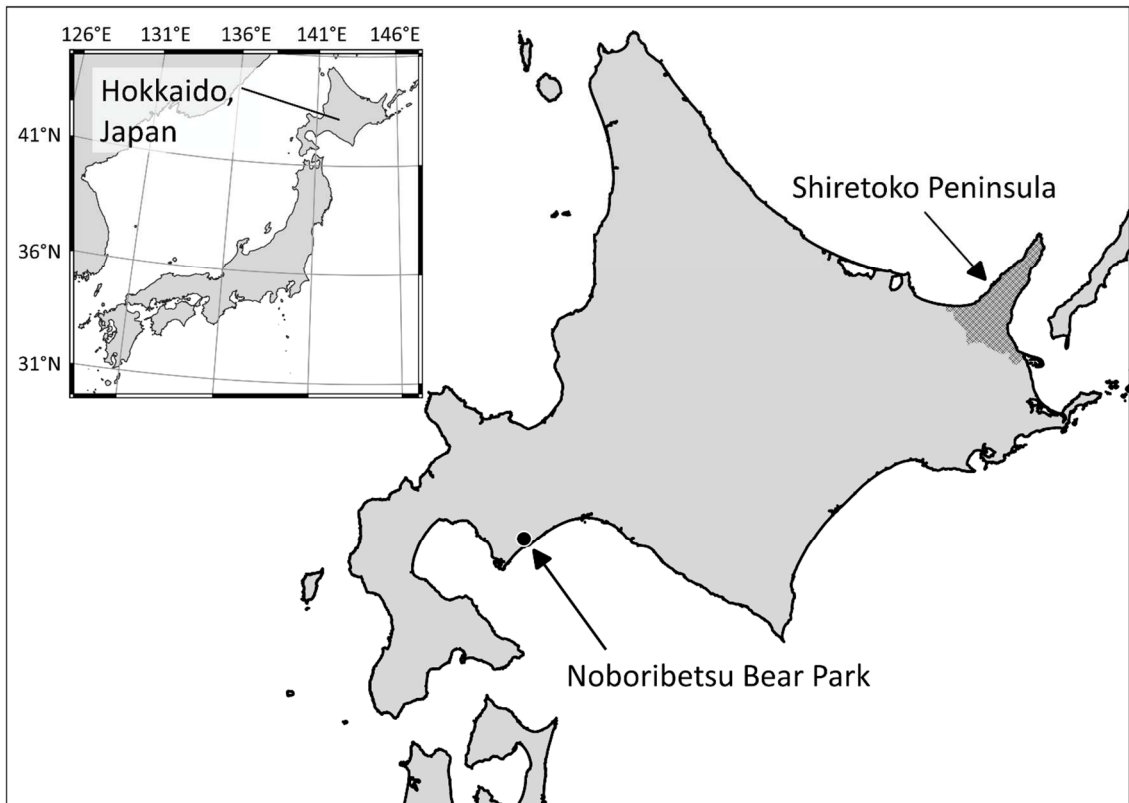


Figure 2-1. Map of Hokkaido, Japan, including the Shiretoko Peninsula and Noboribetsu Bear Park.

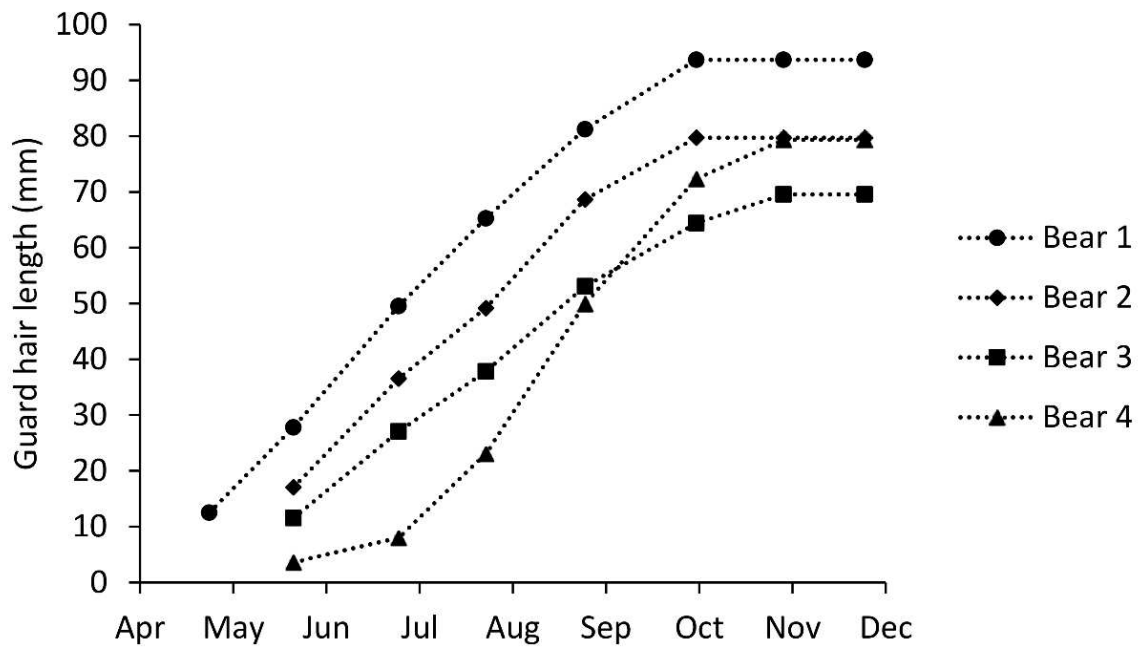


Figure 2-2. Cumulative length plots of guard hairs of captive bears; Bear 1 (●), Bear 2 (◆), Bear 3 (■), and Bear 4 (▲). The guard hair length was measured each month in patch A.

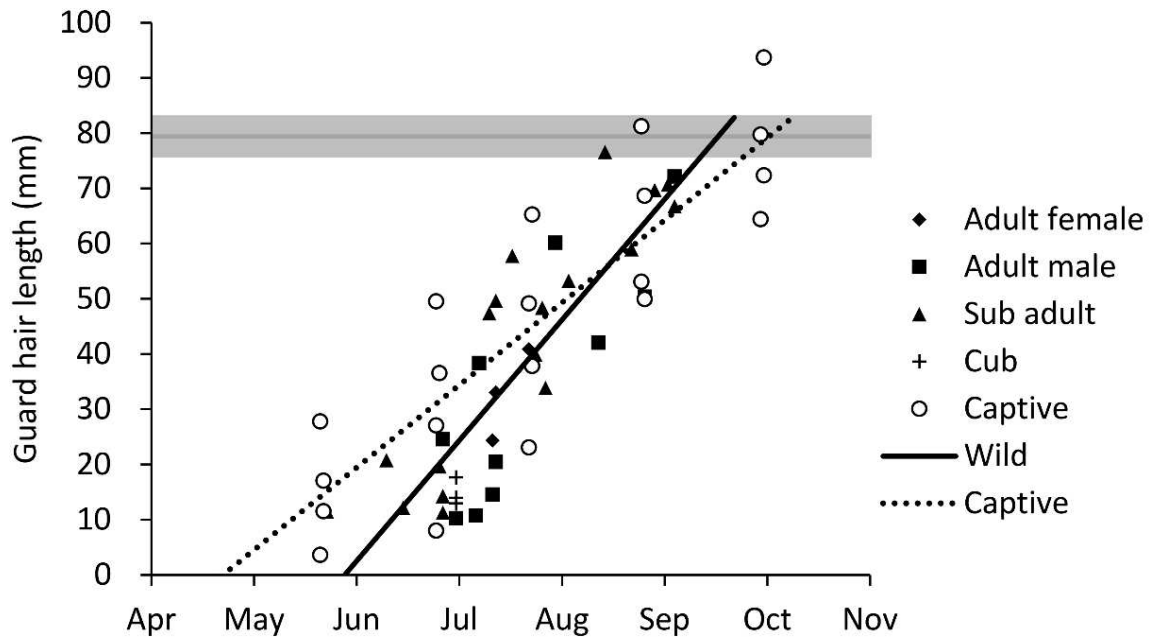


Figure 2-3. Length of guard hairs in captive bears (○), wild adult females (◆), wild adult males (■), wild sub-adults (▲), and wild cubs (+) are plotted sequentially. Grey dotted horizontal line indicates the mean length of guard hairs, with a 95% confidence interval (grey zone). The slope values of linear regression which indicate the growth rate (mm / day) were 0.5 for captive bears ($r^2 = 0.75$), and 0.7 for wild bears ($r^2 = 0.74$).

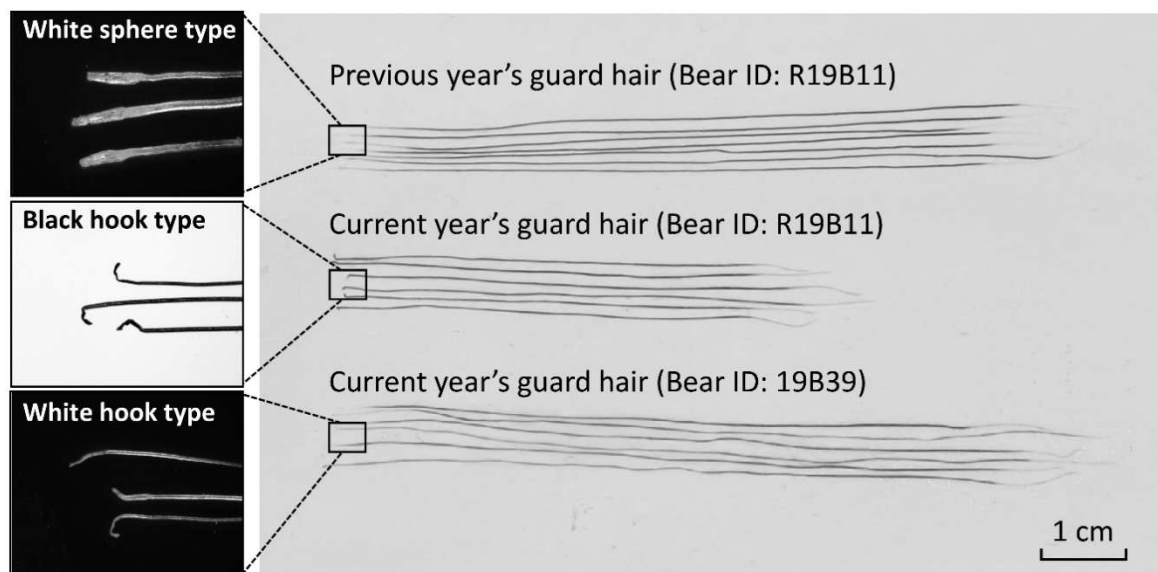


Figure 2-4. Guard hairs of different lengths were observed in the same fur sample (upper right image is an example from Bear ID: R19B11). White sphere type bulbs (upper left image) were observed in the previous year's hair, while black hook type bulbs (middle left image) were observed in the current year's hair. From late summer to autumn, white hook type bulbs (lower left picture) were observed in the current year's guard hair (lower right image is an example from Bear ID: 19B39).

Chapter 3 | Role of asocial learning in high-energy food consumption for male brown bears.

3.1 Introduction

Diet selection is a complex process resulting from genetic inheritance (Arnold 1981), asocial or social learning (Heyes and Galef 1996, Galef and Laland 2005), and the interaction of inheritance and learning (Kandel et al. 2021). Social learning involves animals learning by observing and interacting with others, such as parents and conspecifics (Heyes and Galef 1996, Galef and Laland 2005). Representative mother-offspring social learning has been studied extensively in many mammalian species, and mothers have been shown to directly influence the feeding styles of their young offspring (e.g., rodents: Peacock and Jenkins 1988, Lupfer et al. 2003, primates: Lynch et al. 2020, and cetaceans: Mann et al. 2007). Even in solitary animals that live independently after spending a certain period of time with their mothers, mother-offspring social learning partially influences the diet selection of young offspring (e.g., black rat: Terkel 1996, black bear: Mazur and Seher 2008, and orangutan: Jaeggi et al. 2010). The question is how long the effects of mother-offspring social learning continue to influence the diet selection of solitary animals.

Asocial learning is defined as animals learning based on their own experiences through trial and error (Heyes and Galef 1996, Kendal et al. 2005). Animals need to adaptively rely on social and asocial learning based on a trade-off between the cost of collecting information and its reliability (Kendal et al. 2005). Echeverría and Vassallo (2012) found that although diet selection during weaning might be influenced by the mother, maternal influence was not essential for diet selection during dispersal in solitary rodents (*Ctenomys talarum*). Additionally, Galef and Whiskin (2001) provided experimental evidence that socially enhanced food preferences weaken after animals have experienced alternative foods, especially high-quality alternatives. These studies indicate that animals can change their diet selection as they gain independent foraging experience, but few studies have addressed this point in free-ranging solitary animals. The foraging experience that animals acquire after becoming independent may play a significant role in their diet selection later in life, especially for brown bears that forage solitarily for a decade or more after becoming independent from their mothers.

Bears (*Ursus*) spend the first 1-2 years of life with their mothers (Schwartz et al. 2006, Shimozuru et al. 2017) and forage independently thereafter. It has been suggested that mother-offspring social learning influences the feeding style of offspring (e.g., Aumiller and Matt 1994), whereas the influence of genetic

inheritance has not been supported (e.g., Hopkins 2013, Morehouse et al. 2016). The habitats in which offspring spend time with their mothers (hereinafter natal habitat), rather than their mother's food preferences, influence the feeding style of offspring for up to two years after independence (Mazur and Seher 2008). Brown bears (*U. arctos*) are generally opportunistic predators, and their diet is generally associated with environmental conditions (i.e., the local availability of food) (Bojarska and Selva 2012). In addition, it is widely recognized that sex and age can influence brown bear diets (Ben-David et al. 2004, Mowat and Heard 2006, Edwards et al. 2011, Adams et al. 2017). Such dietary variations characterized by intrinsic factors of individuals are generally considered to result from differences in energy requirements (mainly based on body size and reproductive costs) and/or interindividual interactions (e.g., food competition and social hierarchy) (Hilderbrand et al. 1999, Ben-David et al. 2004, Mowat and Heard 2006).

The Shiretoko Peninsula, contains an abundance of energy-rich salmon and pine nuts and has one of the highest densities of brown bear populations worldwide (Hokkaido Government 2017). Nevertheless, the availability of these food resources is unevenly distributed within the peninsula. Therefore, this peninsula is an appropriate study site to examine how the natal habitat influences diet selection by brown bears later in life. Here, I analyzed a large amount of genetic and dietary data of brown bears inhabiting the Shiretoko Peninsula, Hokkaido, Japan, to examine 1) factors influencing diet and 2) dietary shifts later in life relative to each natal habitat.

3.2 Materials and Methods

Field site description

The Shiretoko Peninsula (43°50'–44°20'N, 144°45'–145°20'E) is located on eastern Hokkaido, Japan, protruding north-northeast into the Sea of Okhotsk (Fig. 3-1). The peninsula is long and narrow, measuring approximately 70 km long and approximately 25 km wide at its base. The Shiretoko Peninsula has three towns, Shari, Rausu and Shibetsu, and the area from the center to the tip of the peninsula is Shiretoko National Park (hereinafter SNP). Most of the land area within the national park is designated as a special protection zone, and human activities that affect wildlife and the natural environment are strictly restricted.

Mountains up to 1,660 m high run down the center of the peninsula, and Japanese stone pine (*Pinus pumila*) is distributed in the subalpine zone. A total of 93 river systems flows into the sea from mountains

throughout the peninsula. Pink salmon (*Oncorhynchus gorbuscha*) and chum salmon (*O. keta*) spawn from summer to autumn in some of these rivers (Appendix S3-1). However, salmon are able to swim upstream in only a limited number of rivers due to dams, fish traps, and steep topography (Appendix S3-1). The rivers and the predicted spawning areas of pink salmon based on elevation, slope, and location of artificial structures are shown in Fig. 3-1. Both salmon and pine nuts are known to be important foods for brown bears inhabiting the Shiretoko Peninsula (Shirane et al. 2021); therefore, I divided the study site into five areas in terms of accessibility to salmon spawning rivers and the stone pine zone for bears (Fig. 3-1). The tip SNP area is the area where brown bears can most easily consume both salmon and pine nuts because the distance between the salmon spawning river and the stone pine zone is small and the area experiences minimal human disturbance. The accessibility to the stone pine zone is high in both the western middle (hereinafter Utoro) and eastern middle (hereinafter Rausu) areas. The accessibility to salmon spawning rivers is low in the Utoro area because this area contains few rivers where salmon can swim upstream and most of the spawning areas are close to areas with high human activity, such as residential areas, roadways, and fishing spots. In the Rausu area, the accessibility to salmon spawning rivers is moderate because in this area, brown bears may consume salmon upstream, as residential areas are distributed only along the coastline. The accessibility to the stone pine zone is low in the western base (hereinafter Shari) and eastern base (hereinafter Shibetsu) areas because these areas contain only marginally distributed stone pine zones in the mountains. In the Shari area, most rivers are surrounded by residential and rural areas, providing limited access for brown bears; thus, the accessibility to spawning salmon in the river is low. In the Shibetsu area, salmon can swim far upstream because the topography is less steep. In addition, residential and rural areas are distributed inland in the Shibetsu area, making estuaries relatively accessible for brown bears. Therefore, the accessibility to salmon spawning rivers is moderate in the Shibetsu area. I used the names of the towns as the area names for clarity, but area boundaries and administrative divisions are different.

Bear hair collection and data curation

I collected hair samples from brown bears throughout the Shiretoko Peninsula between March and December from 2010–2020. Most bears were killed for nuisance control or hunting, and some were captured for research purposes. Nuisance control and hunting of bears occur outside the national park with rare exceptions according to a management plan (Kushiro Nature Conservation Office of the

Ministry of the Environment et al. 2017). Within 24 hours after a bear was dead or during immobilization, whole hairs, including hair roots, were plucked from the back of the bear and stored in a paper bag at -30°C. In the first step of sample preparation, the hairs were briefly washed with distilled water to remove large stains, rinsed with 70% ethanol, and dried at 60°C for 48 hours.

Our research group collected genetic samples from all the captured bears. We extracted genomic DNA and genotyped it using 21 microsatellite markers and one sex marker (details of the genotyping were described in Shimozuru et al. 2017). We also conducted parentage analysis to assign bears to parents (details of the parentage analysis can be found in Shirane et al. 2019). The birth area of each bear was defined as the collection site of genetic data for the putative mother according to Shirane et al. (2019) and assigned to one of the above-described five study regions. Genetic data for bears captured from 2010 to 2018 were reported by Shirane et al. (2018, 2019), and Shimozuru et al. (2020a, 2020b), and those for bears captured in 2019–2020 have not yet been published.

The age of each bear was estimated by counting the cementum annuli of the teeth (Yoneda 1976). For individuals whose birth year was recorded by observation and who could be individually identified, the age based on such field observations was adopted. The age of individuals classified as the mother or father of another known individual by parentage analysis was estimated by the age of the offspring plus the average age of the first reproduction (Shimozuru et al. 2017). For three individuals for which neither tooth samples, field observation records, nor relatedness information were available, approximate age was estimated from the growth curve of body length (Shirane et al. 2020). The age data that were used for my analysis were modified based on the age when the hair grew. If a hair was determined to be from the previous year based on the shape of the hair bulb (Jimbo et al. 2020), the age was determined to be the estimated age at the time of capture minus one year. The bears of each sex were classified into three age classes: juvenile, sexually mature adult, and physically mature adult. On the Shiretoko Peninsula, most offspring become independent after 1 or 2 years (Shimozuru et al. 2017). The minimum age of reproductive maturity for brown bears is generally reported to be 3 years for both females and males (e.g., Zedrosser et al. 2004, 2007), and males are known to start dispersing at 3 years old on the Shiretoko Peninsula (Shirane et al. 2019). Females reach 95% of their asymptotic body length at age 4.6 and males at 7.6 years (Shirane et al. 2020). Therefore, I defined juveniles as bears aged 0–2 years (both females and males), sexually mature adults as females aged 3–4 years and males aged 3–7 years, and physically mature adults as females aged ≥ 5 years and males aged ≥ 8 years.

Potential food items

I also collected 33 potential food items of brown bears inhabiting the Shiretoko Peninsula according to a list developed based on previous studies (Ohdachi and Aoi 1987, Sato et al. 2004, Shirane et al. 2021) and information from local wildlife experts. As suggested by Phillips et al. (2005, 2014), I used the ecology of bear foraging to a priori aggregate potential food items into the following five food categories: alpine plants (pine nuts and wild berries), plants (e.g., herbaceous plants, fruits, and acorns), dent corn (*Zea mays*), terrestrial animals (insects and sika deer), and marine animals. The marine animals category included pink salmon, chum salmon, and marine mammals. Dent corn was included as a representative crop eaten by brown bears in eastern Hokkaido (Sato et al. 2004). However, most of the crop damage reported from Shari town included beets and wheat, and dent corn damage was reported from only Shibetsu town. However, I did not include beets and wheat in my analyses because I could not distinguish them from other natural plants by isotopic signatures.

Stable isotope analysis

I separated approximately 10 to 30 guard hairs that were not broken from root to tip and removed their follicles after recording the hair bulb type. After sorting, the hair samples were washed with a 2:1 chloroform:methanol (v/v) mixture according to a standard protocol for stable isotope analysis to remove contamination and surface lipids (Hobson et al. 2000) and then dried at 60°C for 48 hours. Each hair sample was placed in a 2 ml reinforced tube (Precellys Lysing Kit, Bertin Technologies, France) and ground into fine powder using a ball mill (Shakeman 6, Biomedical Science, Japan) for 10 cycles of 90 s at 4350 rpm after adding four ϕ 3.0 mm and one ϕ 5.0 mm zirconia ball bearings. The food samples were washed with distilled water, cut into small pieces, and dried in an oven at 60°C for at least 48 hours. After complete drying, they were ground into fine powder using a ball mill.

Ground hairs (0.8–1.2 mg) and food items (2.8–3.2 mg plant matter, 0.8–1.2 mg animal matter) were weighed and enclosed in tin boats (4 × 4 × 11 mm). The carbon, nitrogen, and sulfur stable isotope ratios ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$) of the samples were analyzed using an isotope ratio mass spectrometer coupled with an elemental analyzer (IsoPrime 100-vario MICRO cube, Elementar, Germany). The results were reported as parts per thousand of the isotopes (‰) relative to a standard as follows:

$$\delta^{13}\text{C}, \delta^{15}\text{N}, \text{ or } \delta^{34}\text{S} = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3$$

where R is $^{13}\text{C}/^{12}\text{C}$, $^{15}\text{N}/^{14}\text{N}$, or $^{34}\text{S}/^{32}\text{S}$ and R_{standard} is Vienna Pee Dee Belemnite (VPDB) for $\delta^{13}\text{C}$, atmospheric nitrogen (AIR) for $\delta^{15}\text{N}$, and Vienna Canyon Diablo Troilite (VCDT) for $\delta^{34}\text{S}$. Since carryover effects were observed in the measurement of sulfur isotopes, I took five measurements for one sample and adopted the latter three values as the measurement values.

I used five amino standards (L-alanine: $\delta^{15}\text{N} = 1.79\text{‰}$ with $\delta^{13}\text{C} = -19.9\text{‰}$, $\delta^{15}\text{N} = 3.7\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, $\delta^{15}\text{N} = 4.99\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, $\delta^{15}\text{N} = 8.72\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, and $\delta^{15}\text{N} = 13.7\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, Shoko Science Co., Ltd., Japan) and three sulfur standards (silver sulfide: IAEA-S-1, IAEA-S-2, and IAEA-S-3, International Atomic Energy Agency, Australia) with $\delta^{34}\text{S} = -32.3\text{‰}$, -0.3‰ , and 22.7‰ as standard reference materials. These standard reference materials were weighed (1.0 mg for the amino standards and 0.2 mg for the sulfur standards), enclosed in tin boats, and run at the start and end of every three samples (i.e., fifteen measurements) to provide reference gas calibrations. Based on these measurements, calibration curves, consisting of straight lines for the nitrogen and sulfur isotopes, were constructed. All the analytical errors in this study were within 0.2‰ for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and 0.3‰ for $\delta^{34}\text{S}$.

