*Limnol. Oceanogr.*, 59(5), 2014, 1581–1589

doi:10.4319/lo.2014.59.5.1581

# Identifying variable sea ice carbon contributions to the Arctic ecosystem: A case study using highly branched isoprenoid lipid biomarkers in Cumberland Sound ringed seals

T. A. Brown,[1,*] C. Alexander,[1] D. J. Yurkowski,[2] S. H. Ferguson,[3] and S. T. Belt [1]

[1] School of Geography, Earth and Environmental Sciences, University of Plymouth, Plymouth, United Kingdom
[2] Great Lakes Institute for Environmental Research, University of Windsor, Windsor, Ontario, Canada
[3] Fisheries and Oceans Canada, Freshwater Institute, Winnipeg, Manitoba, Canada

*Abstract*

We analyzed liver samples from 322 ringed seals (*Pusa hispida*) collected from Cumberland Sound (southeast Baffin Island) to test our ability to differentiate between carbon sources in near apex predators. Highly branched isoprenoids (HBIs) were present in all samples, and their distributions were consistent with recognized seal habitat use. HBI distributions in mature seals ($\geq$ 5 yr) confirmed a less variable carbon source during winter, consistent with geographically restricted sexually mediated territorialism. In contrast, HBI distributions were more variable for immature seals ($<$ 5 yr old), consistent with increased movements and body growth–mediated habitat selection. The ubiquitous presence of sea ice–derived HBIs (e.g., the 'Ice Proxy with 25 carbons') in every seal collected throughout January–December indicates that springtime sea ice primary production remains important for ringed seals throughout the year. HBI distributions remain largely unaltered by trophic transfer, enabling them to document short-term ($<$ 4 weeks) and seasonal changes in carbon. This important characteristic of HBIs facilitated interpretation of sea ice–derived carbon use by seals over annual and interannual timeframes and identified strong associations between sea ice carbon use and insolation as well as sea ice extent. Analysis of HBI distributions could be used to monitor and predict the response of Arctic organisms to reducing sea ice extent and the associated decline in future sea ice primary production over a range of temporal scales.

The strong seasonality in the Arctic is defined by changes in insolation at high latitudes (Walsh 2008). Winter is broadly characterized as being a dark period with frozen seas and limited biological activity. In contrast, summer is characterized by large quantities of daylight, is much warmer, is generally ice-free, and is biologically more productive. The springtime transition from winter to summer represents one of the most important periods of time in the Arctic for animals that have gone through a negative energy balance. A combination of factors, including increasing insolation, temperature, and accumulated nutrients, facilitates large sympagic (ice-associated) algal blooms that stimulate biological activity (Brown et al. 2011). These sympagic blooms play an important role in the initiation of life-stage activities of certain key organisms (Weydmann et al. 2013), thereby defining the timing and structure of the Arctic ecosystem. As sea ice declines under changing climate conditions (Parkinson and Comiso 2013), it is unclear how Arctic ecosystems will be affected by an associated reduction, and potential loss, of spring sympagic primary production. In order to answer this question, it is first necessary to develop methods that are capable of reliably differentiating between springtime sympagic and summertime phytoplanktic primary production throughout the Arctic ecosystem. An emerging approach to achieving this research target is the analysis of highly branched isoprenoid (HBI) diatom lipids, some of which are produced selectively by sea ice diatoms (Brown et al. 2014) during spring (Brown et al. 2011), whereas others are most likely produced by pelagic phytoplankton (Belt et al. 2008).

To date, the extensive characterization of diatom-derived HBIs in numerous marine environments has identified that these lipids are sensitive to the environment in which they are biosynthesized, with some demonstrating a degree of source specificity (Belt et al. 2007). Recent studies have also demonstrated that the analysis of HBIs in some Arctic animals can improve our understanding of how some organisms respond to changes in source primary production (Brown and Belt 2012*a*). It has also been identified that HBIs can be transferred across trophic levels, following their detection in fish (Brown and Belt 2012*b*), birds (Brown et al. 2013*b*), and mammals (Brown et al. 2013*a*), and related observations have been made for some components of Antarctic ecosystems (Goutte et al. 2014). A recent development in the analysis of HBIs in Arctic mammals uses a method that is based on the relative abundances of all HBIs within a sample rather than a comparison of absolute concentrations (Brown et al. in press *b*). Such relative distributions of HBIs are considered to represent an HBI fingerprint, or ''H-Print,'' that is characteristic of the conditions of the environment in which they were produced. However, no Arctic studies to date have provided the opportunity for a thorough assessment of the usefulness of HBI lipids in ecosystems, largely because of the unavailability of sufficiently large data sets for meaningful statistical analysis. Thus, although previous analyses of HBI lipids in Arctic organisms have provided some useful binary information with respect to dietary source consumption (i.e., presence or absence), more detailed studies involving larger sample numbers and better-defined sample sets need to be carried out before the corresponding large-scale ecological interpretations can be made with confidence.