Isotopic value correction according to hair growth stage

I classified the growth stages of the hair based on the shapes of hair bulbs according to Jimbo et al. (2020). The shape of the hair bulbs were classified into three types by gross observation: black hook (hereinafter BH-type), which indicated that hairs were growing in the current year of capture; white hook (hereinafter WH-type), which indicated that hairs almost stopped growing during the current year of capture; and white sphere (hereinafter WS-type), which indicated that hairs had grown during the previous year of capture (Jimbo et al. 2020).

Of the total 295 samples, 80 hair samples, which were collected from bears captured between June 27 and October 28, were classified as having BH-type bulbs. Forty-five hair samples, which were collected between September and December, were classified as having WH-type bulbs, and 170 hair samples, which were collected between March and November, were classified as having WS-type bulbs. The mean carbon and sulfur isotopic values were significantly lower in the guard hairs with BH-type bulbs than in those with WH- and WS-type bulbs (Steel-Dwass test, both $p < 0.05$), while the mean nitrogen isotopic values

were not significantly different among the bulb types (Kruskal-Wallis rank sum test, chi-squared = 2.8, $p = 0.25$). There were positive correlations between the sampling date and the three stable isotopic ratios for guard hairs with BH-type bulbs ($\delta^{13}\text{C}$: $p < 0.05$, $R^2 = 0.14$, coefficient = 0.018; $\delta^{15}\text{N}$: $p < 0.05$, $R^2 = 0.043$, coefficient = 0.020; $\delta^{34}\text{S}$: $p < 0.05$, $R^2 = 0.041$, coefficient = 0.024), but no significant correlations were shown for guard hairs with WH- or WS-type bulbs (all $p > 0.05$) (Fig. 3-2). Therefore, I corrected the isotopic values of guard hairs with BH-type bulbs to the full hair growth season diet using the following procedure. Since BH-type bulbs were observed until October 28 in my sample sets, October 28 was used as the baseline based on the regression line between the sampling date and measured isotopic values. The following equation was used to correct the values:

$$\text{Correction } X = \text{Measured } X + \text{SLOPE}_X \times (301 - \text{Sampling date})$$

where X is the measured values of $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, or $\delta^{34}\text{S}$, SLOPE is the slope of the regression line between the sampling date and each isotopic ratio, and $(301 - \text{Sampling date})$ is the number of days between October 28 and the date of hair sample collection. After correction, I again compared the mean values of the isotopic ratios among the hair bulb types, and no significant differences were found (Kruskal-Wallis rank sum test, all $p > 0.05$). The corrected values for the hair samples with BH-type bulbs were used in the subsequent analysis.

Stable isotope mixing model

I first tested the differences in the $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ values among the five food categories using a Kruskal-Wallis rank sum test with Steel-Dwass nonparametric post hoc comparisons. Statistically significant differences in at least one of the stable isotopic values (i.e., $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, or $\delta^{34}\text{S}$) were found in all combinations of four categories (alpine plants, plants, terrestrial animals and marine animals). There were no statistically significant differences between dent corn and alpine plants, terrestrial animals, or marine animals, probably because of small sample sizes, but there were significant differences in the values of $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ between dent corn and plants. With regard to the potential food items used as sources in the mixing models, aggregation of species with similar functional significance has been recommended when the isotopic signatures of food items are not significantly different (Phillips et al. 2005). In this case, I kept dent corn as an independent source because it is clear that crops have different

ecological functions in bear diets.

To estimate the proportional contributions of each food category to the diet of bears at the individual level, I used a stable isotope mixing model in the IsotopeR package (Hopkins and Ferguson 2012). The model runs three MCMC chains with a burn-in of 1,000 draws followed by 10,000 draws from the posterior. For all the models, measurement errors based on my data and discrimination factors were included. I used discrimination factors for hairs in accordance with reports by Mowat et al. (2017) and Hopkins et al. (2017, 2021): $\Delta^{13}\text{C} \pm \text{standard deviation (SD)}$ is $2.7 \pm 0.1\text{‰}$ for C_3 plants (alpine plants and plants), $1.4 \pm 0.5\text{‰}$ for C_4 plants (corn), and $0.0 \pm 0.2\text{‰}$ for animal matter (Hopkins et al. 2021) and $\Delta^{15}\text{N} \pm \text{SD}$ is $2.4 \pm 0.2\text{‰}$ for plant matter and $3.9 \pm 0.3\text{‰}$ for animal matter (Kurle et al. 2014). The value of $\Delta^{34}\text{S}$ of food items was calculated from a regression equation provided by Florin et al. (2011): $\Delta^{34}\text{S} = 1.16 - 0.26 \times \delta^{34}\text{S}$.

Statistical analysis

To examine the factors that affected the three stable isotope ratios, I used linear mixed-effect models, which were implemented using the 'lmer' function in the R package lme4 (Bates et al. 2015). In the linear mixed-effect models, which followed a Gaussian distribution, $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ were used as response variables, and the capture area, sex, age, and their interactions were used as explanatory variables. To account for the fact that the diets of brown bears are potentially affected by annual fluctuations in food resources, I included year as a random effect in all the models. I tested all possible combination of the explanatory variables and selected the best model based on the Akaike information criterion (AIC). Then, I specifically analyzed the variation in the proportional dietary contributions using the Dirichlet regression, a multivariate extension of the beta regression. The Dirichlet regression is appropriate for modeling response variables that represent continuous proportions to any number of categories (Douma and Weedon 2019). I implemented the Dirichlet regression using the explanatory factors that were included in the best model selected among the initial linear mixed-effect models. As mixed-effect models for Dirichlet regression have not been implemented in commonly used software yet (Douma and Weedon 2019), I was unable to include random effects in this analysis. We used the 'DirichReg' function in the R package DirichletReg (Maier 2014).

To quantify dietary changes from natal diet, I calculated the dissimilarity distance between the diets of bears at the time of capture and the average diet of their birth area. Since females have a small home range

(average $26.5 \text{ km}^2 \pm 1.8 \text{ SE}$; Kohira et al. 2006), the diets of females were considered representative of the diet in the capture area. A mean of the three stable isotope ratios for all females captured in each area was used as the average diet in each area. Dietary changes were calculated as the distance between two points in a three-dimensional space including $\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$ values. First, a single regression analysis was performed to examine the correlation between age and dietary change in each birth area. Next, I compared the mean values of dietary changes by sex and age class using the Kruskal-Wallis rank sum test with Steel-Dwass nonparametric post hoc comparisons. Then, I examined correlations between dietary change and the dietary contributions of marine animals using bivariate regression analysis to investigate how the diet shifted.

Ethics statement

Brown bear hair samples were collected as part of a joint research agreement between the Shiretoko Nature Foundation and the Hokkaido University under the Brown Bear Management Plan for the Shiretoko Peninsula, based on a consensus among the Kushiro Nature Conservation Office of the Ministry of the Environment, Hokkaido Regional Forest Office, Hokkaido government, Shari town, Rausu town, and Shibetsu town (formulated in 2012, revised in 2017). The capture of brown bears for the purpose of scientific research was conducted in accordance with the Guidelines for Animal Care and Use of Hokkaido University, and all procedures were approved by the Animal Care and Use Committee of the Graduate School of Veterinary Medicine, Hokkaido University (permit numbers 1152, 15009, and 17005), and by the Hokkaido University Animal Care and Use Committee (18-0083). Plant samplings were approved by the Hokkaido Regional Environmental Office and the Abashiri Southern Forest Management Station (permit numbers 1908131 and 2005201).

3.3 Results

Factors influencing diet

Stable isotope ratios were calculated for a total of 295 hair samples (129 females and 166 males, Table 3-1) and the values of the three stable isotope ratios in the bear hairs were highly variable throughout the peninsula (Table 3-1 and Fig. 3-3). The mean isotopic values of bear food items are shown in Appendix S3-3.

Regional variations in the diets were relatively clear among females. The diet of the females captured

in the SNP area mainly consisted of marine animals (Fig. 3-4). The mean proportional contribution of marine animals was significantly higher in the SNP area than in the Utoro, Rausu, and Shari areas (Steel-Dwass test, all $p < 0.05$) (Appendix S3-2). The diet of the females captured in the Rausu area mainly consisted of alpine plants (Fig. 3-4). The mean proportional contribution of alpine plants was significantly higher in the Rausu area than in the other areas (Steel-Dwass test, all $p < 0.05$). The female diets in the Utoro and Shari areas were similar and mainly consisted of alpine plants and plants (Fig. 3-4). Although the number of samples collected from the Shibetsu area was small, the diets of females in this area were mainly composed of plants and terrestrial animals (Fig. 3-4). Dent corn contributed more than 10% to the diets of four individuals, three of which were captured near dent corn fields in the Shibetsu area.

Similar regional variation was also shown in the diets of males, but were not as clear as those in females. Male bear diets did not have significantly differing contributions of alpine plants (Steel-Dwass test, $p=0.24$) and marine animals ($p=0.14$) among those captured in the SNP and Rausu areas. Marine animals were abundant in the diets of several males, even in the middle and base regions, and individual variations were large with regard to the contribution of marine animals (Fig. 3-4). See Appendix S3-2 for detailed results of the comparisons of the mean contributions of each food category between areas.

Among the linear mixed-effect models for each stable isotope ratio, the models including the capture area, sex, and age had the lowest AIC values (Appendix S3-4). Therefore, these three factors were included in the Dirichlet regression model as explanatory variables. Age-related changes were most pronounced in the contribution of marine animals, and age was positively correlated with the contribution of marine animals in all regions (Fig. 3-5). The contribution of alpine plants was negatively correlated with the age, especially in the SNP and Rausu areas. The contributions of plants, dent corn, and terrestrial animals did not change with bear age (Fig. 3-5).

The effects of natal habitat on diet selection later in life

The birth areas of 245 bears (110 females and 135 males) with assigned mothers were estimated. I could not determine the mothers for the remaining 50 bears. Although I assumed that the mean hair stable isotope values ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$) of female bears captured in each region represented the average diet of the birthplace, it was uncertain whether the average diet in the Shibetsu area was properly represented because only four female samples were obtained from that area. Therefore, 14 bears (4 females and 10 males) born in the Shibetsu area were excluded from the subsequent analysis. I also excluded one male

born outside my study site. The following results were derived from the analysis of 106 females and 124 males whose mothers were identified and which were presumed to have been born in the SNP, Utoro, Rausu, or Shari area.

Significant positive correlations between age and dietary changes were found for the males born in the Utoro, Rausu, and Shari areas (all $p < 0.05$). On the other hand, no significant correlation was found among the males born in the SNP area ($p = 0.407$). Among females, there were no correlations between age and dietary changes in any of the birth areas (all $p > 0.1$; Fig. 3-6). The data for the bears born in the Utoro, Rausu, and Shari areas were combined since the relationships between age and dietary changes were similar for these bears. The average values differed significantly between physically mature adults and juveniles and sexually mature adults among the males born outside the SNP area (Steel-Dwass test, both $p < 0.05$), while there were no significant differences between age classes among the males born in the SNP area (Kruskal-Wallis rank sum test, chi-squared = 3.06, $p = 0.22$) or among the females (Kruskal-Wallis rank sum test: SNP, chi-squared = 4.33, $p = 0.12$; other areas, chi-squared = 0.61, $p = 0.74$) (Fig. 3-6).

For the males born in the Utoro, Rausu or Shari area, positive correlations were found between the contributions of marine animals to the diet and dietary changes (all $p < 0.05$) (Fig. 3-7a). For the males born in the SNP area, there was a significant negative correlation between dietary changes and the contributions of marine animals to the diet ($p = 0.016$, $R^2 = 0.14$, coefficient = -0.037); however, the plots showed two directional tendencies, namely, one group of individuals whose diets shifted to include a higher contribution from marine animals and another group of individuals for which marine animals contributed slightly less to their diets (Fig. 3-7a). Consumption of marine animals considerably varied among juveniles ($n=22$, range: 0.08–0.70) and sexually mature males ($n=13$, range: 0.15–0.65) but was high in all physically mature males ($n=5$, range=0.44–0.79). The mean dietary contributions of marine animals among physically mature males were ranked by birth area as follows: SNP (mean \pm SD = 0.59 ± 0.16 , $n=5$, age range: 10–16) > Rausu (mean \pm SD = 0.52 ± 0.16 , $n=6$, age range: 8–33) > Utoro (mean \pm SD = 0.45 ± 0.26 , $n=5$, age range: 9–28) > Shari (mean \pm SD = 0.38 ± 0.33 , $n=3$, age range: 8–15); however, the differences were not statistically significant (Fig. 3-7b, Kruskal-Wallis rank sum test, chi-squared = 2.25, $p = 0.52$).

3.4 Discussion

The results demonstrated that the dietary selection of solitary large mammals changes later in life, suggesting that dietary selection can be influenced by factors other than mother-offspring social learning during infancy. I focused on the consumption of high-energy natural foods, such as marine animals and alpine plants, to investigate the effects of natal habitat on diet selection in brown bears and how dietary preferences change as individuals grow. The results indicated that females maintained the average diet of their birth area even after reaching sexual and physical maturity, while the diets of males changed significantly after physical maturation (≥ 8 years old) (Fig. 3-6). Males consumed a higher proportion of marine animals at older ages regardless of their birth areas. Sex-biased dietary shifts might be driven by differences between the sexes in foraging experiences after independence. Males have larger home ranges and may have the advantage of acquiring foraging experiences in new habitats, which leads to changes in their food preferences, compared with those of females, which stay in their natal habitats and have smaller home ranges.

I initially expected that the diets of males would change immediately after independence due to habitat changes accompanied by natal dispersal, since brown bears are opportunistic predators (Bojarska and Selva 2012). However, the results indicated that the diets of some males significantly changed after more than 6 years of independence from their mothers. Such a dietary shift later in life might relate to asocial learning after independence. It is known that socially enhanced food preferences wane relatively quickly if animals are exposed to high-quality alternative foods (Galef and Whiskin 2001). Additionally, Pokrovskaya (2015) suggested that the food preferences of bears are developed by trial and error. Marine animals are certainly a high-quality food for brown bears; however, males that were born in the middle or base areas and did not learn to feed on marine animals while with their mothers may not easily acquire foraging experience for this prey item. Males may take several years to roam around the peninsula, discover an area with high availability of marine animals, and visit the area during the appropriate season. Individuals born at the base of the peninsula (i.e., in the Shari or Shibetsu areas) may need more time to discover the SNP area and its high availability of marine animals than those born in the middle of the peninsula (i.e., in the Utoro and Rausu areas). The physical distances between the birth areas and the SNP area may have led to the differences in the ultimate dependency of each male on marine animals (Fig. 3-7b). This argument is still conjectural, and therefore, further research on the dispersal pathways of males is needed.

On the other hand, the dietary shift showed different tendencies in males born in the SNP area; the diet of some individuals changed to consume more marine animals, while that of other individuals changed to consume fewer marine animals (Fig. 3-7a). A large individual variation in marine animal consumption was found among bears less than 7 years old. There are three possible reasons for lower marine animal consumption among some young males born in the SNP area: (i) the young males dispersed to areas with a low accessibility to marine animals, or they stayed in or near the SNP area even after natal dispersal, but (ii) they could not catch salmon because of underdeveloped hunting techniques, or (iii) they could not acquire marine animals because of interindividual competition with older individuals. Some studies on bears have shown that social dominance plays an important role in the acquisition of the most caloric food sources in a landscape, and older males usually have an advantage in the consumption of animal matter (e.g., Mowat and Heard 2006, Hatch et al. 2019, Naganuma et al. 2020). The results also showed a positive correlation between age and the contribution of marine animals to male diets (Fig. 3-5). If social dominance has the strongest influence on diet, young individuals should generally consume fewer marine animals; however, some young individuals consumed very large amounts of marine animals. Therefore, I believe that several factors, including foraging locations, hunting techniques, and social dominance, may combine to influence the individual variation in marine animal consumption at a younger age.

One limitation of the method used was that I could not be certain that the capture area was the foraging area. While 42% of the samples used in this study were hairs from the previous year, I do not have any information about the area in which each bear foraged in the year before capture. However, the dietary characteristics of female bears showed a clear regional change in the capture areas, which generally corresponded to the regional variations in food availability predicted from salmon spawning river distributions and vegetation features. This suggests that females had foraged at or near their capture areas. The annual home range of females inhabiting the Shiretoko Peninsula is small (Kohira et al. 2006), and daily foraging trips are assumed to be at most 3 km (equal to the radius of the home range). Therefore, it is unlikely that females used areas far from their capture areas as their primary foraging sites. On the other hand, the home range of males is much larger (Kohira et al. 2006), which is approximately 8–12 km in radius. Therefore, it is quite possible that males migrated to neighboring areas during the active season. The lack of coincidence between capture and foraging areas may be one reason why regional differences in the diets among males were more unclear than those among females and why some males that were captured outside the SNP area had very high levels of marine animal consumption.

I considered the Utoro and Rausu areas to provide similar geographical access to the stone pine zone; however, there was a large difference in the dietary contribution of alpine plants between bears captured in the Utoro area and those captured in the Rausu area, contrary to my expectations (Appendix S3-2). Alpine plants were dominant in the diet in the Rausu area, whereas the consumption of plants and alpine plants was approximately the same in the Utoro and Shari areas (Fig. 3-4). According to the vegetation map (Appendix S3-1), the Utoro and Shari areas are dominated by deciduous multilayered or single-layered forests with tall to intermediate height trees (*Abieti sachalinensis-Quercus crispula* and *Aceri glabri-Q. crispula* communities). In these communities, acorns (*Q. crispula*, etc.) and fruits (cherry, *Prunus sargentii*; wild vine, *Vitis coignetiae*; etc.) are abundant, and bears can consume these plant materials from spring to autumn (Shirane et al. 2021). On the other hand, the Rausu area is dominated by deciduous single-layered forests with short trees and relatively rich forest floor vegetation (*Betula ermanii* and *Sasa sp.-B. ermanii* communities). This means that herbaceous plants such as fuki (*Petasites japonicus*) and *Apiaceae* are abundant in the Rausu area, and bears can consume these plants from spring to early summer (Shirane et al. 2021). Stable isotope analyses estimate the proportional contributions to an assimilated diet and therefore does not represent the actual amount eaten. In addition, stable isotopes analyses of hair samples are thought to integrate the assimilation of protein, carbohydrate, and lipids (Mowat and Heard 2006). Acorns (a source of lipids, Naves et al. 2006), fruits (a source of carbohydrates, Hertel et al. 2018), and pine nuts (a source of lipids, López-Alfaro et al. 2015) contribute more to the assimilated diet because these food items are more digestible and absorbable for brown bears than herbaceous plants (e.g., Dahle et al. 1998, Persson et al. 2001, Bojarska and Selva 2013, López-Alfaro et al. 2015). Perhaps, this was the reason why the contribution of plant categories was higher in the Utoro and Shari areas and that of alpine plants was higher in the Rausu area. I am also aware that crops (beets and wheat) might provide some valuable food for bears in the Utoro and Shari areas, especially in the summer when food is scarce. Although beets and wheat were excluded from the present analysis because they could not be distinguished from natural plants by the isotopic signatures, they may have contributed to a greater proportion of natural plants in the diet of some areas.

Bears attempt to regulate total protein, lipid, and carbohydrate intake to maximize dietary efficiency and growth (Robbins et al. 2007, Erlenbach et al. 2014). A large body size has advantages in reproduction and hibernation for both males and females. Marine animals (i.e., salmon and marine mammals) are a source of lipids and protein and are the highest energy food for brown bear population on the Shiretoko

Peninsula. The dietary shift observed in males emphasizes that the use of marine animals can improve their fitness. However, for some individuals with extremely high marine animal consumption (maximum contribution of marine animals was estimated to be 0.82), it is questionable whether such feeding habit is optimal in terms of the nutritional balance. Pine nuts are the second highest energy food after meat, which provide nearly optimal levels of protein and lipids (Costello et al. 2016). It is also known that bears tend to avoid inter- and intra-specific competition by changing their dietary niche if alternative food resources, such as pine nuts and fruits, are sufficiently available (e.g., Mowat and Heard 2006, Belant et al. 2006, Fortin et al. 2007). The positive correlations between age and marine animal contribution and the negative correlations between age and alpine plant contribution observed in the SNP and Rausu areas (Fig. 3-5) suggest that alpine plants serve as an alternative or complimentary food source in areas where the availability of marine animals is moderate to high.

The correction procedure of stable isotope ratio values based on hair growth stages used in this study can be applied to ecological studies of bears in other regions with some notes. Hairs are metabolically inert after formation and thus preserve the isotopic record of an animal's diet during the hair growing period (Tieszen et al. 1983, Ayliffe et al. 2004). Although the correction procedure used in this study was based on the average trend for isotopic values of whole guard hair to continue increasing during the hair growing periods, it should be noted that the fluctuation in isotopic values of an individual does not necessarily match this trend. Therefore, I expect that this correction would not inaccurate inference when examining changes in the diet by area or age, as in this study, but it may not be appropriate for studying details of the diet of an individual. Although using only fully grown hairs collected before a molt has been a common practice in previous analyses (e.g., Jones et al. 2006, Adams et al. 2017, Careddu et al. 2021), the inability to use hairs in the growth stage is inefficient and a severe limitation to collecting a sufficient sample size in wildlife species such as bears, where opportunities to collect hairs are restricted. The correction procedure adopted in this study offers a novel approach that expands the types of hair samples that can be used for analysis and can be applied to ecological studies using hair biomarker analysis not only for bears but also for a variety of wildlife.

Conclusion

My prediction that dietary selection by large solitary mammals may change later in life was supported for male bears. While the results support the generally accepted hypothesis that social dominance is an

advantage in the ability to acquire high-energy food, I also observed individual variation that cannot be explained by social dominance alone. I believe that post-independence foraging experiences (i.e., the exploration of available foraging habitats and improved hunting techniques) may play a role in dietary shifts, and the results do not contradict this prediction. I examined age-related dietary shifts by correlating dietary changes from the average diets of the birth areas of bears with the age of each individual. This can be a useful indicator for studying the development of diet selection in free-ranging animals, for which it is difficult to track individual dietary shifts continuously. The results suggested that the development of dietary preferences after dispersal varies depending on natal habitat and sex and hence deserves more attention.

Table 3-1. Numbers of samples (individual bears, n) and carbon ($\delta^{13}\text{C}$), nitrogen ($\delta^{15}\text{N}$), and sulfur ($\delta^{34}\text{S}$) stable isotope ratios (mean \pm SD) in brown bear hairs for each capture area.

Capture area	n	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	$\delta^{34}\text{S}$ (‰)
Female	129	-23.31 \pm 1.09	3.85 \pm 2.78	11.41 \pm 3.19
SNP	19	-22.11 \pm 1.05	7.05 \pm 3.72	15.43 \pm 2.51
Utoro	38	-23.53 \pm 0.92	3.53 \pm 2.25	10.99 \pm 2.98
Rausu	31	-23.23 \pm 0.88	2.90 \pm 2.40	12.00 \pm 2.43
Shari	37	-23.81 \pm 0.71	3.15 \pm 1.66	9.58 \pm 2.23
Shibetsu	4	-23.02 \pm 2.14	5.37 \pm 1.40	8.59 \pm 1.28
Male	166	-23.02 \pm 1.31	5.08 \pm 3.17	11.85 \pm 3.57
SNP	12	-21.82 \pm 0.80	7.38 \pm 3.72	15.68 \pm 2.10
Utoro	31	-23.22 \pm 1.30	4.76 \pm 3.11	12.33 \pm 3.38
Rausu	48	-22.68 \pm 1.20	4.71 \pm 3.30	13.62 \pm 2.39
Shari	55	-23.32 \pm 1.25	4.64 \pm 2.73	9.74 \pm 3.44
Shibetsu	20	-23.41 \pm 1.36	6.28 \pm 2.77	10.34 \pm 2.85
Total	295	-23.15 \pm 1.23	4.54 \pm 3.07	11.66 \pm 3.42

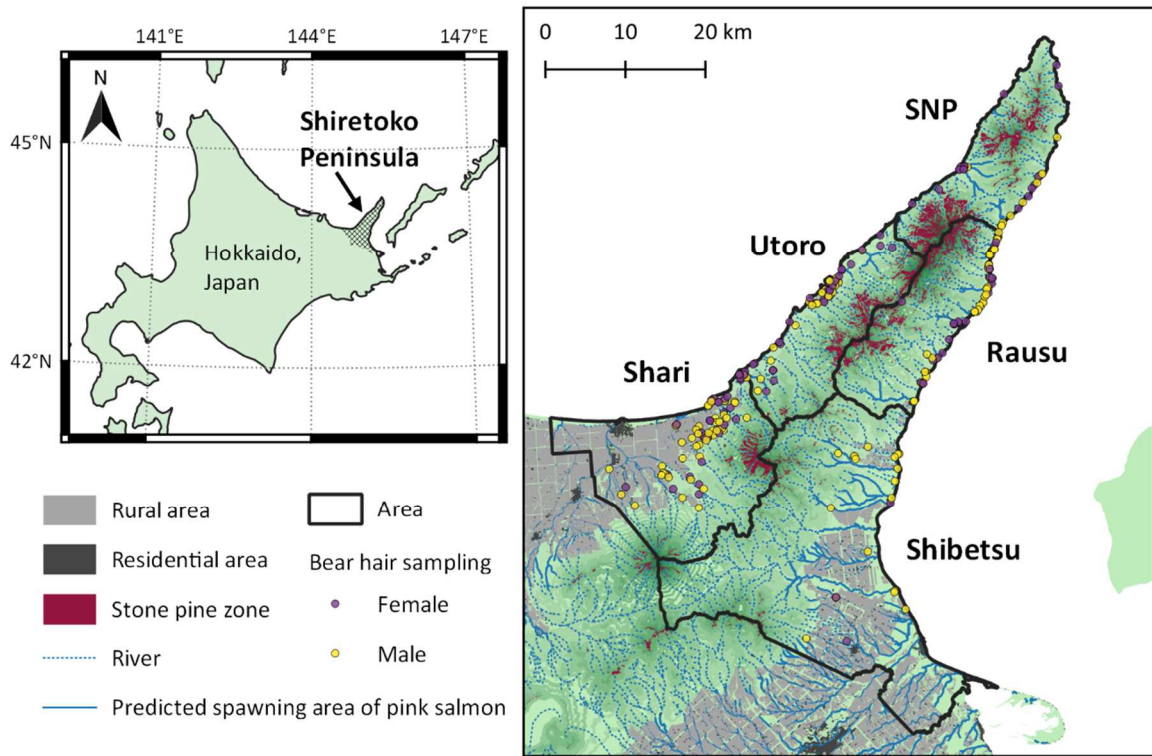


Figure 3-1. Map of the Shiretoko Peninsula. The whole peninsula was divided into five areas for the analysis: SNP, Utoro, Rausu, Shari, and Shibetsu. The stone pine zone is *Pinus pumila* zone in the vegetation map.

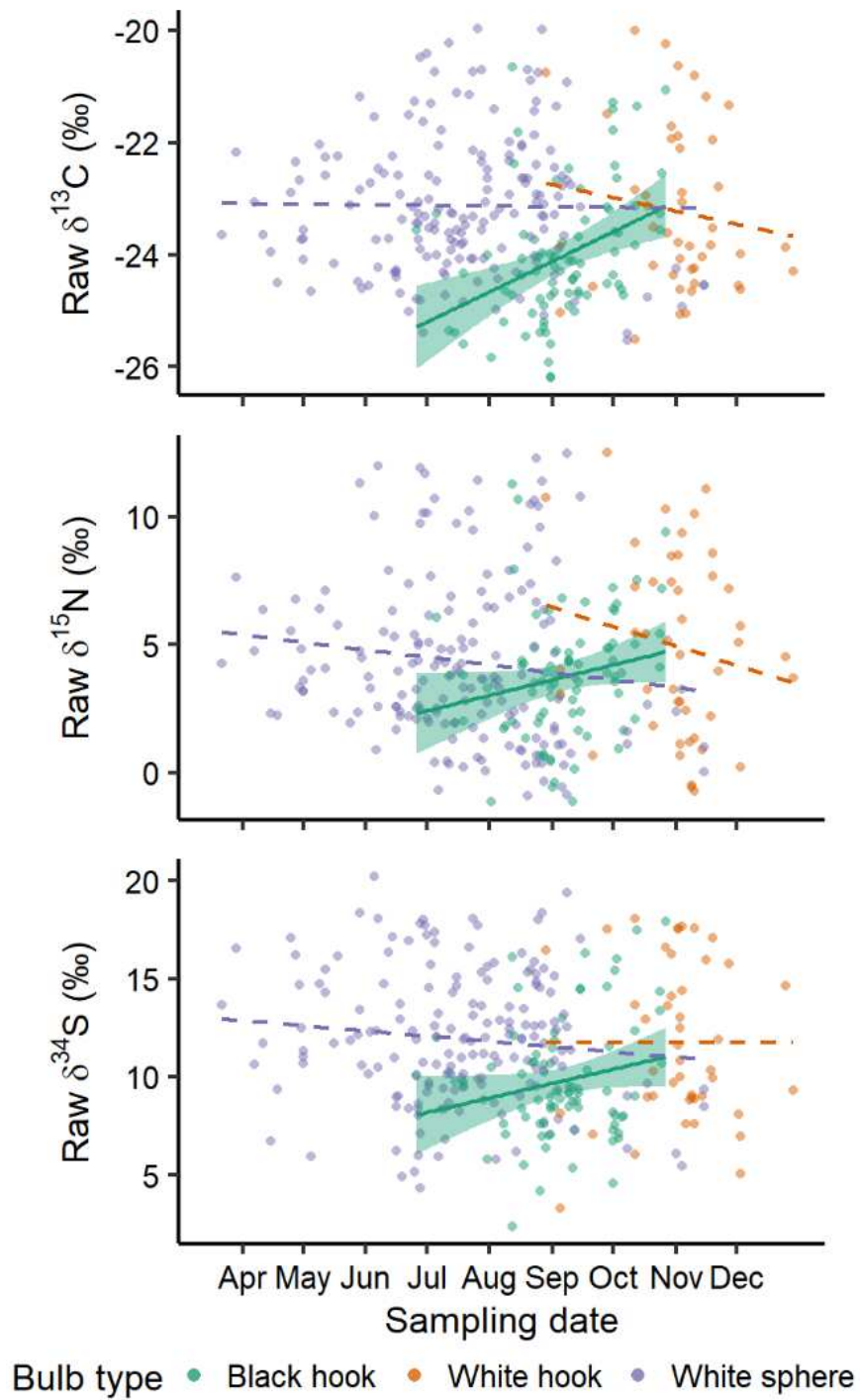


Figure 3-2. The relationship between sampling date (day of year) and isotopic values of whole guard hairs of each bulb type. The correlations that were significant ($p < 0.05$) are shown as solid lines with 95% CIs, and those that were not significant are shown as dashed lines.

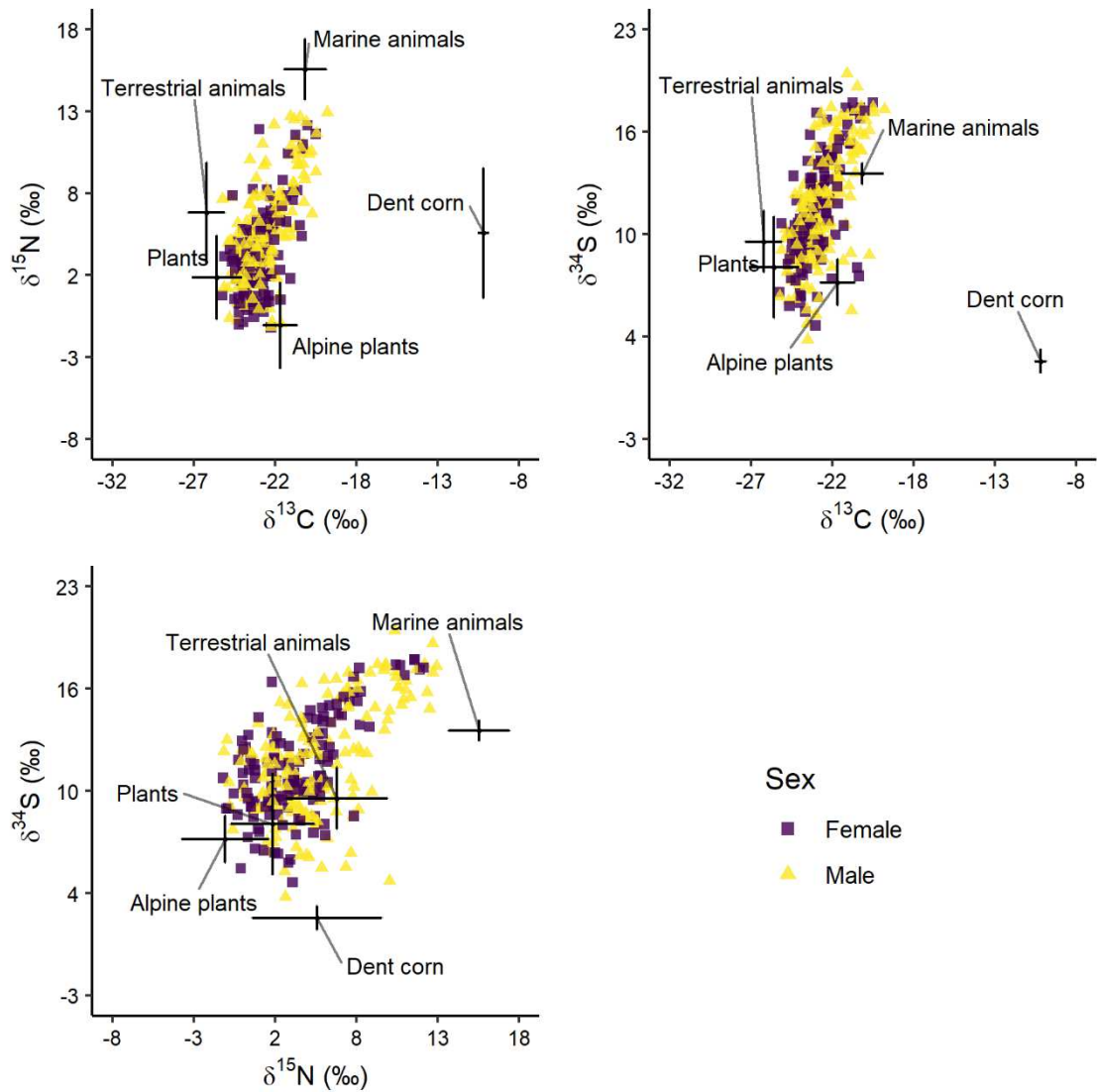


Figure 3-3. Carbon ($\delta^{13}\text{C}$), nitrogen ($\delta^{15}\text{N}$), and sulfur ($\delta^{34}\text{S}$) stable isotope ratios in brown bear hairs (130 females and 167 males) and potential food items (mean \pm SD) from the Shiretoko Peninsula, Hokkaido, Japan. Isotopic signatures of food items were corrected for tissue-diet discrimination.

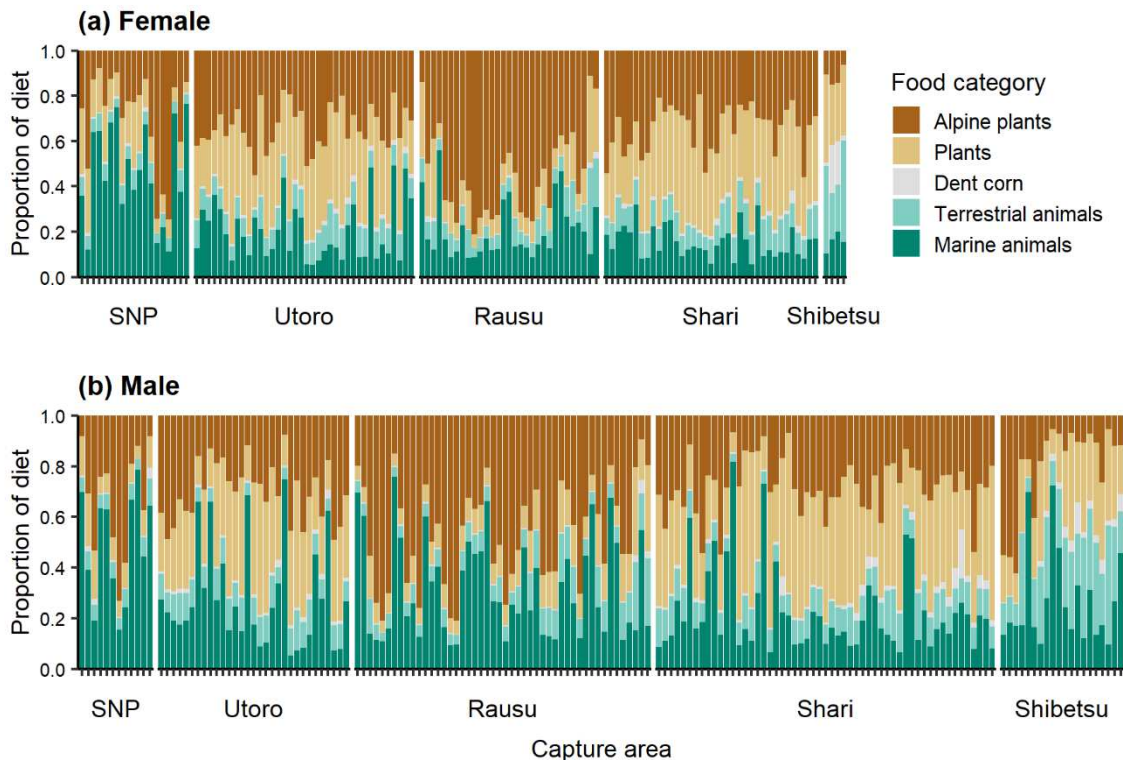


Figure 3-4. The proportional contributions estimated by IsotopeR stable isotope mixing models of five food categories to the diets of brown bears captured in each area of the Shiretoko Peninsula. Each bar represents the diet of an individual bear: (a) shows females and (b) shows males.

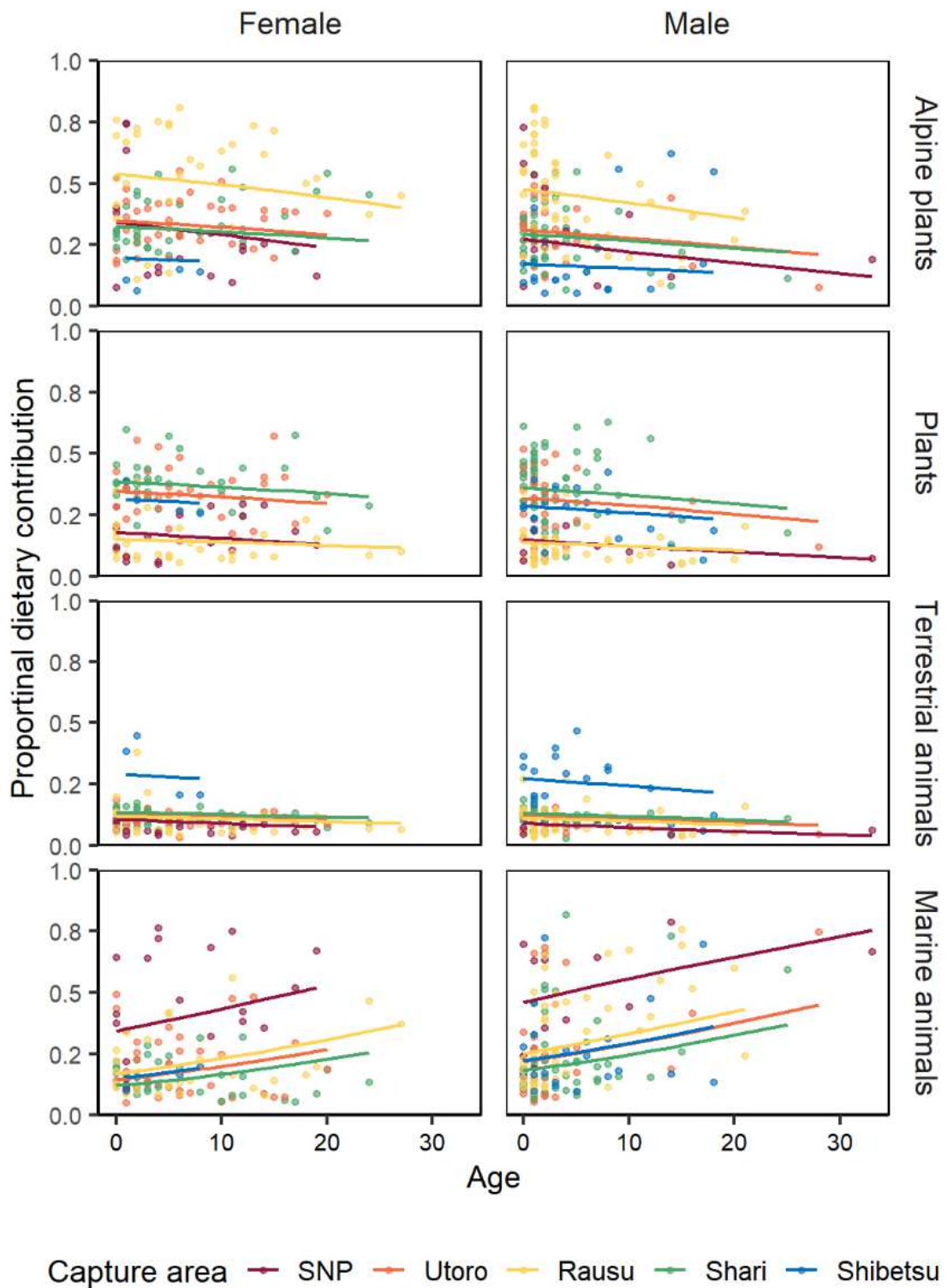


Figure 3-5. Age-related dietary shift in each area, as predicted by the Dirichlet regression model. The horizontal axes indicate the bear age, and the vertical axes represent the proportional dietary contribution of each food category. The contribution of dent corn was little for all bears and was not shown here.

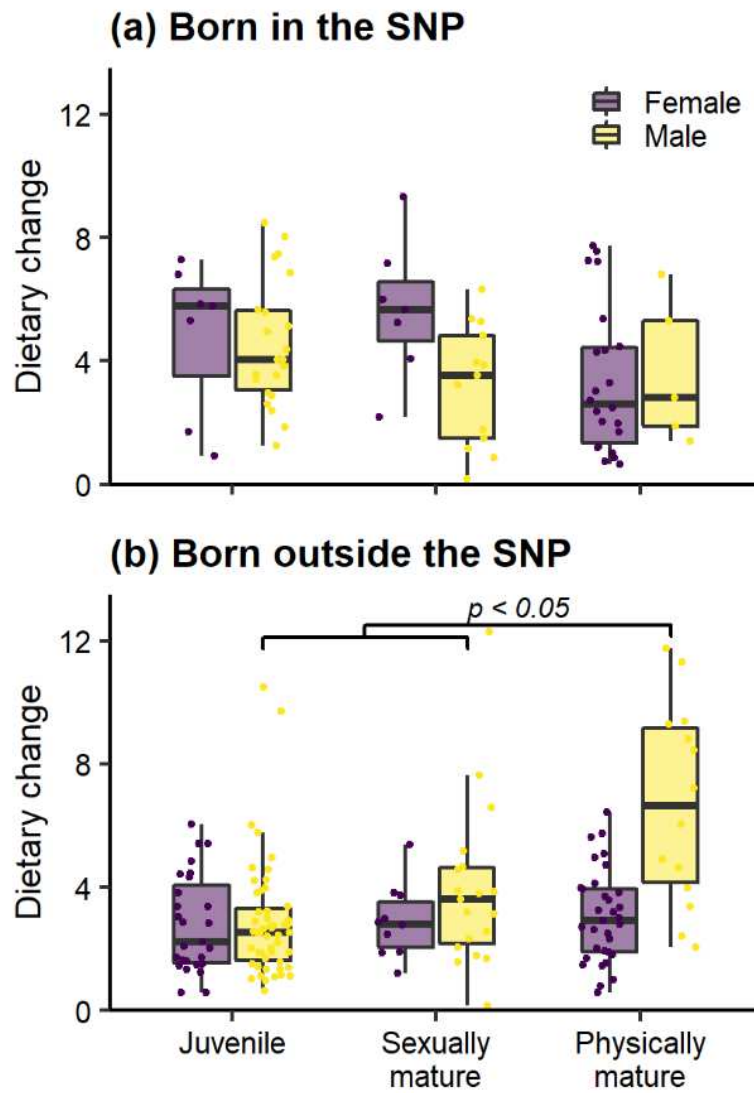


Figure 3-6. Dietary changes from the average diet of the birth area by age class. Only the diets of males born outside the SNP area were significantly changed after physical maturity.