* Corresponding author: thomas.brown@plymouth.ac.uk

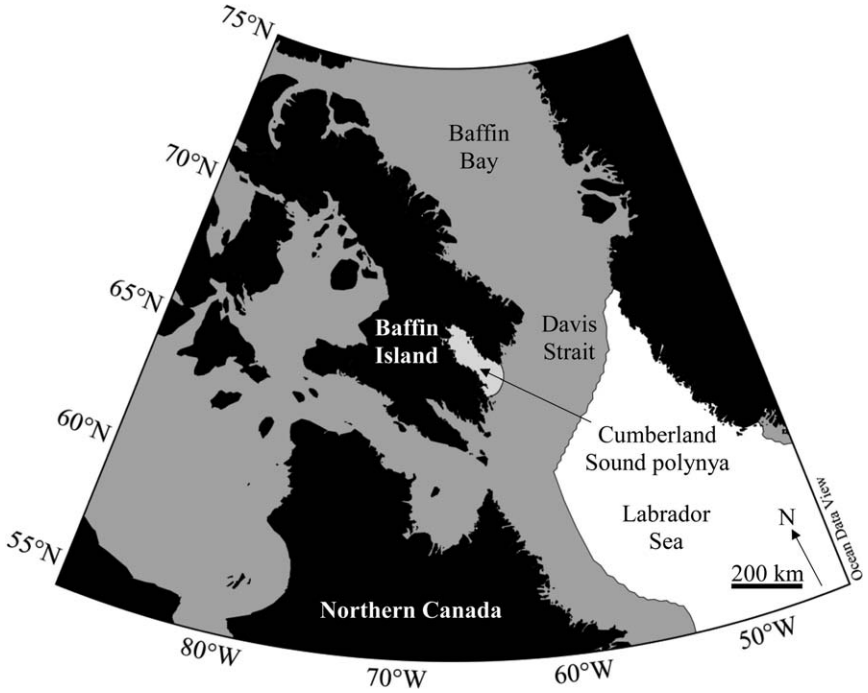


Fig. 1. Map of the study region. Typical March sea ice extent is shaded dark gray. Map from Ocean Data View.

The current study represents a large temporal- and spatial-scale analysis of HBIs within a key Arctic marine mammal (ringed seals [*Pusa hispida*]) and represents the first investigation into the sensitivity of the H-Print approach to reflect known environmental and biometric variables. At the outset, we hypothesized that the dominant influence on HBI distributions in a marine mammal would be determined by seasonal changes in sympagic and phytoplanktic primary production. To test this hypothesis, we measured the H-Prints of 322 ringed seals, and these were compared to available biometric (seal age, length, and mass) and environmental data (month and year sampled as well as the corresponding changes in sea ice and insolation). The H-Print–derived outcomes were also compared to previously defined habitat use identified for ringed seals from other Arctic ecosystems in order to identify spatial patterns.

## Methods

*Geographical setting*—Cumberland Sound, Nunavut, Canada, lies on the southeast coast of Baffin Island at approximately 65°N, 65°W. At 250 km long and 80 km wide, Cumberland Sound is a large inlet containing numerous small islands and glacial fjords.

The Baffin Island Current, comprising the West Greenland Current and Arctic Ocean outflow, flows southward past Cumberland Sound into the Labrador Sea, exporting sea ice and icebergs. As a peripheral Arctic region, Cumberland Sound is characterized by seasonally variable sea ice and is in close proximity to the winter ice edge ($<$ 400 km). During winter, the sea ice conditions in Cumberland Sound are also characterized by the presence of a polynya (Hannah et al. 2009), which maintains thin ice or open-water conditions on a local scale (Fig. 1). Monthly mean sea ice concentrations for the period 1990 to 2011 were obtained for Davis Strait (Canadian Ice Service), Cumberland Sound, and the Arctic (U.S. National Ice Centre) for the years during which seal samples were collected.

*Sample collection*—Three hundred twenty-two *Pusa hispida* (ringed seal) liver samples were obtained from animals collected from Cumberland Sound between 1990 and 2011 by Inuit hunters as part of their subsistence harvests (Table 1). Sampling was carried out in accordance with the community-based monitoring program coordinated by Fisheries and Oceans Canada in Winnipeg, Manitoba, Canada. Liver subsamples were frozen onsite in a freezer at −20°C and were then shipped to Fisheries and Oceans Canada, where they were stored at −30°C. Samples were then freeze-dried (−45°C; 20 Pa; 72 h), ground using a mortar and pestle, and, following homogenization, were further subsampled and sent to Plymouth University prior to analysis.

To group individuals into age classes, seal age has been determined by counting the annual growth layer groups in the cementum of decalcified, stained, longitudinal thin sections of the lower right canine by Barbara Stewart (Fisheries and Oceans Canada; samples prior to 1996) and Matson's Laboratory in Montana (samples from 2007–2011 period) following the method of Bernt et al. (1996) and Stewart et al. (1996). All seals have been grouped into separate age classes: adults ($\geq$ 5 yr of age), sub-adults (1–4 yr of age), and young of the year ($<$ 1 yr of age). Standard length (cm; straight-line distance between nose

Table 1. Summary of ringed seal biometric and H-Print data. SD, standard deviation.