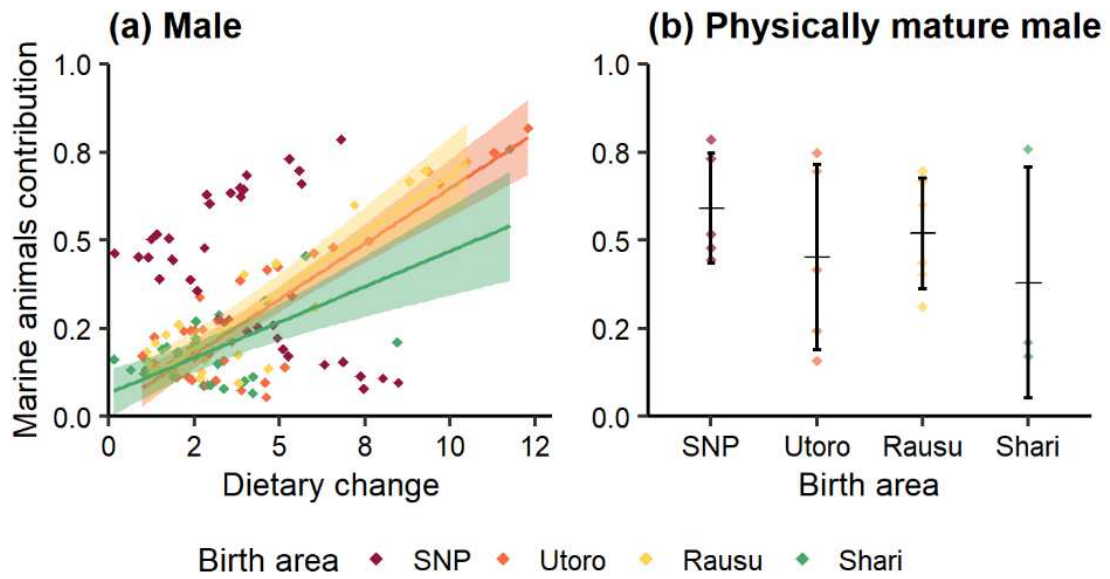
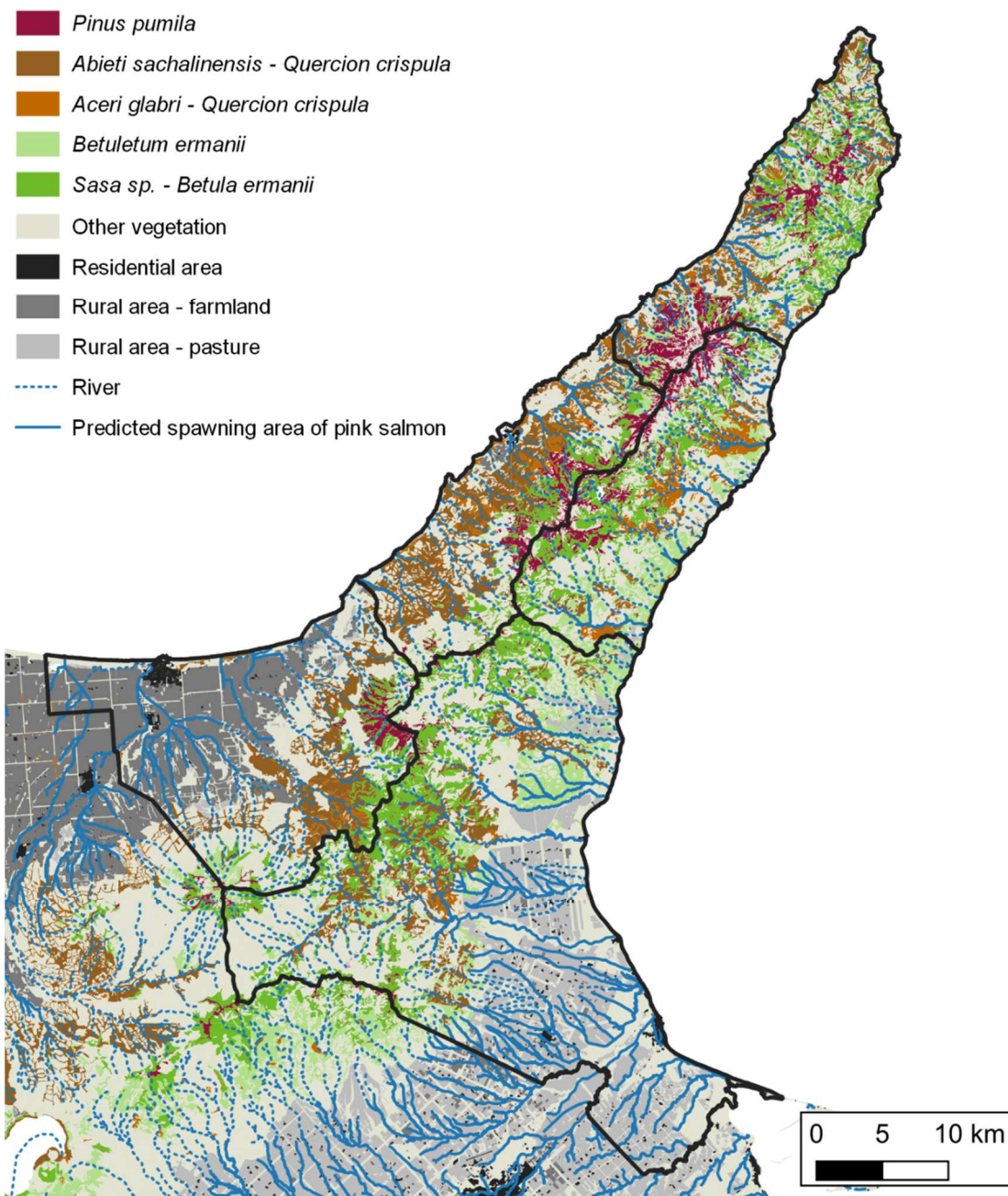


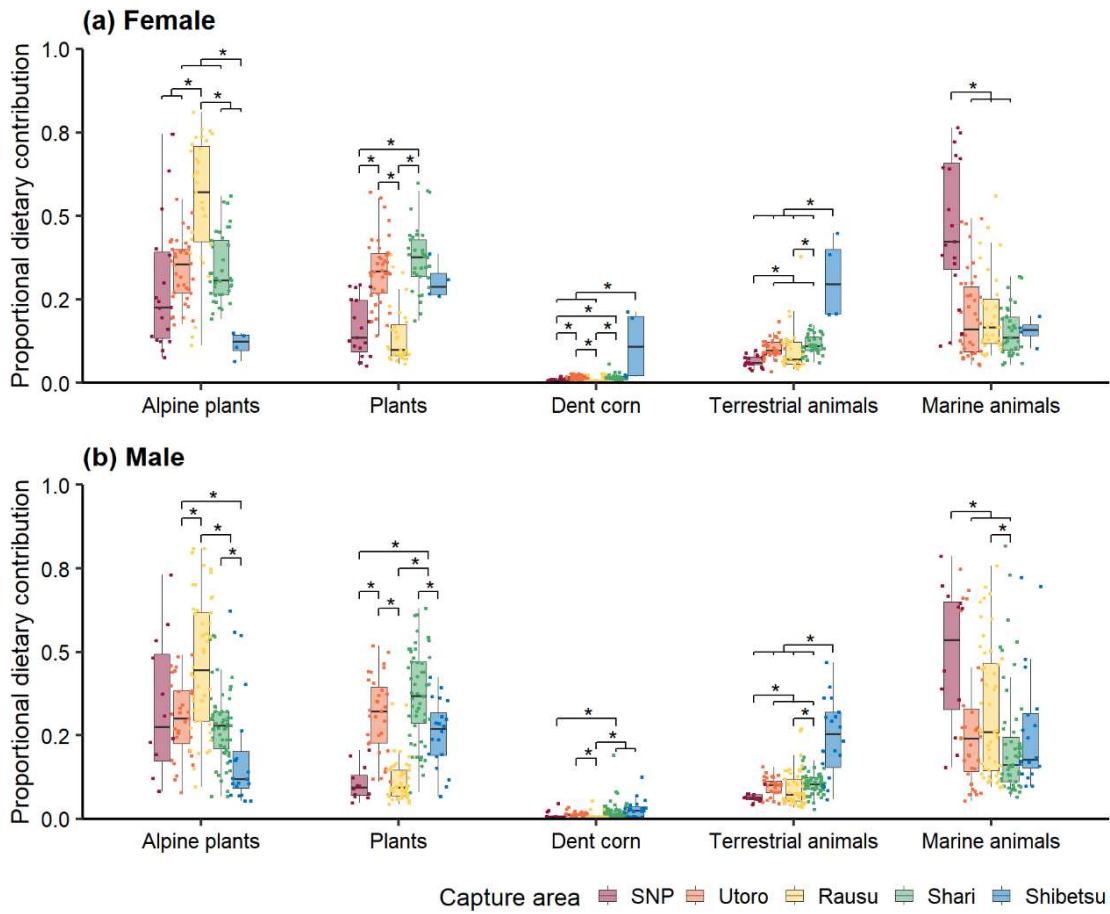
Figure 3-7. (a) The relationship between dietary change and dietary contributions of marine animals to the diets of males by birth area. Single regressions that were significant ($p < 0.05$) are shown as solid lines with 95% CIs. Although a statistically negative correlation was found for the tip area, this pattern is not graphically shown because it differs from the two directional tendencies of the plot. (b) Estimated value (rhombus) and mean \pm SD of the dietary contributions of marine animals to physically mature males by birth area. The vertical axis represents marine animal dietary contributions, and the horizontal axis represents birth area.



Appendix S3-1. The four typical vegetation types of the Shiretoko Peninsula (*Abieti sachalinensis*–*Quercion crispula* community, *Aceri glabri*–*Quercion crispula* community, *Betula ermanii* community, and *Sasa sp.*–*Betula ermanii* community) and *Pinus pumila* community are shown in different colors. The vegetation map was created and processed by the authors using the 1:25,000 GIS-based vegetation map "Hokkaido" from the "The 6-7th National Survey on the Natural Environment: Vegetation Survey (2000–2012)" (Biodiversity Center of Japan, Ministry of the Environment, Japan; <http://gis.biodic.go.jp/webgis/sc-025.html?kind=v67>). The river distribution was created and processed

by the authors using "National Land Numerical Information (River Data, Hokkaido)" (Ministry of Land, Infrastructure, Transport and Tourism, Japan; <https://nlftp.mlit.go.jp/ksj/gml/datalist/KsjTmplt-W05.html#prefecture01>).

Since pink salmon run upstream more than chum salmon in the same river (Hokkaido Government 2013), I used the map of predicted the spawning area of pink salmon for the proper division of the study site in terms of accessibility to salmon for bears. Although the upper limits of salmonid runs are often described by elevation and distance from the sea, I used elevation as a threshold because the Shiretoko Peninsula has a narrow and steep topography. Based on a survey of pink salmon on the Shiretoko Peninsula (Komiyama, unpublished), I assumed that an elevation of 200 m was a possible run for pink salmon. In addition, the locations of geographical barriers, such as waterfalls of 1.5 m or more, an extremely low water volume, and small pools, and artificial barriers, such as dams and Urai, were established through geographical information and interviews with local staff. The area downstream of these barriers was predicted to be the river section where pink salmon can run. The GIS data for the predicted salmon spawning river was created and provided by Mr. Tomoki Sakiyama, a former member of our research project (currently affiliated with the Graduate School of Environmental Science, Hokkaido University).



Appendix S3-2. Box plots of estimated bear diets for each capture area. The Kruskal-Wallis rank sum test with Steel-Dwass nonparametric post hoc comparisons was used to compare dietary contributions of each food category between capture areas, and significant differences ($p < 0.05$) are indicated by *.

Appendix S3-3. Stable isotope values and discrimination factors for potential food items of brown bears on the Shiretoko Peninsula, Hokkaido, Japan. All units are in ‰.

	n	Stable isotope value			Isotopic discrimination factor		
		$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{34}\text{S}$	$\Delta^{13}\text{C}$	$\Delta^{15}\text{N}$	$\Delta^{34}\text{S}$
Alpine plants	13	-24.79 ± 0.98	-3.19 ± 2.67	7.82 ± 1.93	2.7 ± 0.1	2.4 ± 0.2	-0.87 ± 0.50
<i>Empetrum nigrum</i>	2	-25.91 ± 0.23	-5.81 ± 0.34	9.18 ± 0.60	2.7 ± 0.1	2.4 ± 0.2	-1.23 ± 0.16
<i>Vaccinium uliginosum</i>	1	-24.49	-3.08	3.76	2.7 ± 0.1	2.4 ± 0.2	0.18
<i>Vaccinium vitis-idaea</i>	3	-24.64 ± 0.15	-5.65 ± 1.40	6.87 ± 1.41	2.7 ± 0.1	2.4 ± 0.2	-0.63 ± 0.37
<i>Pinus pumila</i>	6	-24.50 ± 1.24	-1.08 ± 2.16	8.48 ± 1.64	2.7 ± 0.1	2.4 ± 0.2	-1.04 ± 0.43
<i>Sorbus matsumurana</i>	1	-25.07	-3.32	8.07	2.7 ± 0.1	2.4 ± 0.2	-0.94
Plants	48	-28.53 ± 1.47	-0.15 ± 2.64	9.10 ± 4.34	2.7 ± 0.1	2.4 ± 0.2	-1.21 ± 1.13
<i>Actinidia arguta</i>	2	-29.26 ± 1.03	0.28 ± 0.24	6.22 ± 0.86	2.7 ± 0.1	2.4 ± 0.2	-0.46 ± 0.22
<i>Actinidia kolomikta</i>	1	-29.89	-3.19	5.34	2.7 ± 0.1	2.4 ± 0.2	-0.23
<i>Aegopodium alpestre</i>	2	-29.79 ± 0.20	2.16 ± 2.41	10.94 ± 2.85	2.7 ± 0.1	2.4 ± 0.2	-1.68 ± 0.74
<i>Angelica genuflexa</i>	2	-27.51 ± 0.58	1.39 ± 0.11	9.05 ± 0.24	2.7 ± 0.1	2.4 ± 0.2	-1.19 ± 0.06
<i>Angelica sachalinensis</i>	1	-29.46	5.04	11.32	2.7 ± 0.1	2.4 ± 0.2	-1.78
<i>Coelopleurum gmelinii</i>	4	-27.81 ± 0.70	0.02 ± 1.72	11.32 ± 2.87	2.7 ± 0.1	2.4 ± 0.2	-1.78 ± 0.75
<i>Heracleum lanatum</i>	4	-29.32 ± 0.87	0.78 ± 2.80	5.86 ± 4.66	2.7 ± 0.1	2.4 ± 0.2	-0.36 ± 1.21
<i>Ligusticum hultenii</i>	1	-25.48	2.67	13.75	2.7 ± 0.1	2.4 ± 0.2	-2.42
<i>Pleurospermum uralense</i>	2	-28.24 ± 0.02	2.79 ± 3.41	6.43 ± 4.41	2.7 ± 0.1	2.4 ± 0.2	-0.51 ± 1.15
<i>Aralia cordata</i>	4	-28.63 ± 1.22	-1.24 ± 0.91	13.46 ± 3.44	2.7 ± 0.1	2.4 ± 0.2	-2.34 ± 0.90
<i>Petesites japonicus</i>	3	-28.29 ± 1.91	0.91 ± 2.59	11.43 ± 5.31	2.7 ± 0.1	2.4 ± 0.2	-1.81 ± 1.38
<i>Quercus crispula</i>	4	-26.13 ± 0.96	-1.58 ± 0.95	9.32 ± 1.77	2.7 ± 0.1	2.4 ± 0.2	-1.26 ± 0.46
<i>Magnolia obovata</i>	1	-29.77	-3.06	8.40	2.7 ± 0.1	2.4 ± 0.2	-1.02
<i>Morus australis</i>	1	-30.18	0.69	11.50	2.7 ± 0.1	2.4 ± 0.2	-1.83
<i>Fallopia sachalinensis</i>	2	-28.30 ± 0.29	1.58 ± 4.43	2.91 ± 0.82	2.7 ± 0.1	2.4 ± 0.2	0.40 ± 0.21
<i>Cerasus sargentii</i>	1	-29.05	-1.61	7.34	2.7 ± 0.1	2.4 ± 0.2	-0.75
<i>Sorbus commixta</i>	4	-29.45 ± 1.33	-2.43 ± 1.38	5.20 ± 0.74	2.7 ± 0.1	2.4 ± 0.2	-0.19 ± 0.19
<i>Acer pictum Thunb.</i>	2	-28.53 ± 0.12	0.61 ± 0.75	12.60 ± 3.86	2.7 ± 0.1	2.4 ± 0.2	-2.11 ± 1.00
<i>Urtica platyphylla</i>	4	-29.23 ± 0.98	-1.97 ± 1.43	9.97 ± 5.88	2.7 ± 0.1	2.4 ± 0.2	-1.43 ± 1.53
<i>Vitis coignetiae</i>	3	-28.48 ± 1.54	-0.83 ± 2.81	9.43 ± 1.15	2.7 ± 0.1	2.4 ± 0.2	-1.29 ± 0.30
Corn (<i>Zea mays</i>)	3	-11.50 ± 0.28	2.68 ± 3.39	1.05 ± 0.86	1.4 ± 0.5	2.4 ± 0.2	0.89 ± 0.22
Terrestrial animals	7	-26.44 ± 1.01	2.46 ± 2.98	11.31 ± 2.44	0.0 ± 0.2	3.9 ± 0.3	-1.78 ± 0.63
<i>Bombus hypocrita sapporoensis</i>	2	-25.33 ± 0.75	-0.82 ± 1.71	9.68 ± 0.98	0.0 ± 0.2	3.9 ± 0.3	-1.36 ± 0.26
<i>Cervus nippon yesoensis</i>	2	-26.28 ± 0.12	6.53 ± 0.12	13.92 ± 2.29	0.0 ± 0.2	3.9 ± 0.3	-2.46 ± 0.60
<i>Lyristes bihamatus</i>	2	-26.85 ± 0.19	1.73 ± 0.50	11.81 ± 0.78	0.0 ± 0.2	3.9 ± 0.3	-1.91 ± 0.20
Ant of unknown species	1	-28.19	2.38	8.40	0.0 ± 0.2	3.9 ± 0.3	-1.02
Marine animals	6	-20.63 ± 1.15	11.56 ± 1.78	17.13 ± 0.86	0.0 ± 0.2	3.9 ± 0.3	-3.29 ± 0.22
<i>Phocoena dalli</i>	2	-19.60 ± 0.08	13.76 ± 0.17	17.08 ± 0.17	0.0 ± 0.2	3.9 ± 0.3	-3.28 ± 0.05
<i>Oncorhynchus gorbuscha</i>	2	-22.02 ± 0.80	9.97 ± 1.11	18.19 ± 0.10	0.0 ± 0.2	3.9 ± 0.3	-3.57 ± 0.03
<i>Oncorhynchus keta</i>	2	-20.26 ± 0.41	10.95 ± 0.70	16.11 ± 0.10	0.0 ± 0.2	3.9 ± 0.3	-3.03 ± 0.03

Appendix S3-4. Parameter estimate, standard error, t value, and the 95% confidence intervals (CIs) of three isotope ratios estimated by linear mixed-effect models. The capture area SNP was reference.

Model	Variable	Estimate	Std. Error	t value	95% CIs
$\delta^{13}\text{C}$	(Intercept)	-22.57	0.23	-99.98	-23.03, -22.14
	Capture area (Utoro)	-1.25	0.23	-5.35	-1.66, -0.82
	Capture area (Rausu)	-1.00	0.23	-4.37	-1.43, -0.57
	Capture area (Shari)	-1.53	0.23	-6.78	-1.96, -1.07
	Capture area (Shibetsu)	-1.55	0.30	-5.13	-2.19, -0.94
	Sex (Male)	0.51	0.13	3.92	0.24, 0.76
	Age	0.05	0.01	4.86	0.03, 0.07
$\delta^{15}\text{N}$	(Intercept)	5.85	0.58	10.03	4.76, 6.96
	Capture area (Utoro)	-3.07	0.58	-5.28	-4.2, -1.87
	Capture area (Rausu)	-3.21	0.56	-5.69	-4.36, -2.07
	Capture area (Shari)	-3.35	0.56	-6.03	-4.41, -2.25
	Capture area (Shibetsu)	-1.94	0.75	-2.58	-3.44, -0.42
	Sex (Male)	1.67	0.32	5.21	1.05, 2.26
	Age	0.12	0.03	4.65	0.07, 0.17
$\delta^{34}\text{S}$	(Intercept)	14.76	0.56	26.41	13.61, 15.89
	Capture area (Utoro)	-3.93	0.61	-6.47	-5.06, -2.72
	Capture area (Rausu)	-2.74	0.60	-4.58	-3.99, -1.46
	Capture area (Shari)	-5.98	0.59	-10.17	-7.11, -4.77
	Capture area (Shibetsu)	-5.91	0.78	-7.59	-7.48, -4.33
	Sex (Male)	1.07	0.34	3.11	0.38, 1.74
	Age	0.06	0.03	2.00	0, 0.12

Chapter 4 | Triggers of human-bear conflicts: Insights from regional characteristics of diets and conflicts

4.1 Introduction

Conflicts between humans and bears (*Ursus*) occur in various places worldwide (Elfström et al. 2012, Can et al. 2014). In many human-bear conflicts, bears intrude into human residential areas to use anthropogenic foods (Can et al. 2014). Except for special scenarios such as human habituation (Shimozuru et al. 2020b), it is generally supported that food shortages are the cause of bears entering human areas (e.g., Elfström et al. 2012, Artelle et al. 2016). Interindividual competition for food is greater in low-quality habitats, and young bears and females with cubs are more likely to enter human areas, either to reduce or to avoid competition (Elfström et al. 2012). Consequently, these bears might incidentally use anthropogenic foods or learn to use them as cubs when accompanying their mothers into these areas (e.g., Mazur and Seher 2008, Hopkins 2013). Individuals who learn to use anthropogenic food are the most likely to become problem individuals that repeatedly intrude into human areas.

Researchers have proposed that stable isotope analysis of hair, liver, bone, and other bear tissues is a useful tool for determining their history of anthropogenic food use (Mizukami et al. 2005a, Narita et al. 2011, Bentzen et al. 2014, Sakiyama et al. 2021). Higher carbon isotope ratios indicate a diet that includes substantial portions of C₄ plants, such as corn. High carbon and nitrogen stable isotope ratios also indicate the use of garbage in some cases (Mizukami et al. 2005b). Bentzen et al. (2014) showed that the carbon and nitrogen stable isotope ratios of hairs from bears feeding on anthropogenic food were higher than those from bears feeding solely on natural foods. Other than the use of anthropogenic foods such as crops and garbage, problematic bear behaviors are associated with stable isotope values. For example, bears that risk close encounters with people can acquire high-quality large mammals and thus have higher nitrogen isotope values (Hobson et al. 2000, Careddu et al. 2021).

On the Shiretoko Peninsula, conflicts between humans and brown bears (*U. arctos*) have recently become a serious problem. There are several types of conflicts including crop damage, intrusion into residential areas, and encounters with residents and tourists. Bears are monitored under a management plan (Kushiro Nature Conservation Office of the Ministry of the Environment et al. 2017); field staff identify bears that enter human settlements based on their genetic and physical characteristics and evaluate their problem behaviors. If bears exhibit serious problem behaviors such as intentionally approaching

people, repeatedly damaging crops, or relying on anthropogenic food such as garbage, they are killed as a form of nuisance control. Other bears have been recorded entering human settlements for years, but they are only eliminated if they exhibit dangerous behavior. Based on the reports from the Shiretoko Nature Foundation (2021), the average number of brown bears killed for nuisance control (excluding hunting) in Shari town and Rausu town between 2000 and 2020 was 22 per year. The years 2012 and 2015 were extraordinary, as the number of bears killed for nuisance control was more than double the normal number. Shirane et al. (2021) analyzed the annual changes in the diet of brown bears inhabiting the Rusha area in Shiretoko National Park and found that they consumed less salmon and pine nuts in 2012 and 2015. They suggested that a simultaneous shortage of these resources, which are the main food sources for bears in summer, led to bear intrusions into residential areas (Shirane et al. 2021). However, the diets of brown bears within the peninsula exhibit regional variation (Jimbo et al. in press), and the number of human-bear conflicts each year also varies by region. These conflicts increased in Rausu town in 2012 and in Shari town in 2015. Such regional variations in human-bear conflicts might be related to regional variation in the feeding habits of brown bears.

In addition, human habituation has become a serious problem in Utoro, eastern Shari town and Rausu town, which are adjacent to the national park (Shimozuru et al. 2020b). Especially during the salmon spawning season in Utoro, bears can be seen hunting salmon near the river mouth regardless of the presence of tourists. Moreover, some bears feed garbage at the fishing spot. These brown bears are bold individuals and may thus more easily access highly nutritious food than brown bears that do not approach human areas.

In this study, I tested the following three hypotheses related to the occurrence of brown bears in human areas by analyzing the stable isotope ratios of hairs from brown bears inhabiting the Shiretoko Peninsula.

- (i) Bears that intrude into human residential areas consume more salmon than those that remain in the forest habitat.
- (ii) The diets of bears differ between Shari town and Rausu town, causing regional differences in bear intrusions.
- (iii) In Shibetsu town, the problematic bear that evaded capture for two years subsisted on dent corn.

4.2 Materials and Methods

Hair sample collection

I conducted stable isotope analysis on bear hair collected on the Shiretoko Peninsula (43°50′–44°20′N, 144°45′–145°20′E), eastern Hokkaido, Japan. Less-invasive hair trapping systems (hereafter hair traps) were used to collect bear hair in the forest region. These hair traps consisted of a single strand of barbed wire wrapped around a natural tree at a height of 30 to 230 cm above the ground at 30 cm intervals. To induce tree-rubbing behavior in brown bears, organic solvent (Creosote R, Yoshida refinery, Tokyo, Japan) was partially applied to the trunk of the tree. Hair traps were installed throughout the peninsula except in the central mountainous area, which was difficult to access. We established them at 63 sites in 2019 and 67 sites in 2020 (Fig. 4-1). In 2020, we removed 7 traps that performed poorly in 2019 and added 11 new sites. In both years, the traps were set up from May to October, and researchers visited them every two weeks to collect hair samples. The bundle of hairs entangled in one sting of barbed wire were regarded as a single sample. Each sample was stored in envelopes and kept frozen until it was used in the genetic analysis. After each collection, the wires were burned off to remove any leftover hairs. I monitored each trap with automatic cameras (HykeCam SP2, Hyke Co., Ltd, Asahikawa, Japan) that were placed in front of each trap tree (Fig. 4-2).

I used hairs collected during late May–August for the stable isotope analysis. To clarify regional variations in bear diet, I targeted female bears over 3 years of age that had a small home range (Kohira et al. 2006) and were independent of their mothers (Shimozuru et al. 2017). I used hairs from females that were confirmed to have offspring based on parentage analysis (Shirane et al. 2019) or camera images and were confirmed to be at least 4 years old at the time of hair collection. First, I classified the shape of the hair bulbs by gross observation according to Jimbo et al. (2020); then, I selected hairs with white sphere bulbs that had grown in the previous year of collection. From each sample, some hairs were used for genetic analyses by clipping 5 mm of hair root, including the bulb (Shirane et al. 2019). Genomic DNA was extracted and genotyped by using 21 microsatellite markers and one sex marker to identify individual bears (Shimozuru et al. 2017).

I assumed that the bundle of hairs entangled in one sting belonged to the same bear, but I did not include samples that contained a mixture of hairs of different colors, forms, or growth stages or samples that showed genetic contamination of multiple individuals in the stable isotope analysis. I also excluded hairs covered with organic solvents or tree sap. Approximately 10 to 30 guard hairs that were intact from root

to tip and were used in the stable isotope analysis. After removing their follicles, the hair samples were washed with a 2:1 chloroform:methanol (v/v) mixture according to a standard protocol for stable isotope analysis to remove contamination and surface lipids (Hobson et al. 2000) and then dried at 60 °C for 48 hours. Each hair sample was placed in a 2 ml reinforced tube (Precellys Lysing Kit, Bertin Technologies, France) and ground into fine powder using a ball mill (Shakeman 6, Biomedical Science, Japan) with 10 cycles at 4,350 rpm for 90 s after adding four ϕ 3.0 mm and one ϕ 5.0 mm zirconia ball bearings.

Stable isotope analysis

The carbon, nitrogen, and sulfur stable isotope ratios ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, and $\delta^{34}\text{S}$) of hair samples were measured following the same procedure as in Chapter 3. Ground hairs (0.8–1.2 mg) were weighed and enclosed in tin boats ($4 \times 4 \times 11$ mm) and were analyzed using an isotope ratio mass spectrometer coupled with an elemental analyzer (IsoPrime 100-vario MICRO cube, Elementar, Germany). Since carryover effects were observed in the measurement of sulfur isotopes, I took five measurements for one sample and adopted the latter three values as the measurement values. I used five amino standards (L-alanine: $\delta^{15}\text{N} = 1.79\text{‰}$ with $\delta^{13}\text{C} = -19.9\text{‰}$, $\delta^{15}\text{N} = 3.7\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, $\delta^{15}\text{N} = 4.99\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, $\delta^{15}\text{N} = 8.72\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, and $\delta^{15}\text{N} = 13.7\text{‰}$ with $\delta^{13}\text{C} = -19.6\text{‰}$, Shoko Science Co., Ltd., Japan) and three sulfur standards (silver sulfide: IAEA-S-1, IAEA-S-2, and IAEA-S-3, International Atomic Energy Agency, Australia) with $\delta^{34}\text{S} = -32.3\text{‰}$, -0.3‰ , and 22.7‰ as standard reference materials. Based on these measurements, calibration curves, consisting of straight lines for nitrogen and sulfur isotopes were constructed. All the analytical errors in this study were within 0.2‰ for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and 0.3‰ for $\delta^{34}\text{S}$.

Stable isotope data on potential food items of bears on the Shiretoko Peninsula (Chapter 3) were also used for the analysis. I estimated the dietary contributions of categories of food items for individuals using a stable isotope mixing model in the IsotopeR package (Hopkins and Ferguson 2012). The five food categories (alpine plants, plants, dent corn, terrestrial animals, and marine animals) were aggregated a priori according to the ecology of bear prey. Dent cone was not included in the analysis of bears sampled in Shari town, Rausu town and the national park because there are few dent corn fields in these areas. The model runs three MCMC chains with a burn-in of 1,000 draws followed by 10,000 draws from the posterior. For all the models, I included measurement errors (based on my data) and discrimination factors. The discrimination factors for hairs were used in accordance with reports by Mowat et al. (2017) and

Hopkins et al. (2017, 2021): $\Delta^{13}\text{C} \pm$ standard deviation (SD) is $2.7 \pm 0.1\text{‰}$ for C_3 plants (alpine plants and plants), $1.4 \pm 0.5\text{‰}$ for C_4 plants (corn), and $0.0 \pm 0.2\text{‰}$ for animal matter (Hopkins et al. 2021) and $\Delta^{15}\text{N} \pm$ SD is $2.4 \pm 0.2\text{‰}$ for plant matter and $3.9 \pm 0.3\text{‰}$ for animal matter (Kurle et al. 2014). The value of $\Delta^{34}\text{S}$ of food items was calculated from a regression equation provided by Florin et al. (2011): $\Delta^{34}\text{S} = 1.16 - 0.26 \times \delta^{34}\text{S}$. The procedure for this analysis also followed Chapter 3.

Statistical analysis

I used the Kernel Density Estimator to create a spatial map of bear diets through the heatmap plugin in Quantum GIS (QGIS) software ver. 3.4.7. The estimated contribution of alpine plants and marine animals to the diet of each bear was used as a weight parameter to visualize dietary variation in relation to location. I selected 6 km as a radius parameter that determined the range of influence of each point, as the average home range of female brown bears on the Shiretoko Peninsula is $26.5 \text{ km}^2 \pm 1.8 \text{ SE}$ (Kohira et al. 2006), which can be approximated as a circle with a diameter of approximately 6 km. I assumed that females were foraging within a 6 km radius around the sampling point, taking into account the possibility that the sampling point might be at the edge of their home range. The estimation results were output as scaled values and displayed on a map using color ramps. Darker colored areas indicated a higher concentration of samples or greater consumption of each food. In addition, linear distances from the sampling sites to the nearest stone pine zone and predicted salmon spawning rivers (Appendix S3-1) were measured by using the NNJoin plugin in QGIS. Then, I plotted the relationship between the distance to each food resource and the contribution of alpine plants and marine animals to bear diets.

Data on the hair stable isotope ratios of 46 captured female bears were over 4 years of age and whose hairs with WS-type bulbs were referred to Chapter 3. Hereafter, I refer to female bears whose hairs were collected by hair traps as trapped female bears and those whose hairs were collected by nuisance control, hunting, or research capture as captured female bears. The three stable isotope ratios and the contribution of each food category were compared between sampling types using the Wilcoxon rank sum test in the R package `exactRankTests` (Hothorn and Hornik 2021).

Ethics statement

All procedures involved in hair trapping sample collection were conducted in accordance with the Guidelines for Animal Care and Use of Hokkaido University, and all procedures were approved by the

Hokkaido University Animal Care and Use Committee (permit numbers 19-0047). The installation of hair traps was approved by the Hokkaido Regional Environment Office and Hokkaido Regional Forest Office.

4.3 Results

Using hair traps, I obtained a total of 62 bear hairs belonging to 50 different female bears that were processed for stable isotope analysis (Table 4-1). In three areas except Shibetsu town, the contribution of alpine plants was higher in 2018 than in 2019 (dietary year) (Table 4-1), but significant annual changes were shown only in Shari town (Wilcoxon rank sum test, $p < 0.05$). The contribution of marine animals also tended to be higher in 2018 than in 2019 in both areas, but none of the differences were significant (Wilcoxon rank sum test, $p > 0.05$).

Spatial analysis showed that marine animals were consumed at high levels only in the national park area (Fig. 4-4). The consumption of alpine plants was not highly biased by region, with alpine plants contributing to bear diet in similar proportions from the tip to the base of the peninsula, except in Shibetsu town (Fig. 4-4). The relationship between the distance from the sampling site to the food resource and the dietary contribution is shown in Figs. 4-5 and 4-6. Most female bears were trapped close (< 6 km) to the stone pine zone (Fig. 4-5). The dietary contribution of alpine plants in trapped female bears in the national park was significantly higher closer to the stone pine zone ($p < 0.05$, $R^2=0.41$, Fig. 4-5). Although the relationship was not significant for trapped females in Shari town and Rausu town, the contribution of alpine plants tended to be higher closer to the stone pine zone in each area and in each year. In Shibetsu town, the contribution of alpine plants was low even at sites close to the stone pine zone. In addition, most trapped female bears were also very close (< 3 km) to the predicted salmon spawning rivers (Fig. 4-6). A weak positive correlation ($p < 0.05$, $R^2=0.23$) between the distance to the predicted salmon spawning rivers and the contribution of marine animals was observed in Shari town (Fig. 4-6). The contribution of marine animals varied widely among individuals trapped close to rivers, especially in the national park (Fig. 4-6).

In the area outside the national park in Shari town, $\delta^{13}\text{C}$ was significantly higher in the trapped bear hair (mean \pm SD; $-23.30 \pm 0.74\text{‰}$) than in the captured bear hair ($-23.92 \pm 0.81\text{‰}$) (Wilcoxon rank sum test, $p < 0.05$), whereas $\delta^{15}\text{N}$ ($p = 0.10$) and $\delta^{34}\text{S}$ ($p = 0.89$) did not differ significantly. In the area outside the national park in Rausu town, $\delta^{13}\text{C}$ ($p = 0.43$), $\delta^{15}\text{N}$ ($p = 0.08$), and $\delta^{34}\text{S}$ ($p = 0.31$) did not differ significantly between sampling types (trapped or captured bears). In both Shari town and Rausu town,

there were no statistically significant differences in the dietary contribution of each food category between sampling types (Wilcoxon rank sum test, all $p > 0.05$) (Fig. 4-7). In Rausu town, the mean contribution of alpine plants to the diet of captured female bears (mean \pm SD; 0.50 ± 0.13) tended to be higher than that of trapped female bears (0.41 ± 0.09). In Shibetsu town, I obtained dietary data from only seven trapped female bears. Since there were no hair stable isotope data from females over 4 years old with WS-type hair bulbs, I was not able to obtain dietary data for captured female bears in this town. Among the bears whose hairs were collected by the hair trap in Shibetsu town, the proportional dietary contribution of dent corn was estimated to be 0.009 for the lowest individual and 0.014 for the highest individual.

4.4 Discussion

Alpine plants consumed by brown bears on the Shiretoko Peninsula include wild berries such as lingonberry (*Vaccinium vitis-idaea*) and crowberry (*Empetrum nigrum*), in addition to pine nuts. However, these wild berries were rarely detected in a dietary analysis of scat in the Rusha area, whereas pine nuts were frequently detected (Shirane et al. 2021), indicating that the consumption of these wild berries is much lower than that of pine nuts. Therefore, the estimated dietary contribution of alpine plants in this study is considered to approximately reflect the contribution of pine nuts.

The hair stable isotope ratios of trapped female bears collected in 2019 and 2020 reflected their diets in 2018 and 2019, respectively. A survey on the abundance of pine nuts from Japanese stone pines reported that on average the stock was slightly more abundant in 2018 than in 2019 across the peninsula, although trends varied by region (Shiretoko Nature Foundation, unpublished). The results of hair stable isotope analysis showed that the consumption of pine nuts was higher in 2018 than in 2019, reflecting the annual resource fluctuations. Most individuals were trapped in close proximity to the high pine zone (< 6 km), and it was estimated that female bears in the national park, Shari town, and Rausu town consumed more pine nuts the closer they were to the stone pine zone. Therefore, these results suggest that resource abundance and the distance to resources directly affected the availability of pine nuts for female bears on the Shiretoko Peninsula except for Shibetsu town.

Unlike the results of Chapter 3, no regional differences in alpine plants consumption between Shari and Rausu areas were indicated. However, a comparison of the diets of captured and trapped female bears in Rausu town indicated that individual food preferences may influence the use of pine nuts. Most of the captured female bears in Rausu town included in this study (9 of 12) were killed in 2012; thus, their hair

stable isotope ratios reflected their diet in 2011. A survey on the abundance of pine nuts on Mt. Onnebetsudake (43°59'36"N, 145°00'48"E) and Rausu-daira (44°04'54"N, 145°07'42"E) near Rausu town reported that the estimated abundance was similar in 2011, 2018, and 2019, and there was a moderate level of fruiting in each of these three years (Shiretoko Nature Foundation, unpublished). However, in Rausu town, my results showed that the captured female bears consumed more pine nuts in 2011 than the trapped female bears consumed in 2018 and 2019. Trapped female bears consumed pine nuts and other plants in roughly equal proportions, whereas captured female bears consumed pine nuts at a higher proportion. These results suggest that the female bears captured in Rausu town in 2012 had a high preference for pine nuts. During summer, brown bears in the Shiretoko Peninsula consume pine nuts in July–September, the Sargent cherry (*Prunus sargentii*) in July, and Japanese bird cherry (*P. ssiori*) in August–September (Shirane et al. 2021). Pine nuts provide nearly optimal levels of proteins and lipids (Costello et al. 2016), and the use of pine nuts is suggested to be effective in maintaining body condition in summer (Shirane et al. 2021). However, since Japanese stone pine are distributed in the subalpine zone, focusing on foraging for pine nuts would reduce the opportunity for foraging for other plants, such as cherries, in the basal zone. Bears that are highly dependent on pine nuts during the summer might be more affected by scarce pine nuts resources, as occurred in 2012, compared to bears that also use fruits in a balanced manner. However, because pine nut abundance fluctuations show different local trends, it is possible that female bears in Rausu town encountered a very abundant pine nut stock in 2011. Future work is needed to compare the diets of captured and trapped bears sampled in the same year to take these factors into account.

In my analysis, both salmonids and marine mammals were included in the marine animals category. High-energy, high-quantity marine mammals are expected to greatly affect the stable isotope ratio of the individuals that consume them. However, because marine mammal strandings are localized and unpredictable, marine mammals are not as stable a food resource as salmon. Thus, I considered the use of marine mammals by brown bears as a special case and focus on the use of salmon in this discussion. Pink salmon (*Oncorhynchus gorbuscha*) resources were abundant throughout the peninsula in 2018 and poor in 2019 (Shiretoko Nature Foundation, unpublished). However, the results of hair stable isotope analysis did not show significant annual changes in the consumption of salmon by trapped female bears in any of the areas. Regional and individual differences in the consumption of salmon seemed to be greater than the annual changes. On the Shiretoko Peninsula, brown bears mostly use salmon in the national park. Although this analysis included only the diets of females older than 3 years, bear age (Jimbo et al. in

press), reproductive status (i.e., solitary, with cubs, or with yearlings), and other internal factors may be related to variation in female salmon consumption in each area. Female bears with cubs are known to occasionally change their foraging behavior (Dahle and Swenson 2003; Ben-David et al. 2004). Multiyear hair trapping, camera trapping, and genetic-based identification surveys are needed to determine the reproductive status of female bears in the year prior to hair sample collection.

For most trapped female bears, salmon spawning rivers were likely included in their home ranges (average 26.5 km², 3 km radius, Kohira et al. 2006); however, the contribution of marine animals varied widely among individuals. Because the predicted salmon spawning rivers are based on the upper boundary of the pink salmon run based on artificial and geographic barriers, elevation, etc. (Appendix S3-1), local stock conditions were not considered. For example, not only the annual abundance but also the possible run distances of salmon, which vary depending on river water levels during the run season, will have a significant impact on the availability for bears. In addition to stock status, it is also important to consider whether brown bears are actually able to access to the river. Even if a salmon spawning river is nearby, bears would not be able to access the river if it was surrounded by human settlements; conversely, bears may easily access the river in some locations even if it is far from the river. Notably, the three female bears were trapped in 2019 at a distance of approximately 7.5~11 km from predicted salmon spawning rivers, yet they were estimated to have consumed many salmon in 2018. All of these female bears were sampled adjacent to the upper Saruma River (western Shari town) and the upper Churui River (Shibetsu town), inland of the peninsula base. One of the three female bears was also trapped at the same trapping site in 2020, but she consumed very few salmon in 2019 (Fig. 4-6). As I mentioned earlier, salmon resources were abundant in 2018; in particular, the number of pink salmon runs was confirmed to be at one of the highest levels since 2010 in the Shari River, western Shari town (based on interviews from the Kitami Salmon Enhancement Program Association by the Shiretoko Nature Foundation, unpublished) and the Churui River, Shibetsu town (Churui River Salmon Fishing Report, 2019). Therefore, I expected that the availability of salmon for bears in the upper stream of the river was higher in 2018 compared to that in 2019, when the resource was poor. In western Shari town and Shibetsu town, most of the lower parts of the river are surrounded by residential or rural areas (Appendix S3-1). Therefore, for female bears living at the base of the peninsula, the availability of salmon may be affected by the abundance of salmon and whether or not salmon run far upstream.

The higher marine animal dietary contributions in captured female bears in the national park compared

to that in trapped female bears might be due to the sampling bias: 8 of the 10 captured female bears in the national park were captured alive for research purposes in the Rusha area. The Rusha area is an open coast with three rivers where pink salmon and chum salmon (*O. keta*) spawn naturally. Because in the last few decades, fishermen have used this land without excluding bears, the females inhabiting this area are highly habituated to humans (Shirane et al. 2020, Shimozuru et al. 2020b). In contrast, cautious adult males rarely appear along the coastline, most frequently at noon. Since the behavioral response of bears to the presence of humans varies among individuals (Aumiller and Matt 1994), the presence of people can act as a barrier to food competition between bears (Hobson et al. 2000, Careddu et al. 2021). In the Rusha area, human presence and human habituation might allow females and young bears to forage for salmon near the river mouth without being threatened by adult males. Since we targeted female bears frequently observed near the coastline for our research capture, these bears might be particularly dependent on salmon compared to other bears in the national park.

Since salmon consumption outside of the national park was not as high, the scarcity of salmon resources might not be a strong trigger for bears inhabiting these areas to intrude into human settlements. In eastern Shari town (i.e., the Utoro area), the availability of salmon for bears is predicted to be poor because salmon are inhibited by dams in most rivers from travelling upstream, and the river mouth areas are surrounded by residential areas. Therefore, I expected that the bears that repeatedly entered human areas would be able to consume more salmon near the river mouth than bears staying in the forests. However, the diets of captured and trapped female bears did not differ, indicating that even captured bears consumed natural foods in the forest until the year before they were captured. Behavioral strategies of bears living near human settlements include avoiding humans when resources are accessible at other times and locations or concentrating foraging activities when resource availability and quality are high (Rode et al. 2006). The results of this study suggest that bears that originally avoided people and consumed natural foods began to enter human areas for some reason (e.g., food scarcity or unintentional feeding). Thus, bear intrusion into human settlements might not be induced by individual foraging strategies but might be triggered by incidental events.

Female bears rarely consumed pine nuts in Shibetsu town even they were close to the stone pine zone; instead, plants dominated more than 50% of their diets. Since small stone pine zones are sparsely distributed in Shibetsu town, some dominant bears may occupy them or the foraging efficiency may be low. The abundance of fruits and herbs is likely to be more important for the female bears in Shibetsu

town than that of pine nuts. Therefore, future research should focus on the availability of fruits such as hard masts and cherries to investigate the effects of food resources on the human-bear conflicts in Shibetsu town.

In Shibetsu town, dent corn damage caused by brown bears and records of brown bears approaching the dent corn fields have been increased recently (Shibetsu town 2017). However, all the trapped female bears analyzed in this study had a very low dietary contribution of dent corn. Jimbo et al. (in press) reported only two females with a dent corn contribution greater than 0.1. Both of these bears were captured and killed in October around the dent corn fields in Shibetsu, indicating that they had used a high amount of dent corn in the year that they were captured (Jimbo et al. in press). These results suggest that bears are immediately eliminated if they use dent corn in this area, few individuals that have used dent corn for more than two years have not been captured. The use of anthropogenic foods is often inherited by cubs through mother-offspring social learning (Hopkins 2013; Morehouse et al. 2016); therefore, the immediate elimination of females that use anthropogenic food is effective in preventing the increase in problem individuals.

Notably, however, that this study did not assess each bear's dependence on beets and wheat. In Shari town, brown bear-caused crop damage is the highest for beets and wheat (Shari town 2021). However, natural plants, such as fruits and herbs, and beets and wheat could not be distinguished from the isotopic signatures (Jimbo et al. in press), so the values estimated as plant contributions reflect the consumption of both of these foods. The nitrogen stable isotope ratio of agricultural crops is influenced by fertilizer and becomes relatively high when manure or high amounts of fertilizer are applied (Bateman et al. 2005, Bateman and Kelly 2007). However, on the Shiretoko Peninsula, the nitrogen stable isotope ratio of plants in the riparian zone is also high because brown bears play a role in transporting marine-derived nitrogen to land by feeding on salmon (Koshino et al. 2013). Therefore, the stable isotope ratios of crops and natural plants on the Shiretoko Peninsula probably have a very wide range and overlap (Appendix S4-1). However, the sulfur stable isotope values of wheat tended to be lower than those of natural plants (Appendix S4-1), although the sulfur stable isotope values of beets were not measured. Therefore, it would be worthwhile to increase the number of wheat samples throughout the peninsula and examine the possibility of wheat classification by sulfur stable isotope ratios in the future. Bear intrusions into human areas and the use of agricultural crops by bears may be different scenarios. Further research is needed to understand bear use of beets and wheat.

The sulfur isotope ratio is considered a good indicator of the consumption of whitebark pine nuts by brown bears in the Greater Yellowstone Ecosystem, since sulfur stable isotope ratios of pine nuts are higher than those of other plants (Felicetti et al. 2003, Hopkins et al. 2017). However, the sulfur stable isotope ratios of plant matter varied so widely on the Shiretoko Peninsula, and the range of sulfur stable isotope values of Japanese stone pine nuts completely overlapped with those of other natural plants (Appendix S4-1). Sulfur isotope ratios on land are known to vary with the geological age; sulfur stable isotope ratios are higher in more newly formed areas (Thode et al. 1953). On the Shiretoko Peninsula, the formation age of the subalpine zone where Japanese stone pines are distributed and the lower elevation zone where herbaceous plants and fruits are distributed are not clearly separated (Appendix S4-2; based on data from the Seamless Geological Map of Japan V2 (GSJ, AIST)). This may explain why pine nuts and other plants could not be distinguished by sulfur stable isotope values. In addition, sulfur stable isotopes are known to be higher in marine organisms than in terrestrial organisms (Felicetti et al. 2003). On the Shiretoko Peninsula, brown bears play a role in transporting marine-derived nutrients to the land by foraging on upstream salmon (Koshino et al. 2013). As some herbaceous plants collected in the Rurua area, where salmon consumption is particularly high in the peninsula, had very high sulfur stable isotope ratios that were equal to those of marine animals (Appendix S4-2) may indicate that the transport of marine-derived sulfur by brown bears is involved in the fluctuation of sulfur stable isotope ratios in terrestrial organisms. Future comprehensive surveys of sulfur stable isotope ratios in peninsula soils could verify these theories.

Table 4-1. Numbers of hair samples (n) and the estimated proportional dietary contribution of each food category in trapped female bears, summarized by diet year. Bear hairs collected in 2019 and 2020 reflected the diets of 2018 and 2019, respectively.