| | | | Biometrics (mean ± 1 SD) | | | | Mean H-Print (% ± 1 SD) | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Year | Season | Frequency | Sex (male : female) | Age (yr) | Mass (kg) | Length (cm) | $IP_{25}$ | IIa | IIb | IIIa | IIIb | IIIc | IIId | Phyto/ice |
| 1990 | Apr–Aug | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| | Sep–Mar | 25 | 1 : 0.4 | 2.0±2.5 | 36±14 | 102±15 | 19±5 | 4±3 | 46±11 | 14±11 | 8±5 | 4±3 | 5±4 | 0.5±0.3 |
| 1991 | Apr–Aug | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| | Sep–Mar | 11 | 1 : 1.8 | 1.2±1.3 | 36±13 | 101±9 | 25±9 | 6±3 | 51±12 | 7±10 | 4±5 | 3±4 | 3±4 | 0.3±0.4 |
| 1992 | Apr–Aug | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| | Sep–Mar | 11 | 1 : 1.2 | 8.1±4.1 | 52±11 | 122±9 | 25±6 | 5±2 | 57±10 | 3±4 | 4±3 | 4±4 | 3±2 | 0.2±0.2 |
| 1993 | Apr–Aug | 7 | 1 : 0.2 | 3.8±2.5 | 39±12 | 112±15 | 22±10 | 5±3 | 48±14 | 6±6 | 8±10 | 5±3 | 6±5 | 0.5±0.8 |
| | Sep–Mar | 72 | 1 : 0.9 | 3.9±3.4 | 41±15 | 111±13 | 22±6 | 4±3 | 55±10 | 5±5 | 5±5 | 4±3 | 5±3 | 0.3±0.3 |
| 1994 | Apr–Aug | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| | Sep–Mar | 2 | 1 : 1 | nd | 48 | 119 | 22 | 3 | 51 | 8 | 8 | 6 | 3 | 0.3 |
| 1996 | Apr–Aug | 32 | 1 : 0.9 | 8.0±8.2 | 39±14 | 114±12 | 21±3 | 7±4 | 43±7 | 6±3 | 9±4 | 8±3 | 6±3 | 0.5±0.1 |
| | Sep–Mar | 57 | 1 : 0.5 | 6.2±6.3 | 41±14 | 113±12 | 21±4 | 5±3 | 52±10 | 4±4 | 6±4 | 6±3 | 5±3 | 0.3±0.2 |
| 2002 | Apr–Aug | 92 | 1 : 0.9 | 3.9±6.1 | nd | 109±17 | 20±7 | 3±3 | 56±12 | 4±5 | 4±3 | 5±5 | 7±6 | 0.3±0.4 |
| | Sep–Mar | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| 2008 | Apr–Aug | 7 | 1 : 0.4 | 0.4±1.1 | nd | 104±17 | 15±6 | 6±4 | 37±4 | 11±10 | 7±4 | 11±2 | 13±6 | 0.8±0.3 |
| | Sep–Mar | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| 2010 | Apr–Aug | 5 | 1 : 0.3 | 1.0±1.2 | nd | 125±7 | 18±7 | 0±1 | 47±5 | 6±3 | 6±4 | 6±2 | 16±8 | 0.5±0.1 |
| | Sep–Mar | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| 2011 | Apr–Aug | 1 | 0 : 1 | 1 | nd | 135 | 23 | 5 | 50 | 5 | 5 | 8 | 4 | 0.3 |
| | Sep–Mar | 0 | — | — | — | — | — | — | — | — | — | — | — | — |
| | Total | 322 | 1 : 0.7 | | | | | | | | | | | |
| | Mean±1 SD | | | 4.4±4.9 | 41±14 | 110±14 | 21±6 | 4±3 | 53±11 | 5±6 | 5±4 | 5±4 | 6±5 | 0.4±0.3 |
| | Median | | | 3 | 40 | 110 | 20 | 4 | 54 | 4 | 4 | 5 | 5 | 0.3 |
| | Maximum | | | 38 | 85 | 147 | 52 | 21 | 100 | 33 | 33 | 26 | 29 | 2.5 |
| | Minimum | | | 0 | 16 | 79 | 4 | 0 | 20 | 0 | 0 | 0 | 0 | 0.1 |

and tail; Mclaren 1993) and body mass (kg) were measured in the field.

*Lipid extraction and purification*—Extraction of HBI lipids from liver subsamples (0.01–1 g) was carried out using established techniques (Belt et al. 2012; Brown et al. 2013*a*). An internal standard (9-octylheptadec-8-ene; 10 $\mu$L; 10 $\mu$g mL$^{-1}$) was added to enable the quantification of HBIs (if required at a later date), according to the method of Belt et al. (2012). Samples were saponified in a methanolic KOH solution (~ 4 mL $H_2O$ : MeOH, 1 : 9; 20% KOH) for 60 min (80°C). Hexane (3 × 4 mL) was added to the saponified solutions, which were then vortexed (1 min) and centrifuged (1 min; 2000 revolutions per minute). Supernatant solutions containing non-saponifiable lipids (NSLs) were transferred to clean vials with glass pipettes and dried ($N_2$ stream) to remove traces of $H_2O$ and MeOH. NSLs were then resuspended in hexane (0.5 mL) and fractionated, providing non-polar (5 mL hexane) lipids using column chromatography ($SiO_2$; 0.5 g), while more polar lipids (e.g., cholesterol) were retained on the columns.

*Lipid analysis*—Analysis of purified non-polar lipid extracts containing HBIs was carried out using gas chromatography–mass spectrometry, according to the method of Belt et al. (2012). Total ion current chromatograms were used to determine the retention time and mass spectra of HBIs, and these were compared with those of authentic standards and published literature for identification purposes.

*Lipid quantification*—HBIs were quantified by measurement of the mass spectral intensities of the molecular ion for each HBI in selective ion monitoring mode (i.e., *m/z* 350.3 for Ice Proxy with 25 carbons [$IP_{25}$], *m/z* 348.3 for IIb, *m/z* 346.3 for IIIa–d;). The analytical intensities of individual HBIs were then normalized according to totals derived from all seven HBIs, with the resulting distribution providing the basis for the H-Print (Brown et al. in press *b*). Here, we adapted the H-Print approach further, by defining it as the ratio of the HBI contributions from planktonic diatoms (Σ IIIa–d) vs. those from sea ice diatoms (Σ $IP_{25}$ and IIb), according to Eq. 1:

$$\text{H-Print} = \frac{(\Sigma\ \text{IIIa} + \text{IIIb} + \text{IIIc} + \text{IIId})}{(\Sigma\ \text{IP}_{25} + \text{IIb})} \quad (1)$$

Individual H-Prints (Eq. 1) were then compared to available biometric and environmental variables.