Area	Dietary year	n	Alpine plants	Plants	Terrestrial animals	Marine animals	Dent corn
National Park	2018	11	0.34 ± 0.07	0.25 ± 0.11	0.02 ± 0.01	0.38 ± 0.14	-
	2019	11	0.28 ± 0.07	0.35 ± 0.09	0.03 ± 0.01	0.34 ± 0.11	-
Shari	2018	15	0.44 ± 0.10	0.30 ± 0.09	0.02 ± 0.00	0.24 ± 0.12	-
	2019	9	0.31 ± 0.05	0.44 ± 0.05	0.03 ± 0.01	0.21 ± 0.06	-
Rausu	2018	8	0.43 ± 0.07	0.33 ± 0.10	0.02 ± 0.01	0.22 ± 0.08	-
	2019	1	0.23	0.60	0.04	0.13	-
Shibetsu	2018	4	0.07 ± 0.01	0.56 ± 0.06	0.05 ± 0.01	0.31 ± 0.07	0.01 ± 0.00
	2019	3	0.07 ± 0.02	0.62 ± 0.04	0.06 ± 0.01	0.24 ± 0.05	0.01 ± 0.00
Total	2018	38	0.37 ± 0.14	0.32 ± 0.13	0.02 ± 0.01	0.28 ± 0.13	-
	2019	24	0.26 ± 0.09	0.43 ± 0.12	0.03 ± 0.01	0.27 ± 0.11	-

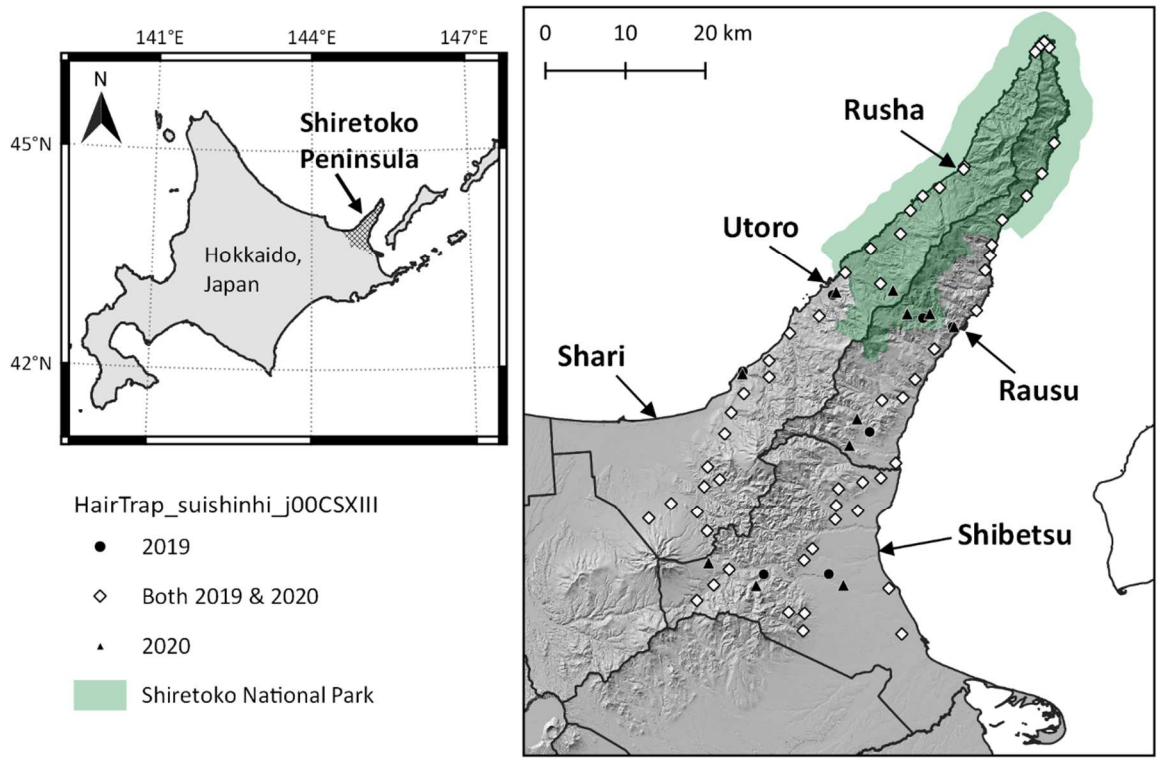


Figure 4-1. Locations of hair traps in 2019 and 2020 on the Shiretoko Peninsula, eastern Hokkaido, Japan.



Figure 4-2. A female bear rubbing on a hair trap tree as recorded by an automatic camera. This female had cubs, indicating that she was at least four years old.

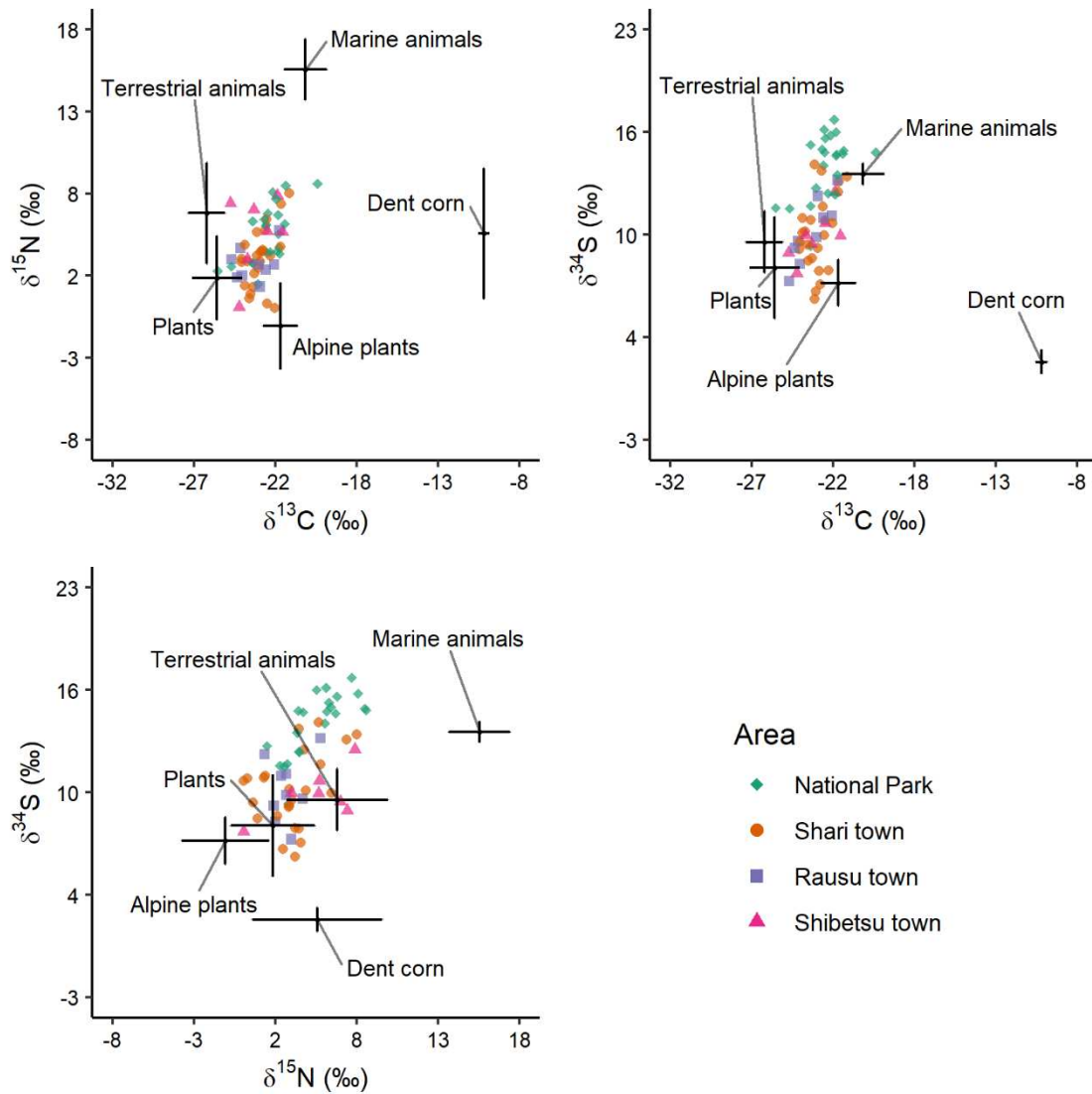


Figure 4-3. Carbon ($\delta^{13}\text{C}$), nitrogen ($\delta^{15}\text{N}$), and sulfur ($\delta^{34}\text{S}$) stable isotope ratios in trapped female bear hairs and potential food items (mean \pm SD) from the Shiretoko Peninsula, Hokkaido, Japan. Isotopic signatures of food items were corrected for tissue-diet discrimination.

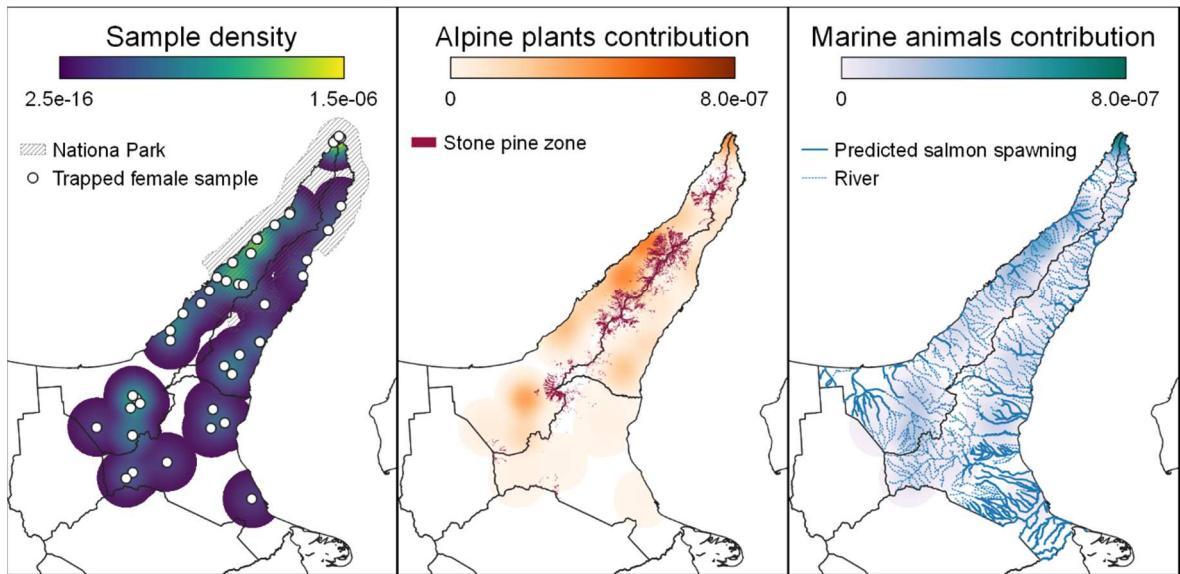


Figure 4-4. Spatial patterns of consumption of alpine plants and marine animals determined by weighted kernel density estimation in the trapped female bears on the Shiretoko Peninsula.

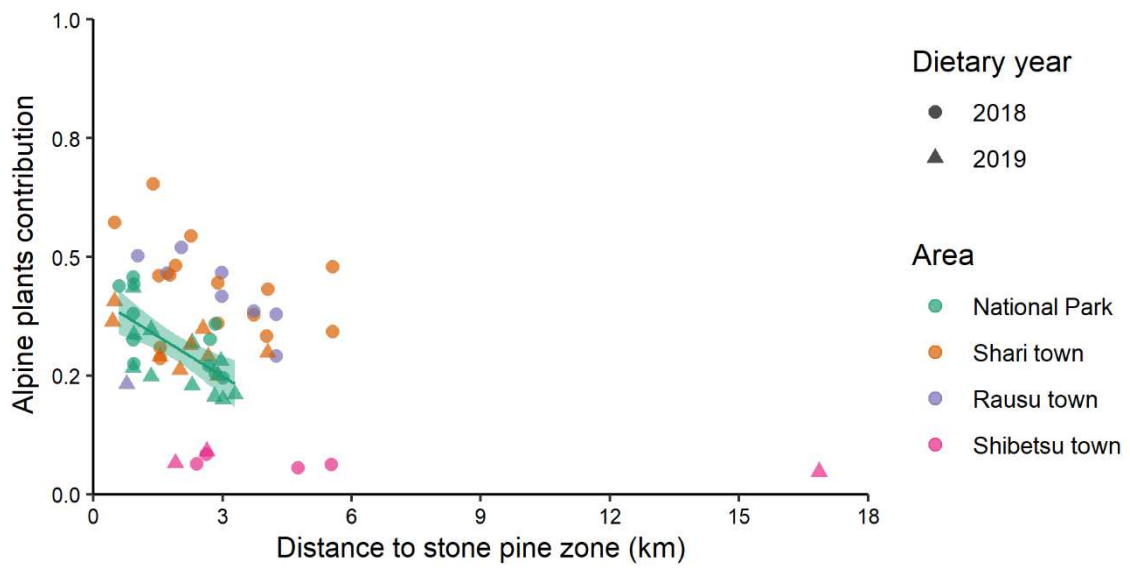


Figure 4-5. Relationship between the distance from the sample collection location to the nearest Japanese stone pine zone and the dietary contribution of alpine plants contribution in the trapped female bears. The significant linear regression is shown as a solid line with a 95% confidence intervals.

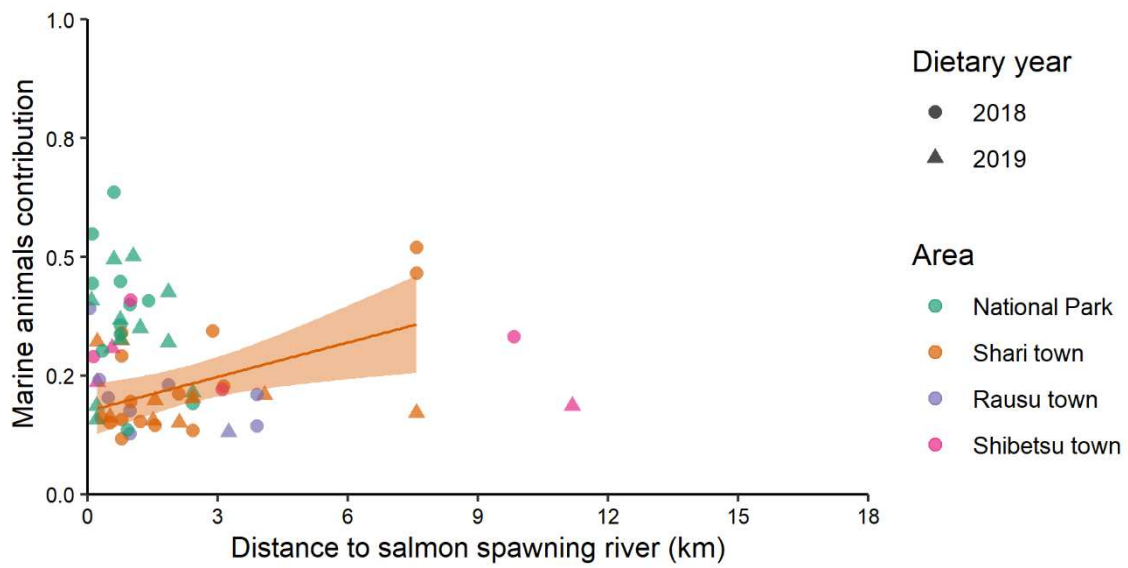


Figure 4-6. Relationship between the distance from the sample collection location to the nearest predicted salmon spawning river and the dietary contribution of marine animal contribution in the trapped female bears. The significant linear regression is shown as a solid line with a 95% confidence intervals.

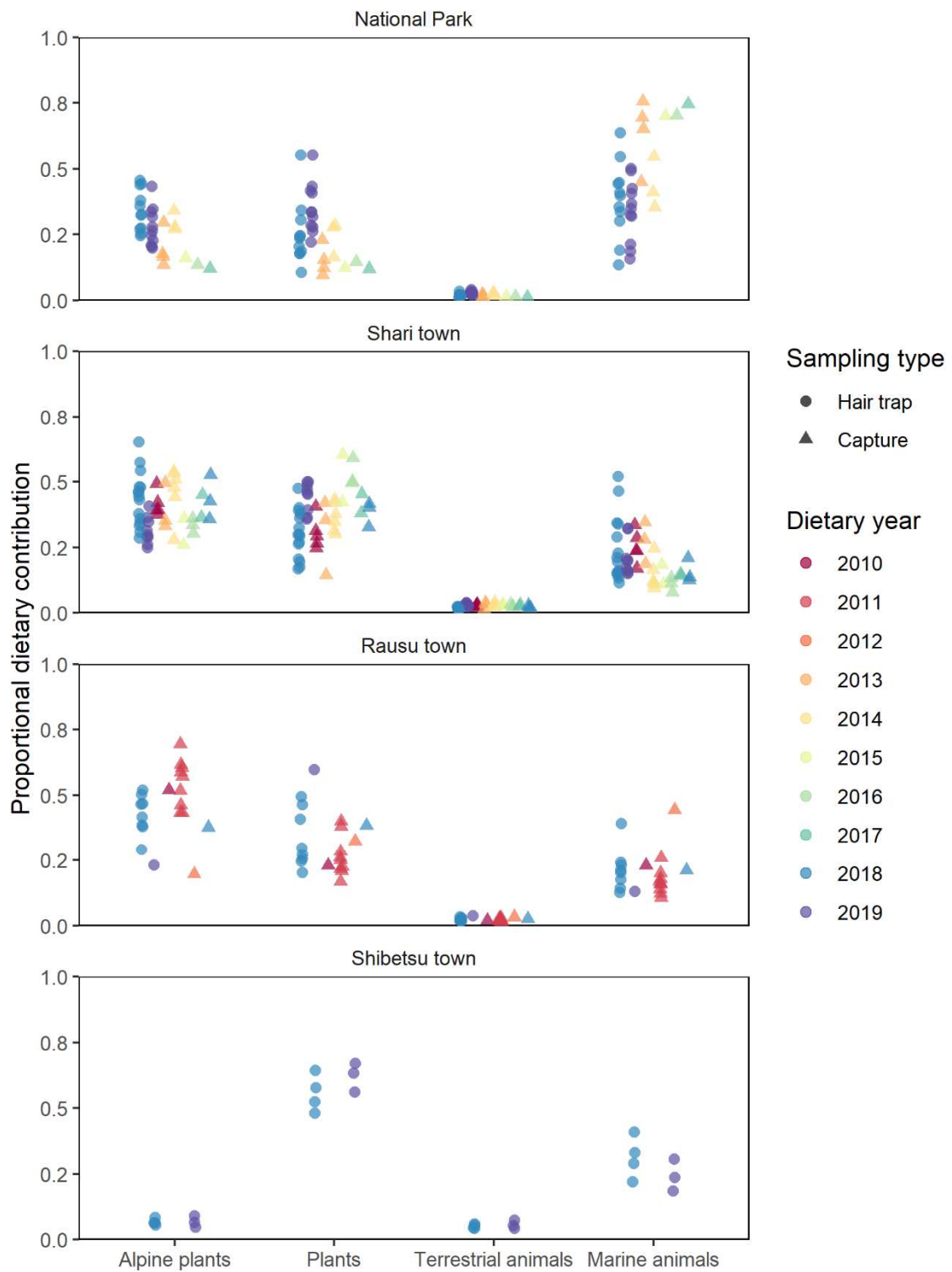
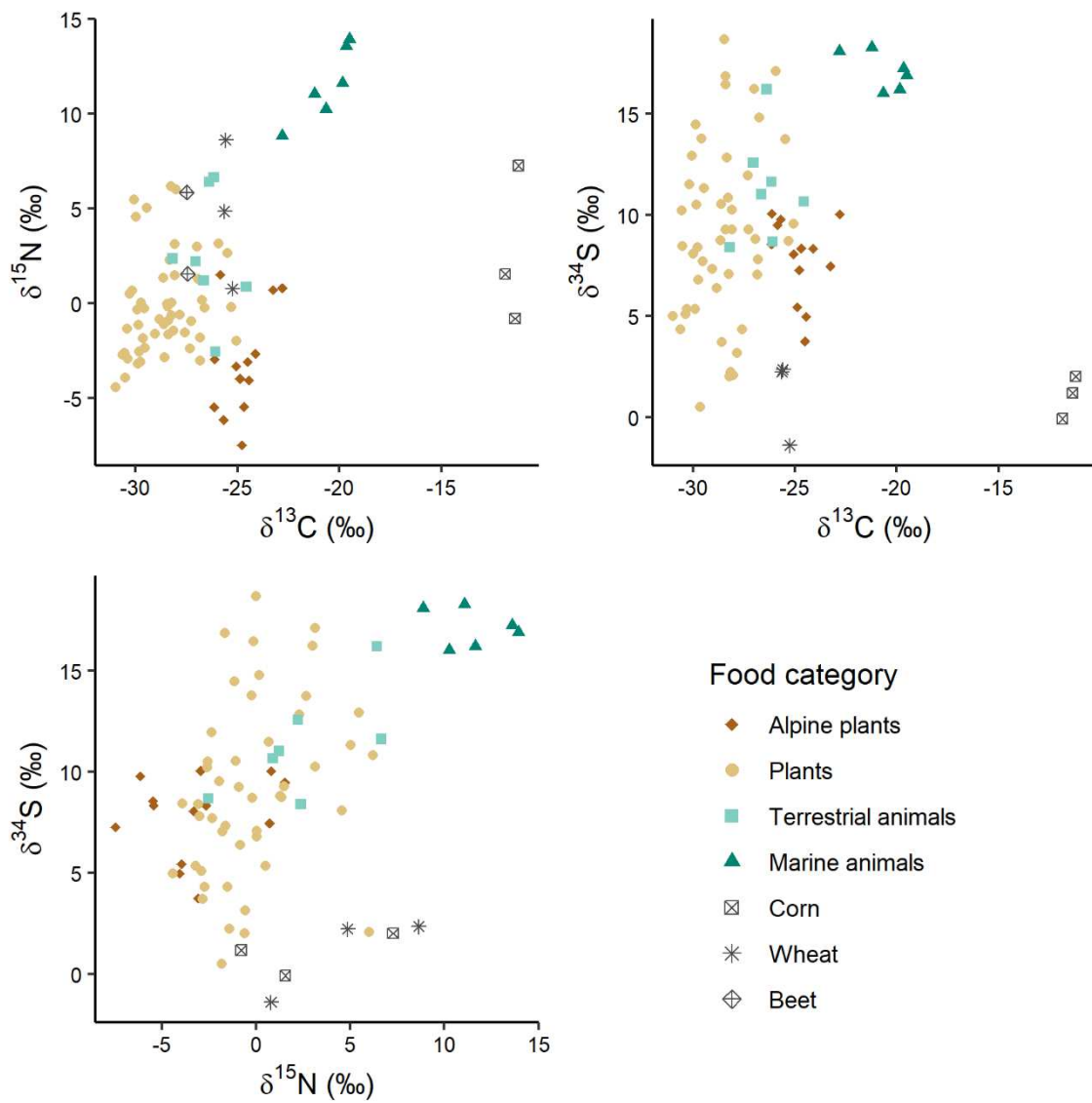
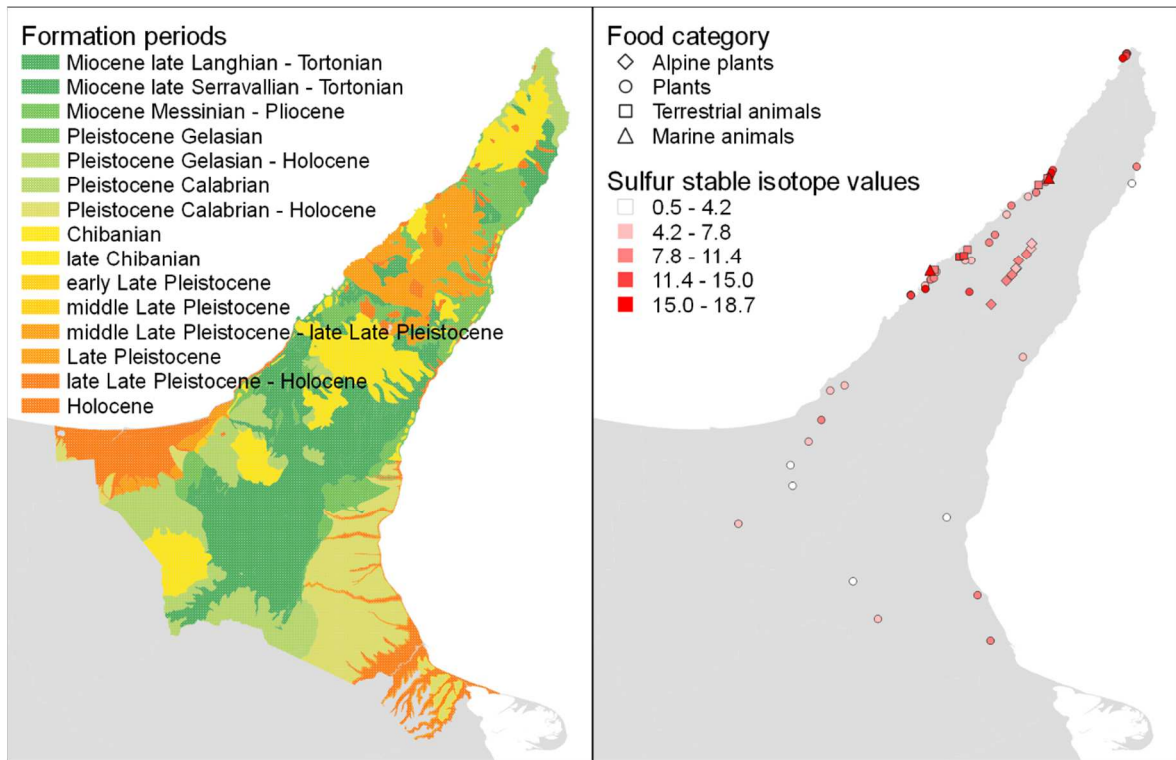


Figure 4-7. The estimated proportional dietary contributions of each food category, plotted by sampling type for each area.