## Results

*Ringed seal biometrics*—The 322 ringed seal liver samples spanned a 20 yr period between 1990 and 2011. Samples from both males and females (male : female sex ratio = 1 : 0.7) were obtained and comprised a representative range of seal age (0–38 yr), mass (16–85 kg), and length (79–147 cm). All biometric data are summarized in Table 1 and Fig. 2.

Both seal length and mass were normally distributed (Fig. 2) and related (Pearson's correlation; $r$ = 0.6; $p$ = <0.001; $n$ = 219). The age distribution of ringed seal samples was positively skewed toward younger specimens (Fig. 2), and age was found to correlate significantly with mass (Pearson's correlation; $r$ = 0.7; $p$ = <0.001; $n$ = 204). The biometric data confirmed that the chosen samples covered a cross section of individual specimens that is likely to be representative of the broader Cumberland Sound population. This observed generality of biometric distributions

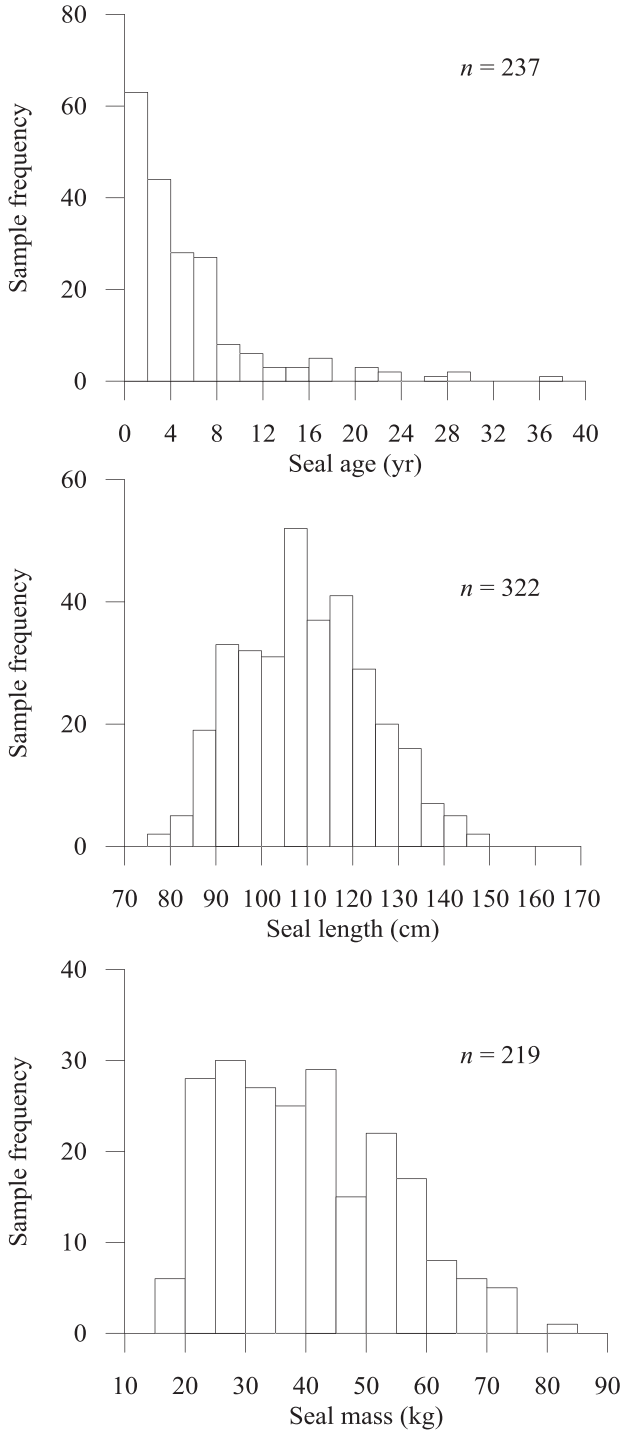


Fig. 2. Frequency plots of biometric data for the ringed seals used in this study.

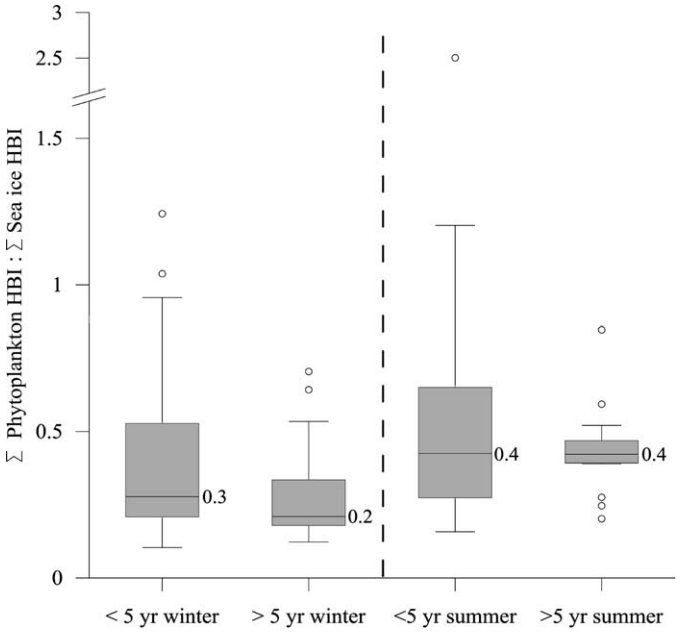


Fig. 3. Box plot of ringed seal H-Prints comparing available data for seal maturity and season collected. Median values are indicated.

increases the likelihood that HBI data obtained from these samples will be representative of the whole community and will not be adversely biased.

*Highly branched isoprenoid lipids in ringed seals*—All ringed seal liver samples contained HBIs, with the sea ice diatom biomarker $IP_{25}$ and a related diene (IIb) being present in every sample. The planktic HBIs (IIIa–d) were present in 282 (87%) samples. Potentially, HBIs IIIa–d may have been present, but below the limit of detection, in some samples as a result of the small sample sizes available ($\sim$ 14% samples = $<$ 50 mg tissue) and their generally lower abundance, compared to $IP_{25}$ and IIb, for the majority of samples (Table 1). However, IIIa–d were also not detected in some samples where much larger sample masses (e.g., 0.5–1 g) were available.

*Lipid variation within individual ringed seals*—Based on previous analyses of southern Arctic (including Cumberland Sound) ringed seal communities, sexual maturity of these seals is reached at 4.6 $\pm$ 0.2 yr of age ($n$ = 195; data on file). Ringed seal H-Prints were found to vary with seal sexual maturity (Mann–Whitney $U$-test; $<$ 5 yr $\neq$ $>$ 5 yr; 95% confidence interval; $p$ = 0.03; $n$ = 172) but not with seal sex (Mann–Whitney $U$-test; 95% confidence interval; $p$ = 0.9; $n$ = 218) and were not strongly correlated with seal mass (Pearson's correlation; $r$ = $-0.2$; $p$ = 0.005; $n$ = 213) or length (Pearson's correlation; $r$ = 0.2; $p$ = 0.001; $n$ = 328).

Although seal maturity is identified as an important consideration, the data resolution did not permit separation of seal maturity groups for a detailed seasonal analysis. As such, we considered seals of all ages in a comprehensive seasonal comparison. Over the 20 yr period, age-related H-Print variation was most evident during winter (October–March; Fig. 3), during which immature seal ($<$ 5 yr old) H-Prints were higher than those of mature seals ($\geq$ 5 yr old; Mann–Whitney $U$-test; 95% confidence interval; $p$ = $<$0.01; $n$ = 116). For the years in which winter and summer seasons were directly comparable (1993 and 1996), immature seals retained a generally higher H-Print than did mature seals. No difference between H-Print and equivalent age class was observed during summer (April–September; Fig. 3; Mann–Whitney $U$-test; 95% confidence interval; $p$ = 0.7; $n$ = 56) over any period of sampling. For both summer and winter, seal H-Prints ranged more widely in immature seals (relative standard deviation $<$ 5 yr; summer = 66%; winter = 77%) compared to mature seals (relative standard deviation $\geq$ 5 yr; summer = 52%; winter = 33%).

Seasonal ringed seal H-Prints were compared to average sea ice concentration and insolation between 1990 and 2011 (Fig. 4). Although positive correlations between the H-Print and sea ice concentration data sets (Davis Strait and Cumberland Sound) were identified, both were lagged by 2 months (lagged correlation; $r$ = $-0.7$; $n$ = 12), with changes in H-Print occurring before the spring melting of sea ice. A further significant correlation was identified whereby increases to the H-Print coincided more closely with a period of daily insolation of $>$ 12 h (Pearson's correlation; $r$ = 0.7; $p$ = 0.02; $n$ = 12), consistent with the expected increased contribution of phytoplanktic HBIs during summer (April–September), both within the Cumberland Sound polynya and at the nearby ice edge (Fig. 1). Finally, a significant interannual relationship (Pearson's correlation; $r$ = $-0.9$; $p$ = 0.05; $n$ = 5) was identified between summer H-Prints and Arctic sea ice concentration (Fig. 5) and was most clearly illustrated by the occurrence of the highest measured summer H-Print (2008) proceeding the lowest recorded sea ice minimum during sampling (2007).

In contrast to these changes in the relative concentrations of phytoplanktic and sea ice diatom HBIs, the ratio of the two sea ice–derived HBIs (IIb and $IP_{25}$) remained remarkably constant in all seals (mean $IIb:IP_{25}$ = 2.7; Pearson's correlation; $r$ = 0.97; $p$ = $<$0.0001; $n$ = 322; Fig. 6).

## Discussion

Previous studies employing HBI analyses within marine ecosystems have represented mainly conceptual studies, using largely opportunistic and somewhat–limited scale data sets (Brown and Belt 2012*b*). As such, the full extent and utility of the ecological advances that could be gained from this type of analysis have remained largely unexplored. The outcomes of the current study provide two significant advances.

We identified HBI lipids in each of a large number (322) of seals analyzed in this study, which advances the initial report by Brown et al. (2013*a*), in which only six ringed seals were analyzed. Further, since samples spanned seals of all ages and were collected from each month of the year over a period of two decades, we have also identified that the incorporation of HBI lipids into ringed seal diet is likely universal.