Appendix S4-1. Plot of stable isotope values of potential food for brown bears collected on the Shiretoko Peninsula. Sulfur stable isotope values could not be measured in beets.



Appendix S4-2. A map of the geological formation periods of the Shiretoko Peninsula (left) was generated based on the 1:200,000 Seamless Geological Map of Japan V2 (Geological Survey of Japan, AIST, Data update: April 6, 2020, <https://gbank.gsj.jp/seamless/>). Areas marked in green are older formation periods, while areas marked in orange are more recently formed. The association between the sampling sites and sulfur stable isotope values of each natural food item was mapped (right).

Chapter 5 | Conclusion

In this study, we used hair stable isotope analysis to reveal interindividual variation in the diet of a brown bear population on the Shiretoko Peninsula. It is difficult to obtain a sufficient number of samples for assessing interindividual variations in the diet of wild animals, especially in large mammals. Many studies of dietary analysis by stable isotope ratios have used samples collected by capturing animals. However, this collection method is very expensive, time-consuming, and invasive, thus limiting its feasibility. In contrast, noninvasive sampling, such as hair traps, can be useful. At least about 5 mg of guard hairs (10-30 hairs) per sample are required to assess the carbon, nitrogen, and sulfur stable isotope ratios. Although the use of three isotope ratios probably improves the accuracy of dietary estimates, the use of only the carbon and nitrogen isotopes might be sufficient for estimating the approximate contribution of the five food categories in this study. If the stable isotope ratios of only these two elements are assessed, approximately 1 mg of guard hairs is sufficient, thereby allowing the inclusion of more samples collected by the hair trap in the analysis. In addition, the method of classifying hair growth stages and correcting the isotopic values according to the collection date that I proposed in Chapter 2 and 3 will considerably expand the range of samples that can be included in the stable isotope analysis. Since interindividual variation in feeding habits is an important factor in the proper management of bear populations, the techniques that I have proposed are expected to advance research in this field.

The Shiretoko Peninsula has a brown bear population with one of the highest densities worldwide and contains high-quality habitat with rivers where salmonids naturally spawn (Hokkaido Government 2017). I found that the diet of male bears shifted later in life to consume more salmon, suggesting that salmon is a preferred food for bears in this peninsula. Additionally, I found regional variations in bear diet that indicated that salmon consumption by bears was concentrated in Shiretoko National Park. I also suggested that young bears and bears living outside of the national park consume pine nuts as an energy-rich food alternative to salmon. These results highlighted that the national park is a hotspot for brown bears inhabiting the peninsula, but other areas may also be good habitats. I suggested that young bears and bears living outside of the national park consume pine nuts as an energy-rich food alternative to salmon.

Poor body condition due to food scarcity is one of the factors accelerating the human-bear conflict. The body condition of brown bears inhabiting the Shiretoko Peninsula declines from spring to summer and recovers from late summer to autumn (Shirane et al. 2021). Bears consume pine nuts from July to September and salmon from August to November, as well as cherries from July to September and acorns

(*Quercus crispula*) in October (Shirane et al. 2021). In addition to salmon and pine nuts, fruits and acorns are recognized as important foods for bear survival and reproductive success in some bear populations worldwide (e.g., Costello et al. 2016, Hertel et al. 2018). These foods may also be important for bears on the Shiretoko Peninsula, especially for bears in Shibetsu town and for bears throughout the peninsula when pine nut resources are scarce. Therefore, I propose monitoring the abundance of cherries, hard masts and other fruits in the future. The ability of stable isotope analysis to classify food categories in detail is limited. Dietary analysis of scat can discriminate fruit species and detect crop use (e.g., Sato et al. 2004; Shirane et al. 2021), but may be less efficient in terms of sampling. Since each method has advantages and disadvantages, I recommend continuing dietary monitoring using a combination of these methods.

On the Shiretoko Peninsula, brown bears scavenge dead marine mammals that wash ashore. The stranding of marine mammals (e.g., Dall's porpoise, *Phocoena dalli*; harbor porpoise, *Phocoena phocoena*; and Steller's sea lion, *Eumetopias jubatus*) occurs across the peninsula (Stranding Network Hokkaido 2021). Although the stranding of marine mammals provides only incidentally available food for brown bears, it offers a great deal of nutrition. Through bear foraging behaviors and defecation, large amounts of marine-derived nutrients are transported to land, even in areas where salmon runs are scarce. However, dead marine mammals that drift ashore near residential areas are immediately removed by humans, because such energy-rich foods strongly attract brown bears, and situations dangerous to nearby residents may occur. Therefore, it is assumed that brown bears can acquire marine mammals only in the national park or from a very limited number of shores outside of the national park. The use of marine mammals by brown bears is a subject of great ecological interest for assessing the impact of the anthropogenic food removal on brown bears and ecosystems. Although both salmon and marine mammals were included in the same food category in this study, the consumption of these animals at different trophic levels will be clearly distinguished by using nitrogen stable isotope analysis on the amino acids (Chikaraishi et al. 2010).

This study suggested that the triggers of human-bear conflicts varied regionally even within a small local population. Dietary differences between captured and trapped female bears in Rausu town could support the idea that the characteristic feeding habits of individual bears are associated with human-bear conflicts. However, the fact that no significant differences in diet were found between captured and trapped female bears in Shari town suggests that, in terms of their feeding habits, any bear has the potential to appear in human settlements. In such cases, it is important to construct an environment that reduces the

risk of forest-dwelling bears appearing in human settlements, even accidentally. For example, installing electric fences around areas where crops and other anthropogenic food that may attract bears, establishing buffer zones, maintaining and forming forests with abundant food for bears, and not feeding or approaching bears even when they are encountered. Thorough efforts are required to ensure that all brown bears do not experience harmless encounters with humans. Human habituation, food scarcity, and crop dependence are completely different scenarios; thus, it is important to consider the local environment, context of the conflict, and individual variations in bear feeding habits to construct a proper monitoring system.

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References

- Adams, Megan S., Christina N. Service, Andrew Bateman, Mathieu Bourbonnais, Kyle A. Artelle, Trisalyn Nelson, Paul C. Paquet, Taal Levi, and Chris T. Darimont. 2017. "Intrapopulation Diversity in Isotopic Niche over Landscapes: Spatial Patterns Inform Conservation of Bear–Salmon Systems." *Ecosphere* 8 (6): e01843.
- Arnold, Stevan James. 1981. "The Microevolution of Feeding Behavior." In *Foraging Behavior: Ecological, Ethological and Psychological Approaches*, 409–53. Garland Press.
- Artelle, Kyle A., Sean C. Anderson, John D. Reynolds, Andrew B. Cooper, Paul C. Paquet, and Chris T. Darimont. 2016. "Ecology of Conflict: Marine Food Supply Affects Human-Wildlife Interactions on Land." *Scientific Reports* 6 (1): 25936.
- Aumiller, Larry D., and Colleen A. Matt. 1994. "Management of McNeil River State Game Sanctuary for Viewing of Brown Bears." *Ursus* 9: 51–61.
- Ayliffe, L. K., T. E. Cerling, T. Robinson, A. G. West, M. Sponheimer, B. H. Passey, J. Hammer, B. Roeder, M. D. Dearing, and J. R. Ehleringer. 2004. "Turnover of Carbon Isotopes in Tail Hair and Breath CO₂ of Horses Fed an Isotopically Varied Diet." *Oecologia* 139 (1): 11–22.
- Bastille-Rousseau, Guillaume, Daniel Fortin, Christian Dussault, Réhaume Courtois, and Jean-Pierre Ouellet. 2011. "Foraging Strategies by Omnivores: Are Black Bears Actively Searching for Ungulate Neonates or Are They Simply Opportunistic Predators?" *Ecography* 34 (4): 588–96.
- Bateman, Alison S., and Simon D. Kelly. 2007. "Fertilizer Nitrogen Isotope Signatures." *Isotopes in Environmental and Health Studies* 43 (3): 237–47.
- Bateman, Alison S., Simon D. Kelly, and Timothy D. Jickells. 2005. "Nitrogen Isotope Relationships between Crops and Fertilizer: Implications for Using Nitrogen Isotope Analysis as an Indicator of Agricultural Regime." *Journal of Agricultural and Food Chemistry* 53 (14): 5760–65.
- Bates, Douglas, Martin Mächler, Ben Bolker, and Steve Walker. 2015. "Fitting Linear Mixed-Effects Models Using lme4." *Journal of Statistical Software* 67 (October): 1–48.
- Belant, Jerrold L., Knut Kielland, Erich H. Follmann, and Layne G. Adams. 2006. "Interspecific Resource

- Partitioning in Sympatric Ursids.” *Ecological Applications* 16 (6): 2333–43.
- Ben-David, Merav, Kimberly Titus, and LaVern R. Beier. 2004. “Consumption of Salmon by Alaskan Brown Bears: A Trade-off between Nutritional Requirements and the Risk of Infanticide?” *Oecologia* 138 (3): 465–74.
- Bentzen, Torsten W., Richard T. Shideler, and Todd M. O’Hara. 2014. “Use of Stable Isotope Analysis to Identify Food-Conditioned Grizzly Bears on Alaska’s North Slope.” *Ursus* 25 (1): 14–23.
- Bojarska, Katarzyna, and Nuria Selva. 2012. “Spatial Patterns in Brown Bear *Ursus arctos* Diet: The Role of Geographical and Environmental Factors: Biogeographical Variation in Brown Bear Diet.” *Mammal Review* 42 (2): 120–43.
- Bojarska, Katarzyna, and Nuria Selva. 2013. “Correction Factors for Important Brown Bear Foods in Europe.” *Ursus* 24 (1): 13–15.
- Bolnick, Daniel I., Richard Svanbäck, Márcio S. Araújo, and Lennart Persson. 2007. “Comparative Support for the Niche Variation Hypothesis That More Generalized Populations Also Are More Heterogeneous.” *Proceedings of the National Academy of Sciences* 104 (July): 10075–79.
- Can, Özgün Emre, Neil D’Cruze, David L. Garshelis, John Beecham, and David W. Macdonald. 2014. “Resolving Human-Bear Conflict: A Global Survey of Countries, Experts, and Key Factors: Human-Bear Conflict.” *Conservation Letters* 7 (6): 501–13.
- Careddu, Giulio, Paolo Ciucci, Stella Mondovì, Edoardo Calizza, Loreto Rossi, and Maria Letizia Costantini. 2021. “Gaining Insight into the Assimilated Diet of Small Bear Populations by Stable Isotope Analysis.” *Scientific Reports* 11 (1): 14118.
- Chikaraishi, Y., N. O. Ogawa, Y. Takano, M. Tsuchiya, and N. Ohkouchi. 2010. “Food Chain Analysis by Nitrogen Isotopic Composition of Amino Acids.” *Chikyukagaku (Geochemistry)* 44: 233–241.
- Clutton-Brock, T. H., ed. 1990. *Reproductive Success: Studies of Individual Variation in Contrasting Breeding Systems*. Chicago, IL: University of Chicago Press.
- Coogan, Sean C. P., David Raubenheimer, Gordon B. Stenhouse, Nicholas C. Coops, and Scott E. Nielsen. 2018. “Functional Macronutritional Generalism in a Large Omnivore, the Brown Bear.” *Ecology and*

- Evolution 8 (4): 2365–76.
- Cook, Nigel J. 2012. “Review: Minimally Invasive Sampling Media and the Measurement of Corticosteroids as Biomarkers of Stress in Animals.” *Canadian Journal of Animal Science* 92: 227–59.
- Costello, Cecily M., Steven L. Cain, Shannon Pils, Leslie Frattaroli, Mark A. Haroldson, and Frank T. van Manen. 2016. “Diet and Macronutrient Optimization in Wild Ursids: A Comparison of Grizzly Bears with Sympatric and Allopatric Black Bears.” *PLOS ONE* 11 (5): e0153702.
- Dahle, Bjørn, Ole J. Sørensen, Egil H. Wedul, Jon E. Swenson, and Finn Sandegren. 1998. “The Diet of Brown Bears *Ursus Arctos* in Central Scandinavia: Effect of Access to Free-Ranging Domestic Sheep *Ovis aries*.” *Wildlife Biology* 4 (1): 147–58.
- Dahle, Bjørn, and Jon E. Swenson. 2003. “Home Ranges in Adult Scandinavian Brown Bears (*Ursus arctos*): Effect of Mass, Sex, Reproductive Category, Population Density and Habitat Type.” *Journal of Zoology* 260 (4): 329–35.
- Deacy, William W., Joy A. Erlenbach, William B. Leacock, Jack A. Stanford, Charles T. Robbins, and Jonathan B. Armstrong. 2018. “Phenological Tracking Associated with Increased Salmon Consumption by Brown Bears.” *Scientific Reports* 8 (1): 11008.
- DeNiro, Michael J., and Samuel Epstein. 1978. “Influence of Diet on the Distribution of Carbon Isotopes in Animals.” *Geochimica et Cosmochimica Acta* 42 (5): 495–506.
- Diaz, Sandra F., Sheila M. F. Torres, Robert W. Dunstan, and Chalernpol Lekcharoensuk. 2004. “An Analysis of Canine Hair Re-Growth after Clipping for a Surgical Procedure.” *Veterinary Dermatology* 15 (1): 25–30.
- Douma, Jacob C., and James T. Weedon. 2019. “Analysing Continuous Proportions in Ecology and Evolution: A Practical Introduction to Beta and Dirichlet Regression.” *Methods in Ecology and Evolution* 10 (9): 1412–30.
- Echeverría, Alejandra Isabel, and Aldo Iván Vassallo. 2012. “Role of Maternal Odors on Foraging Behavior during Postnatal Development in a Solitary Subterranean Rodent *Ctenomys talarum*.” *Acta Ethologica* 15 (1): 91–99.

- Edwards, Mark A., Andrew E. Derocher, Keith A. Hobson, Marsha Branigan, and John A. Nagy. 2011. "Fast Carnivores and Slow Herbivores: Differential Foraging Strategies among Grizzly Bears in the Canadian Arctic." *Oecologia* 165 (4): 877–89.
- Elfström, Marcus, Andreas Zedrosser, Ole-Gunnar Støen, and Jon E. Swenson. 2012. "Ultimate and Proximate Mechanisms Underlying the Occurrence of Bears Close to Human Settlements: Review and Management Implications." *Mammal Review* 44 (1): 5–18.
- Erlenbach, Joy A., Karyn D. Rode, David Raubenheimer, and Charles T. Robbins. 2014. "Macronutrient Optimization and Energy Maximization Determine Diets of Brown Bears." *Journal of Mammalogy* 95 (1): 160–68.
- Felicetti, Laura A, Charles C Schwartz, Robert O Rye, Kerry A Gunther, James G Crock, Mark A Haroldson, Lisette Waits, and Charles T Robbins. 2004. "Use of Naturally Occurring Mercury to Determine the Importance of Cutthroat Trout to Yellowstone Grizzly Bears." *Canadian Journal of Zoology* 82 (3): 493–501.
- Felicetti, Laura A, Charles C Schwartz, Robert O Rye, Mark A Haroldson, Kerry A Gunther, Donald L Phillips, and Charles T Robbins. 2003. "Use of Sulfur and Nitrogen Stable Isotopes to Determine the Importance of Whitebark Pine Nuts to Yellowstone Grizzly Bears." *Canadian Journal of Zoology* 81 (5): 763–70.
- Florin, Scott T., Laura A. Felicetti, and Charles T. Robbins. 2011. "The Biological Basis for Understanding and Predicting Dietary-Induced Variation in Nitrogen and Sulphur Isotope Ratio Discrimination: Understanding Nitrogen and Sulphur Discrimination." *Functional Ecology* 25 (3): 519–26.
- Fortin, Jennifer K., Sean D. Farley, Karyn D. Rode, and Charles T. Robbins. 2007. "Dietary and Spatial Overlap between Sympatric Ursids Relative to Salmon Use." *Ursus* 18 (1): 19–29.
- Galef, Bennett G., and Kevin N. Laland. 2005. "Social Learning in Animals: Empirical Studies and Theoretical Models." *BioScience* 55 (6): 489–99.
- Galef, Bennett G., and Elaine E. Whiskin. 2001. "Interaction of Social and Individual Learning in Food Preferences of Norway Rats." *Animal Behaviour* 62 (1): 41–46.
- Hata, A., M. B. Takada, R. Nakashita, K. Fukasawa, T. Oshida, Y. Ishibashi, and Y. Sato. 2017. "Stable

- Isotope and DNA Analyses Reveal the Spatial Distribution of Crop-Foraging Brown Bears.” *Journal of Zoology* 303 (3): 207–17.
- Hatch, Kent A., Kimberly A. Kester, Janene Auger, Beverly L. Roeder, Kevin Bunnell, and Hal L. Black. 2019. “The Effect of Sex, Age, and Location on Carnivory in Utah Black Bears (*Ursus americanus*).” *Oecologia* 189 (4): 931–37.
- Hertel, Anne G., Richard Bischof, Ola Langval, Atle Mysterud, Jonas Kindberg, Jon E. Swenson, and Andreas Zedrosser. 2018. “Berry Production Drives Bottom-up Effects on Body Mass and Reproductive Success in an Omnivore.” *Oikos* 127 (2): 197–207.
- Heyes, Cecilia M., and Bennett G. Galef. 1996. *Social Learning In Animals: The Roots of Culture*. Elsevier.
- Hilderbrand, G V, C C Schwartz, C T Robbins, M E Jacoby, T A Hanley, S M Arthur, and C Servheen. 1999. “The Importance of Meat, Particularly Salmon, to Body Size, Population Productivity, and Conservation of North American Brown Bears.” *Canadian Journal of Zoology* 77 (1): 132–38.
- Hobson, Keith A. 1987. “Use of Stable-Carbon Isotope Analysis to Estimate Marine and Terrestrial Protein Content in Gull Diets.” *Canadian Journal of Zoology* 65 (5): 1210–13.
- Hobson, Keith A. 1999. “Tracing Origins and Migration of Wildlife Using Stable Isotopes: A Review.” *Oecologia* 120 (3): 314–26.
- Hobson, Keith A, Bruce N McLellan, and John G Woods. 2000. “Using Stable Carbon ($\Delta^{13}\text{C}$) and Nitrogen ($\Delta^{15}\text{N}$) Isotopes to Infer Trophic Relationships among Black and Grizzly Bears in the Upper Columbia River Basin, British Columbia.” *Canadian Journal of Zoology* 78 (8): 1332–39.
- Hobson, Keith A., and Harold E. Welch. 1992. “Determination of Trophic Relationships within a High Arctic Marine Food Web Using $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ Analysis.” *Marine Ecology Progress Series* 84 (1): 9–18.
- Hokkaido Government. 2017. “Brown Bear Management Plan in Hokkaido (in Japanese).” http://www.pref.hokkaido.lg.jp/ks/skn/higuma/hokkaido_bear_management_plan05.pdf (accessed July 8, 2021).

- Hopkins, John B. 2013. "Use of Genetics to Investigate Socially Learned Foraging Behavior in Free-Ranging Black Bears." *Journal of Mammalogy* 94 (6): 1214–22.
- Hopkins, John B., and Jake M. Ferguson. 2012. "Estimating the Diets of Animals Using Stable Isotopes and a Comprehensive Bayesian Mixing Model." Edited by Lyle Konigsberg. *PLoS ONE* 7 (1): e28478.
- Hopkins, John B, Jake M Ferguson, Cheryl Frederick, and Klemen Jerina. 2021. "Measuring the Impact of Corn on Mammalian Omnivores." Edited by Patrick Zollner. *Journal of Mammalogy* 102 (1): 270–82.
- Hopkins, John B., Jake M. Ferguson, Daniel B. Tyers, and Carolyn M. Kurle. 2017. "Selecting the Best Stable Isotope Mixing Model to Estimate Grizzly Bear Diets in the Greater Yellowstone Ecosystem." Edited by Jordi Moya-Larano. *PLOS ONE* 12 (5): e0174903.
- Hothorn, Torsten, Frank Bretz, and Peter Westfall. 2008. "Simultaneous Inference in General Parametric Models." *Biometrical Journal* 50 (3): 346–63.
- Hothorn, Torsten and Kurt Hornik. 2021. "exactRankTests: Exact Distributions for Rank and Permutation Tests." R package version 0.8-34. <https://CRAN.R-project.org/package=exactRankTests>
- Jaeggi, Adrian V., Lynda P. Dunkel, Maria A. Van Noordwijk, Serge A. Wich, Agnes A.L. Sura, and Carel P. Van Schaik. 2010. "Social Learning of Diet and Foraging Skills by Wild Immature Bornean Orangutans: Implications for Culture." *American Journal of Primatology* 72 (1): 62–71.
- Jimbo, Mina, Naoya Matsumoto, Hideyuki Sakamoto, Yojiro Yanagawa, Yoshiko Torii, Masami Yamanaka, Tsuyoshi Ishinazaka, et al. 2020. "Hair Growth in Brown Bears and Its Application to Ecological Studies on Wild Bears." *Mammal Study* 45 (4): 337–45.
- Jimbo, Mina, Tsuyoshi Ishinazaka, Yuri Shirane, Yoshihiro Umemura, Masami Yamanaka, Hiroyuki Uno, Mariko Sashika, Tsubota Tsubota, and Michito Shimozuru. In press. "Diet Selection and Asocial Learning: Natal Habitat Influence on Lifelong Foraging Strategies in Solitary Large Mammals." *Ecosphere*.
- Jones, Elena S., Douglas C. Heard, and Michael P. Gillingham. 2006. "Temporal Variation in Stable Carbon and Nitrogen Isotopes of Grizzly Bear Guardhair and Underfur." Edited by Jacob Bowman. *Wildlife Society Bulletin* 34 (5): 1320–25.