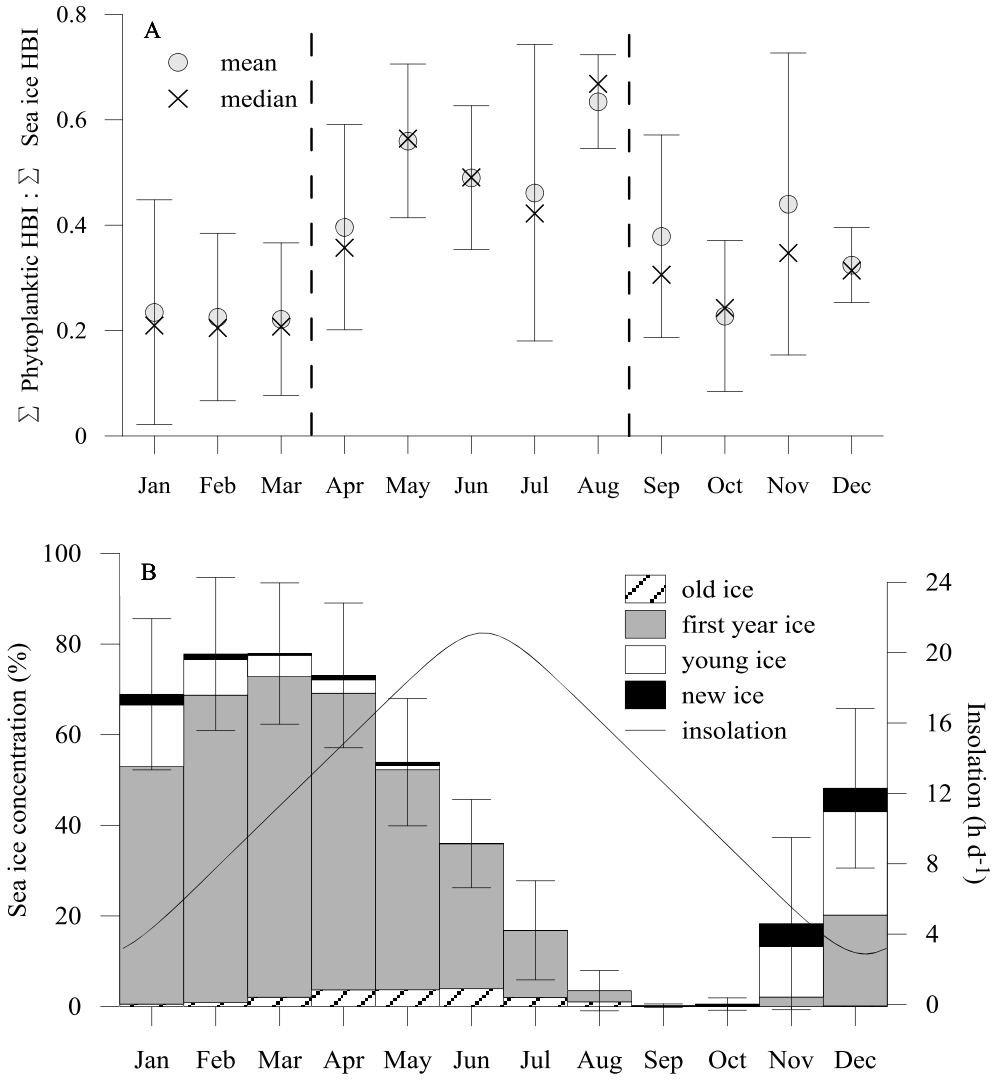


Fig. 4. (A) Mean ± 1 standard deviation (SD) and median H-Prints of ringed seals. Samples from different years are combined to provide a continuous series. Vertical black dashed lines indicate winter and summer seasons. (B) Mean Davis Strait sea ice concentration (Canadian Ice Service) compiled using data only from years during which ringed seals were sampled.

We have shown that distributions of HBIs, as measured using the H-Print approach, can be interpreted positively with respect to habitat use and geographical distributions derived from other methods of analysis including, for example, stable isotopes (Young et al. 2010). Since HBIs were ubiquitous in these near-apex predators, it is also likely that HBIs will be present and ubiquitous in all lower trophic levels, consistent with the findings of previous studies (Brown and Belt 2012*b*; Brown et al. in press *a*,*b*).

*HBI lipid variation and seal biometrics*—The absence of a sex-related difference in H-Prints identified here is consistent with the findings of a similar study that analyzed 192 ringed seals from southern Hudson Bay (Young and Ferguson 2013), in which a seasonal comparison of $\delta^{13}C$ and $\delta^{15}N$ revealed no discernible difference between male and female ringed seals. In contrast, comparison of H-Prints with seal life stages revealed a clear relationship (Fig. 3). Since ringed seal sexual maturity is closely connected with changes in seal habitat use, including increased territorialism (Smith and Hammill 1981; Krafft et al. 2007), this is expected to significantly affect seal feeding habits throughout the year. Indeed, we note that the observed difference in H-Print was more defined during winter (October–March), with mature seals (≥ 5 yr) having consistently lower H-Prints, with a greater relative contribution from the sea ice algal-derived HBIs. In contrast, immature seal (< 5 yr) H-Prints were, on average, higher

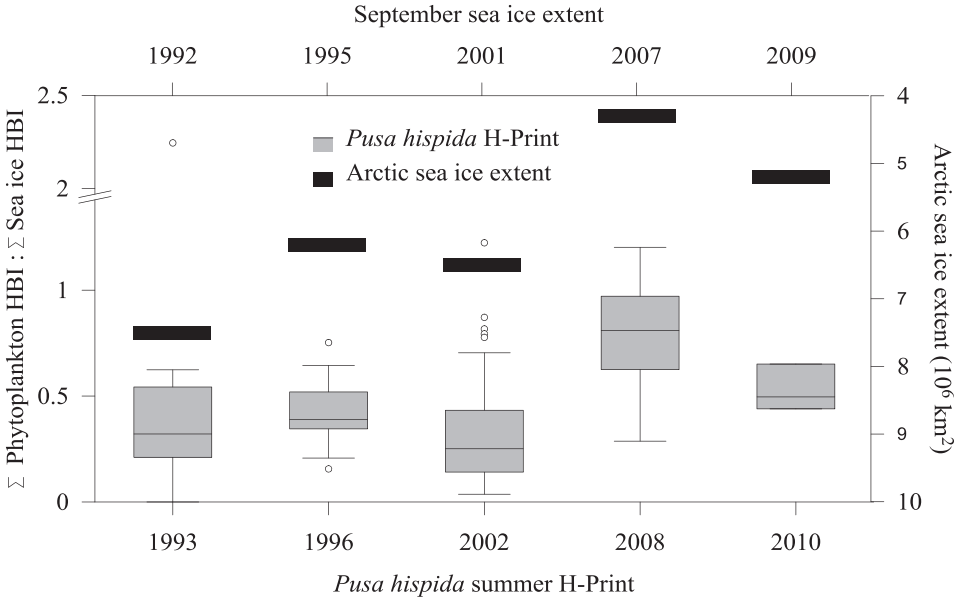


Fig. 5. Interannual comparison of ringed seal H-Prints (gray) with Arctic sea ice extent (black; Pearson's correlation; $r = -0.9$; $p = 0.05$; $n = 5$).

and far more variable during this period, suggesting a substantial difference in habitat use within this cohort. These apparent age-defined H-Prints are consistent with telemetry observations, which identified that mature ringed seal winter ranges in the Western Arctic were typically $< 1$ km, and no more than 30 km (Kelly et al. 2010), resulting in a more consistent under–ice dominated diet. In addition, adult ringed seals exhibited more localized movement behavior than do sub-adults in the Bering and Chukchi Seas during the fall and winter periods (Crawford et al. 2012). Although comprehensive telemetry data were not available for Cumberland Sound seals, one adult male was successfully tagged in Clearwater Fjord in 2011 and remained within 30 km of the tagging location throughout the year (D. J. Yurkowski unpubl.). During summer, the differences between mature and immature ringed seal H-Prints were much less well defined, and both were more variable, indicating increased diversity in diet and habitat use among all seals. Indeed, Kelly et al. (2010) identified that ringed seals were less geographically restricted during summer, with some travelling up to 1800 km from their winter habitats in the western Arctic.

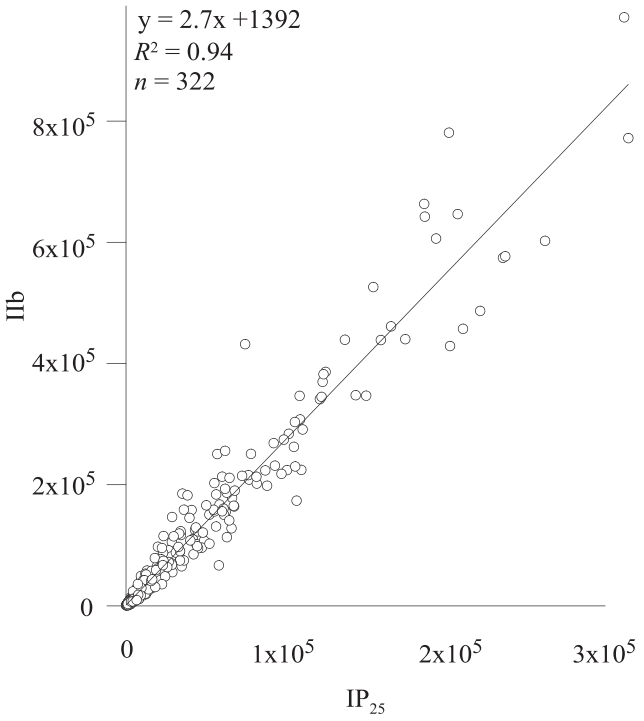


Fig. 6. Plot of $IP_{25}$ vs. IIb showing consistent relationship between these HBIs.

*Seasonal HBI lipid variation in ringed seals*—At the beginning of the calendar year (January–March), seal H-Prints were generally low (0.23), with increased contributions of the sea ice–derived HBIs at this time, consistent with benthic feeding under ice (Brown et al. in press *a*) and limited geographical range. In spring (April–May), there was a distinct increase in the magnitude of the H-Prints (0.39 to 0.56), with greater relative contributions from the phytoplanktic HBIs (Fig. 4), consistent with a hypothesized change in diet to pelagic forage fish at this time (Young and Ferguson 2013). While this transition was not discernable from $\delta^{15}$N data in Hudson Bay ringed seals, a combination of fatty acid and stomach content analysis did indicate a similar change in this nearby community (Young and Ferguson 2013). However, hierarchical cluster analysis using $\delta^{15}$N, $\delta^{13}$C, and fatty acids in Hudson Bay seals suggested that the dietary transition from benthic to pelagic feeding may occur earlier in Cumberland Sound (April–May) than in Hudson Bay (June), despite the fact that ice melt generally occurs later here. This relatively early and

abrupt springtime H-Print transition in Cumberland Sound indicated a consistent early shift in diet from benthic to pelagic sources at this time, which may be related to the greater water depth in Cumberland Sound (cf. Hudson Bay), making pelagic predation preferable to seals. Comparison of these springtime H-Prints to monthly mean Davis Strait and Cumberland Sound sea ice concentration indicated that the spring H-Print transition occurred 1–2 months before significant sea ice decline began. Analysis of satellite-derived sea ice concentrations and Moderate Resolution Imaging Spectroradiometry during this transition, however, revealed the appearance of the Cumberland Sound polynya (Fig. 1), which consistently appears during late March (Barber and Massom 2007), coincident with the transition in measured H-Prints. Since an increased duration of open water is correlated with increased phytoplanktic production (Rysgaard et al. 1999), it is, therefore, possible that the presence of the polynya stimulates sufficient pelagic primary production to induce an ecosystem shift from benthic to pelagic carbon source prior to significant sea ice melt. Comparison of H-Prints with insolation data further supports this idea, since higher H-Prints, indicative of pelagic feeding (0.50), correlated to insolation of $> 12$ h $d^{-1}$, which is generally accepted as the photoperiodic threshold required for significant phytoplanktic cell germination (Eilertsen et al. 1995).