- Kandel, Eric R., John D. Koester, Sarah H. Mack, and Steven A. Siegelbaum. 2021. *Principles of Neural Science*. Sixth Edition. McGraw-Hill Education / Medical.
- Kendal, Rachel L., Isabelle Coolen, Yfke van Bergen, and Kevin N. Laland. 2005. "Trade - Offs in the Adaptive Use of Social and Asocial Learning." *Advances in the Study of Behavior* 35: 333–79.
- Kersey, David C., and Martin Dehnhard. 2014. "The Use of Noninvasive and Minimally Invasive Methods in Endocrinology for Threatened Mammalian Species Conservation." *General and Comparative Endocrinology*, 2nd Meeting of The North American Society for Comparative Endocrinology, 203 (July): 296–306.
- Kohira, Masao, Hideaki Okada, and Masami Yamanaka. 2006. "Controlled Exposure: Demographic Trends, Dispersal Patterns, and Management of Brown Bears in Shiretoko National Park." In *Wildlife in Shiretoko and Yellowstone National Parks*, 238–42. Shari, Hokkaido, Japan: Shiretoko Nature Foundation.
- Koshino, Yosuke, Hideaki Kudo, and Masahide Kaeriyama. 2013. "Stable Isotope Evidence Indicates the Incorporation into Japanese Catchments of Marine-Derived Nutrients Transported by Spawning Pacific Salmon." *Freshwater Biology* 58 (9): 1864–77.
- Kurle, Carolyn M., Paul L. Koch, Bernie R. Tershy, and Donald A. Croll. 2014. "The Effects of Sex, Tissue Type, and Dietary Components on Stable Isotope Discrimination Factors ($\Delta^{13}\text{C}$ and $\Delta^{15}\text{N}$) in Mammalian Omnivores." *Isotopes in Environmental and Health Studies* 50 (3): 307–21.
- Kushiro Nature Conservation Office of the Ministry of the Environment, Hokkaido Regional Forest Office, Hokkaido, Shari town, Rausu town, and Shibetsu town. 2017. "Brown Bear Management Plan in the Shiretoko Peninsula (in Japanese)". http://shiretoko-whc.com/data/management/higuma/higuma_kanri_keikaku.pdf (accessed July 7, 2021).
- Leblond, C. P. 1951. "Histological Structure of Hair, with a Brief Comparison to Other Epidermal Appendages and Epidermis Itself." *Annals of the New York Academy of Sciences* 53 (3): 464–75.
- Lee-Thorp, Julia A. 2002. "Preservation of Biogenic Carbon Isotopic Signals in Plio-Pleistocene Bone and Tooth Mineral." In *Biogeochemical Approaches to Paleodietary Analysis*, edited by Stanley H. Ambrose and M. Anne Katzenberg, vol. 5, 89–115. *Advances in Archaeological and Museum Science*.

Boston, MA: Springer US.

- López-Alfaro, Claudia, Sean C. P. Coogan, Charles T. Robbins, Jennifer K. Fortin, and Scott E. Nielsen. 2015. "Assessing Nutritional Parameters of Brown Bear Diets among Ecosystems Gives Insight into Differences among Populations." Edited by Joseph K. Bump. PLOS ONE 10 (6): e0128088.
- Lupfer, Gwen, Jerome Frieman, and Daniel Coonfield. 2003. "Social Transmission of Flavor Preferences in Two Species of Hamsters (*Mesocricetus auratus* and *Phodopus campbelli*)." Journal of Comparative Psychology 117 (4): 449–55.
- Lynch, Emily C., Caley A. Johnson, Robert F. Lynch, Jessica M. Rothman, Anthony Di Fiore, and Ryne A. Palombit. 2020. "Mothers and Fathers Improve Immature Baboon Foraging Success." Behaviour 157 (5): 387–414.
- Macbeth, B. J., M.R.L. Cattet, G. B. Stenhouse, M. L. Gibeau, and D. M. Janz. 2010. "Hair Cortisol Concentration as a Noninvasive Measure of Long-Term Stress in Free-Ranging Grizzly Bears (*Ursus arctos*): Considerations with Implications for Other Wildlife." Canadian Journal of Zoology 88 (10): 935–49.
- Maier, Marco Johannes. 2014. "DirichletReg: Dirichlet Regression for Compositional Data in R." Research Report Series / Department of Statistics and Mathematics 125.
- Mangipane, Lindsey S., Diana J. R. Lafferty, Kyle Joly, Mathew S. Sorum, Matthew D. Cameron, Jerrold L. Belant, Grant V. Hilderbrand, and David D. Gustine. 2020. "Dietary Plasticity and the Importance of Salmon to Brown Bear (*Ursus arctos*) Body Size and Condition in a Low Arctic Ecosystem." Polar Biology 43 (7): 825–33.
- Mann, Janet, Brooke L. Sargeant, and Matthew Minor. 2007. "Calf Inspections of Fish Catches in Bottlenose Dolphins (*Tursiops* Sp.): Opportunities for Oblique Social Learning?" Marine Mammal Science 23 (1): 197–202.
- Mazur, Rachel, and Victoria Seher. 2008. "Socially Learned Foraging Behaviour in Wild Black Bears, *Ursus americanus*." Animal Behaviour 75 (4): 1503–8.
- McGregor, Ba. 1988. "Effects of Different Nutritional Regimens on the Productivity of Australian Cashmere Goats and the Partitioning of Nutrients between Cashmere and Hair Growth." Australian

- Journal of Experimental Agriculture 28 (4): 459–67.
- Mizukami, Rumiko N., Mitsuaki Goto, Shigeyuki Izumiyama, Hidetake Hayashi, and Muneoki Yoh. 2005a. “Estimation of Feeding History by Measuring Carbon and Nitrogen Stable Isotope Ratios in Hair of Asiatic Black Bears.” *Ursus* 16 (1): 93–101.
- Mizukami, R. N., M. Goto, S. Izumiyama, M. Yoh, N. Ogura, and H. Hayashi. 2005b. “Temporal Diet Changes Recorded by Stable Isotopes in Asiatic Black Bear (*Ursus thibetanus*) Hair.” *Isotopes in Environmental and Health Studies* 41 (1): 87–94.
- Morehouse, Andrea T., Tabitha A. Graves, Nate Mickle, and Mark S. Boyce. 2016. “Nature vs. Nurture: Evidence for Social Learning of Conflict Behaviour in Grizzly Bears.” Edited by Elisabetta Palagi. *PLOS ONE* 11 (11): e0165425.
- Moriwaki, Jun, Ryosuke Omori, Michito Shimozuru, Hifumi Tsuruga, Tsutomu Mano, and Toshio Tsubota. 2018. “Evaluation of Body Condition Using Body Mass and Chest Girth in Brown Bears of Hokkaido, Japan (*Ursus arctos yesoensis*).” *Japanese Journal of Veterinary Research* 66 (2): 71–81.
- Mowat, Garth, P. Jeff Curtis, and Diana J. R. Lafferty. 2017. “The Influence of Sulfur and Hair Growth on Stable Isotope Diet Estimates for Grizzly Bears.” Edited by Zhigang Jiang. *PLOS ONE* 12 (3): e0172194.
- Mowat, Garth, and Douglas C Heard. 2006. “Major Components of Grizzly Bear Diet across North America.” *Canadian Journal of Zoology* 84 (3): 473–89.
- Naganuma, Tomoko, Shinsuke Koike, Rumiko Nakashita, Chinatsu Kozakai, Koji Yamazaki, Shino Furusaka, and Koichi Kaji. 2020. “Age- and Sex-Associated Differences in the Diet of the Asian Black Bear: Importance of Hard Mast and Sika Deer.” *Mammal Study* 45 (2): 155–66.
- Narita, Ryo, Tsutom Mano, Reo Yokoyama, and Atsushi Takayanagi. 2011. “Variation in Maize Consumption by Brown Bears (*Ursus arctos*) in Two Coastal Areas of Hokkaido, Japan.” *Mammal Study* 36 (1): 33–39.
- Naves, Javier, Alberto Fernández-Gil, Carlos Rodríguez, and Miguel Delibes. 2006. “Brown Bear Food Habits at the Border of Its Range: A Long-Term Study.” *Journal of Mammalogy* 87 (5): 899–908.

- Ohdachi, Satoshi, and Toshiki Aoi. 1987. "Food Habits of Brown Bears in Hokkaido, Japan." *Bears: Their Biology and Management* 7: 215–20.
- Parakkal, Paul F. 1970. "Morphogenesis of the Hair Follicle during Catagen." *Zeitschrift Für Zellforschung Und Mikroskopische Anatomie* 107 (2): 174–86.
- Peacock, Mary M., and Stephen H. Jenkins. 1988. "Development of Food Preferences: Social Learning by Belding's Ground Squirrels *Spermophilus beldingi*." *Behavioral Ecology and Sociobiology* 22 (6): 393–99.
- Perrin, M R, and B S Campbell. 1980. "Key to the Mammals of the Andries Vosloo Kudu Reserve (Eastern Cape), Based on Their Hair Morphology, for Use in Predator Scat Analysis." *South African Journal of Wildlife Research* 10 (1): 1–14.
- Persson, Inga-Lill, Steinar Wikan, Jon E. Swenson, and Ivar Myrsterud. 2001. "The Diet of the Brown Bear *Ursus arctos* in the Pasvik Valley, Northeastern Norway." *Wildlife Biology* 7 (1): 27–37.
- Peterson, Bruce J., and Brian Fry. 1987. "Stable Isotopes in Ecosystem Studies." *Annual Review of Ecology and Systematics* 18 (1): 293–320.
- Phillips, Donald L., Richard Inger, Stuart Bearhop, Andrew L. Jackson, Jonathan W. Moore, Andrew C. Parnell, Brice X. Semmens, and Eric J. Ward. 2014. "Best Practices for Use of Stable Isotope Mixing Models in Food-Web Studies." *Canadian Journal of Zoology* 92 (10): 823–35.
- Phillips, Donald L., Seth D. Newsome, and Jillian W. Gregg. 2005. "Combining Sources in Stable Isotope Mixing Models: Alternative Methods." *Oecologia* 144 (4): 520–27.
- Pokrovskaya, Liya. 2015. "Foraging Activity and Food Selection in Asiatic Black Bear Orphaned Cubs in Absence of Social Learning from a Mother." *Mammalian Biology* 80 (4): 355–64.
- QGIS.org, 2022. QGIS Geographic Information System. QGIS Association. <http://www.qgis.org>
- R Core Team. 2019. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>
- R Core Team. 2021. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. <https://www.R-project.org/>

- Rasmussen, Palle V., and Christian F. Børsting. 2000. "Effects of Variations in Dietary Protein Levels on Hair Growth and Pelt Quality in Mink (*Mustela vison*)." *Canadian Journal of Animal Science* 80 (4): 633–42.
- Robbins, Charles T., Jennifer K. Fortin, Karyn D. Rode, Sean D. Farley, Lisa A. Shipley, and Laura A. Felicetti. 2007. "Optimizing Protein Intake as a Foraging Strategy to Maximize Mass Gain in an Omnivore." *Oikos* 116 (10): 1675–82.
- Rode, Karyn D., Sean D. Farley, and Charles T. Robbins. 2006. "Behavioral Responses of Brown Bears Mediate Nutritional Effects of Experimentally Introduced Tourism." *Biological Conservation* 133 (1): 70–80.
- Sakiyama, Tomoki, Junko Morimoto, Jun Matsubayashi, Yasuto Furukawa, Mami Kondo, Hifumi Tsuruga, Tsutomu Mano, and Futoshi Nakamura. 2021. "Factors Influencing Lifespan Dependency on Agricultural Crops by Brown Bears." *Landscape and Ecological Engineering* 17 (3): 351–62.
- Sato, Yoshikazu, Toshiki Aoi, Koichi Kaji, and Seiki Takatsuki. 2004. "Temporal Changes in the Population Density and Diet of Brown Bears in Eastern Hokkaido, Japan." *Mammal Study* 29 (1): 47–53.
- Schoeninger, Margaret J, and Michael J DeNiro. 1984. "Nitrogen and Carbon Isotopic Composition of Bone Collagen from Marine and Terrestrial Animals." *Geochimica et Cosmochimica Acta* 48 (4): 625–39.
- Schwartz, Charles C., Mark A. Haroldson, Gary C. White, Richard B. Harris, Steve Cherry, Kim A. Keating, Dave Moody, and Christopher Servheen. 2006. "Temporal, Spatial, and Environmental Influences on the Demographics of Grizzly Bears in the Greater Yellowstone Ecosystem." *Wildlife Monographs* 161: 1–68.
- Shari town. 2021. "Wild Animals Damage Prevention Plan (in Japanese)." https://www.town.shari.hokkaido.jp/material/files/group/16/R305_sharityoutyoujyuuhigaiboushikei kaku.pdf (accessed February 24, 2022).
- Shibetsu town. 2017. "Wild Animals Damage Prevention Plan (in Japanese)." <https://www.shibetsutown.jp/file/contents/140/2489/H29tyoujyuuhogo.pdf> (accessed February 24,

2022).

Shimozuru, Michito, Yuri Shirane, Mina Jimbo, Masami Yamanaka, Masanao Nakanishi, Tsuyoshi Ishinazaka, Shinsuke Kasai, et al. 2020a. “Male Reproductive Input, Breeding Tenure, and Turnover in High-Density Brown Bear (*Ursus arctos yesoensis*) Populations on the Shiretoko Peninsula, Hokkaido, Japan.” *Canadian Journal of Zoology* 98 (3): 175–85.

Shimozuru, Michito, Yuri Shirane, Masami Yamanaka, Masanao Nakanishi, Tsuyoshi Ishinazaka, Shinsuke Kasai, Takane Nose, et al. 2020b. “Maternal Human Habituation Enhances Sons’ Risk of Human-Caused Mortality in a Large Carnivore, Brown Bears.” *Scientific Reports* 10 (1): 16498.

Shimozuru, Michito, Masami Yamanaka, Masanao Nakanishi, Jun Moriwaki, Fumihiko Mori, Masakatsu Tsujino, Yuri Shirane, et al. 2017. “Reproductive Parameters and Cub Survival of Brown Bears in the Rusha Area of the Shiretoko Peninsula, Hokkaido, Japan.” Edited by Marco Festa-Bianchet. *PLOS ONE* 12 (4): e0176251.

Shirane, Yuri, Mina Jimbo, Masami Yamanaka, Masanao Nakanishi, Fumihiko Mori, Tsuyoshi Ishinazaka, Mariko Sashika, Toshio Tsubota, and Michito Shimozuru. 2021. “Dining from the Coast to the Summit: Salmon and Pine Nuts Determine the Summer Body Condition of Female Brown Bears on the Shiretoko Peninsula.” *Ecology and Evolution* 11 (10): 5204–19.

Shirane, Yuri, Fumihiko Mori, Masami Yamanaka, Masanao Nakanishi, Tsuyoshi Ishinazaka, Tsutomu Mano, Mina Jimbo, Mariko Sashika, Toshio Tsubota, and Michito Shimozuru. 2020. “Development of a Noninvasive Photograph-Based Method for the Evaluation of Body Condition in Free-Ranging Brown Bears.” *PeerJ* 8: e9982.

Shirane, Yuri, Michito Shimozuru, Masami Yamanaka, Hifumi Tsuruga, Saiko Hirano, Natsuo Nagano, Jun Moriwaki, et al. 2018. “Sex-Biased Natal Dispersal in Hokkaido Brown Bears Revealed through Mitochondrial DNA Analysis.” *European Journal of Wildlife Research* 64 (6): 65.

Shirane, Yuri, Michito Shimozuru, Masami Yamanaka, Hifumi Tsuruga, Masanao Nakanishi, Tsuyoshi Ishinazaka, Takane Nose, et al. 2019. “Sex-Biased Dispersal and Inbreeding Avoidance in Hokkaido Brown Bears.” *Journal of Mammalogy* 100 (4): 1317–26.

Shiretoko Nature Foundation. 2021. Breakdown of the total number of individuals killed by hu-mans

- including behavioral stages (in Japanese). FY2021 1st Ezo Deer and Brown Bear Working Group, Reference 2-2. http://shiretoko-whc.com/data/meeting/shikakuma_wg/r03/shikakuma_R0301_shiryo2-2.pdf (accessed February 21, 2022).
- Sponheimer, M., T. F. Robinson, T. E. Cerling, L. Tegland, B. L. Roeder, L. Ayliffe, M. D. Dearing, and J. R. Ehleringer. 2006. "Turnover of Stable Carbon Isotopes in the Muscle, Liver, and Breath CO₂ of Alpacas (*Lama pacos*).” *Rapid Communications in Mass Spectrometry* 20 (9): 1395–99.
- Stranding Network Hokkaido. 2021. "Data from Stranding Information.” <https://kujira110.com/?cat=3> (accessed July 7, 2021)
- Teeri, J. A., and D. A. Schoeller. 1979. "Δ¹³C Values of an Herbivore and the Ratio of C₃ to C₄ Plant Carbon in Its Diet.” *Oecologia* 39 (2): 197–200.
- Terkel, Joseph. 1996. "Cultural Transmission of Feeding Behavior in the Black Rat (*Rattus rattus*).” In *Social Learning in Animals: The Roots of Culture*, 17–47. San Diego, CA, US: Academic Press.
- Thode, H. G., J. Macnamara, and W. H. Fleming. 1953. "Sulphur Isotope Fractionation in Nature and Geological and Biological Time Scales.” *Geochimica et Cosmochimica Acta* 3 (5): 235–43.
- Tieszen, L. L., T. W. Boutton, K. G. Tesdahl, and N. A. Slade. 1983. "Fractionation and Turnover of Stable Carbon Isotopes in Animal Tissues: Implications for Δ¹³C Analysis of Diet.” *Oecologia* 57: 32–37.
- Ueda, Momoko, and Lynne S. Bell. 2019. "Assessing Dual Hair Sampling for Isotopic Studies of Grizzly Bears.” *Rapid Communications in Mass Spectrometry* 33 (18): 1475–80.
- Yamauchi, Kiyoshi, Shigekazu Kurakake, Takahiro Morosawa, Mami Kondo, Reina Uno, Takashi Yuasa, Hifumi Tsuruga, Hidetoshi B. Tamate, and Masaaki Yoneda. 2014. "A Pilot Study of the Hair-Trapping Method in Asiatic Black Bears (*Ursus thibetanus*): Determination of Optimal Survey Period for Estimating Population Size.” *Mammal Study* 39 (4): 191–200.
- Yoneda, Masaaki. 1976. "Age Determination and Age Structure of the Ezo Brown Bear.” *The Journal of the Mammalogical Society of Japan* 7 (1): 1–8.
- Zedrosser, Andreas, Eva Bellemain, Pierre Taberlet, and Jon E. Swenson. 2007. "Genetic Estimates of Annual Reproductive Success in Male Brown Bears: The Effects of Body Size, Age, Internal

Relatedness and Population Density.” *Journal of Animal Ecology* 76 (2): 368–75.

Zedrosser, Andreas, Georg Rauer, and Luise Kruckenhauser. 2004. “Early Primiparity in Brown Bears.”
Acta Theriologica 49 (3): 427–32.

要約 (Summary in Japanese)

これまで、特にヒグマのような大型哺乳類の食性研究では、個体群や集団レベルでの平均的な食性調査が主流であった。しかし、安定同位体比分析などの個体レベルでの食性解析技術が発展したことにより、多くの動物種において採食生態の個体間変異が発見されるようになった。個体間変異は一般的に、食物資源や環境など外的要因に基づくもの、性別や年齢など個体の内的要因に基づくもの、そしてそれらでは説明できないものに分けられる。個体の食物選択は、行動、生存、繁殖を通して個体群動態や生態系に関わる。そのため、個体間変異をもたらす要因と影響を理解することは生態学的に重要な課題である。

第2章では、体毛の安定同位体比分析を生態学的研究に応用するために、飼育および野生ヒグマの体毛伸長様式（伸長速度、伸長期間、換毛期間、毛球の構造変化など）を調べた。ガードヘア（保護毛）は、飼育個体で4月下旬から10月上旬にかけて、野生個体で5月下旬から9月下旬にかけて伸長することが観察された。また、換毛は8月から9月末の間に完了した。毛球の形状は白球型（White sphere: WS タイプ）、黒鉤型（Black hook: BH タイプ）、白鉤型（White hook: WH タイプ）の3種類に分類された。毛球の形状は、採取した前年に伸長した古い体毛（WS タイプ）と採取したその年に新しく伸長した体毛（BH タイプ、WH タイプ）を識別するために利用できることが明らかとなった。

第3章では、知床半島に生息するヒグマの食性の個体間変異を調査し、食性の変異をもたらす要因について検討した。特に、母親から独立した後の採食経験が食物選択に及ぼす影響に注目した。知床半島で2010年から2020年の間に捕獲されたヒグマ（ $n=295$ ）から採取したガードヘアの安定同位体比分析によって、個体レベルで食性を推定した。また、捕獲時点の食性と出生地の平均的な食性を比較することで、ヒグマの食性が成長に伴ってどの程度変化したのかを定量的に評価した。その結果、ヒグマの食性は個体群内において地域、性別、年齢によって変化していることが明らかになった。雄雌ともに、年齢と食性に占める海産動物の寄与率には正の相関が見られた。また、メスは生涯に渡って出生地の平均的な食性を維持していたが、オスの食性は母親から独立して6年以上が経過し、身体的に成熟した段階で大きく変化していた。このような後半生における食性変化は、生息地の探索、採食経験の獲得、社会的優位性など、いくつかの要因によるものであると考えられた。本研究は、単独性の大型哺乳類において、独立後の採食経験がその後の食物選択を左右することを示唆する初めての証拠となる。

第4章では、ヒグマの食性の地域差が、人とヒグマの軋轢問題とどのように関連しているの

かを明らかにした。2019年および2020年に知床半島全域（斜里町、羅臼町、標津町）に立木型ヘアトラップを設置し、ヒグマの体毛を非侵襲的に採取した。食性の地域差を明確にするため、行動圏が小さく、母親から独立した3歳以上のメスを解析対象とした。また、ヘアトラップによって体毛が採取された個体（以下、トラップ個体）と有害駆除・狩猟・学術研究のための捕獲によって体毛が採取された個体（以下、捕獲個体）の食性を国立公園、斜里町、羅臼町のそれぞれで比較した。サケマス遡上河川の多くが市街地に囲まれた斜里町では、人前にくり返し出沒する個体のほうが林内に留まる個体よりも多くのサケマスを消費しているのではないかと予想したが、トラップ個体と捕獲個体の食性に差異は認められなかった。このことは、有害駆除された個体であっても、少なくとも捕殺の前年までは林内の個体と変わらない食物を利用していたことを示しており、斜里町でヒグマが人里に出沒するのは必ずしも高エネルギーな食物を獲得するためではないことを示唆している。羅臼町では、捕獲個体のほうがトラップ個体よりも高山植物（主にハイマツ球果）に強く依存していることが示唆された。これらの個体は、春から夏にかけて草本やサクラの実をバランスよく利用する個体に比べて、ハイマツ球果の資源量変動に敏感である可能性がある。知床半島に生息するヒグマにとって、ハイマツ球果はサケマスに代わる高エネルギー食物であると考えられるが、ハイマツ球果の資源不足が一部のメスヒグマにとっては人里に出沒するトリガーとなり得ることが示唆された。標津町のメスヒグマでは、サケマスやハイマツ球果の利用は少なく、その他の植物の利用が多かった。そのため、標津町でのヒグマと人の軋轢に関わる食物状況を検証するためには、サクラやミズナラなどの結実を考慮すべきである。また、今回の分析では、デントコーンを2年以上に渡って利用していた個体や、デントコーンを利用しながらも捕獲されていない個体は検出されなかった。

本研究では、体毛安定同位体比分析を用いた個体レベルでの食性解析によって、ヒグマの採食生態の個体間変異が生じる要因を明らかにした。羅臼町で示された捕獲個体とトラップ個体の食性差異は、個々の採食生態が人との軋轢に関連することを示唆する。一方、斜里町では捕獲個体とトラップ個体の食性に差異は示されず、採食生態という点ではどのメスも人里に出沒する可能性があることが示唆された。この場合、森林に生息するクマが偶発的にでも人里に現れるリスクを減らすような環境づくりが重要である。人慣れ、食物不足、農作物依存は全く異なる軋轢のシナリオであり、地域の環境、軋轢の状況、クマの採食生態の個体差などを考慮し、適切なモニタリングシステムを構築することが重要である。本研究の成果は、野生ヒグマの生態理解を深め、ヒグマの保護管理に新たな視点を提供するものである。