The long-term sampling strategy also provided the opportunity to examine the sensitivity of HBI lipids to interannual variation in sea ice extent. The close relation identified between summer H-Prints and September sea ice extent is indicative of the sensitivity of HBIs to the variable duration of the open-water period (summer). Indeed, reduced sea ice extent can have an effect on the following year's sea ice conditions, leading to increased periods of open water (Giles et al. 2013) and increased phytoplanktic production (Rysgaard et al. 1999). Such a delay was observed here by increases in the H-Print ratio (Eq. 1), indicative of relatively enhanced phytoplanktic production in summer. This effect was most apparent in 2008, when the highest H-Print followed the lowest sea ice extent (2007) within the sampling interval. These data suggest that, with a predicted further reduction in sea ice extent (Giles et al. 2013), seals will likely respond by supplementing their diet with increasing proportions of pelagic prey. In any case, these observations will provide important baseline data against which future trends can continue to be measured.

*Improved understanding of HBI–ecosystem cycling*—The seasonal transition in H-Prints for Cumberland Sound ringed seals, therefore, supports the hypothesis that H-Prints are sensitive to changes in the marine environment and the associated response of sympagic and phytoplanktic primary production. While our current understanding of HBI metabolism in organisms is, at present, limited, the relationship between H-Prints and the associated environmental changes lead us to propose some additional information regarding the behavior of HBI lipids following ingestion.

For example, although turnover rates of HBIs have yet to be determined in detail, it has been shown previously that Arctic zooplankton incorporated HBIs up to 4–6 weeks after production of these lipids was observed in sea ice (Brown and Belt 2012*a*). Here, we observed (in springtime) that changes in ringed seal H-Prints also appear to be sensitive and sufficiently rapid to record dietary changes on a scale of $\sim 4$ weeks, similar to the proposed 1 month turnover for $\delta^{13}C$ in ringed seal muscle (Young and Ferguson 2013). For this change to be recorded in higher trophic-level organisms, in close association with measured environmental changes, indicates that the trophic transfer of HBIs likely occurs rapidly across trophic levels.

Furthermore, a close relationship between the abundances of the two sea ice–derived lipids (IIb and $IP_{25}$) has already been observed in Arctic sea ice and underlying sediments (Belt and Müller 2013), and we identified a similarly significant correlation ($r = 0.97$) between these two HBIs in the ringed seals in the current study. Although the relative metabolic reactivities of individual HBIs have yet to be determined, the consistency in the relative amounts of IIb and $IP_{25}$ in the seal samples indicates that these lipids are not affected differentially by digestion, which supports the suggestion that H-Prints remain largely unaltered during rapid trophic transfer. Although no sediment was available for Cumberland Sound, the $IIb : IP_{25}$ ratio measured in ringed seals was consistent with data for northern Baffin Bay surface sediments ($IIb : IP_{25} =$ 1.9, 2.8, and 3.1; P. Cabedo-Sanz pers. comm.) as well as those measured from other locations in the Canadian Arctic ($2.0 \pm 0.5$ [$n = 11$]; range, 1.3 to 3.1; Belt et al. 2013). Nevertheless, determination of in vitro or in vivo HBI reactivity should be considered an important research target for future studies.

*Acknowledgments*

T.B. thanks Plymouth University for a Post-Doctoral Research Fellowship. We also thank the Pangnirtung Hunters and Trappers Association for collecting ringed seal samples and Robert Stewart and Blair Dunn for their coordination efforts. Ringed seal sample collections were conducted under a Fisheries and Oceans Canada license to fish for scientific purposes (S-02/03 to 11/12-1010-NU-A1) and were supported by the Nunavut Wildlife Research Trust Fund, Fisheries and Oceans Canada, and Natural Sciences and Engineering Research Council of Canada–Ocean Tracking Network (S.H.F. and D.J.Y.). We also thank two anonymous reviewers for their helpful comments.

## References

Barber, D. G., and R. A. Massom. 2007. The role of sea ice in Arctic and Antarctic polynyas, p. 1–43. *In* W. Smith and D. Barber [eds.], Polynyas: Windows to the world. Elsivier Oceanography Series 74. Elsevier Science.

Belt, S. T., T. A. Brown, A. Navarro-Rodriguez, P. Cabedo-Sanz, A. Tonkin, and R. Ingle. 2012. A reproducible method for the extraction, identification and quantification of the Arctic sea ice proxy $IP_{25}$ from marine sediments. Anal. Methods **4:** 705–713, doi:10.1039/c2ay05728j

———, G. Massé, S. J. Rowland, M. Poulin, C. Michel, and B. Leblanc. 2007. A novel chemical fossil of palaeo sea ice: $IP_{25}$. Org. Geochem. **38:** 16–27, doi:10.1016/j.orggeochem.2006.09.013

———, AND J. MÜLLER. 2013. The Arctic sea ice biomarker $IP_{25}$: A review of current understanding, recommendations for future research and applications in palaeo sea ice reconstructions. Quat. Sci. Rev. **79:** 9–25, doi:10.1016/j.quascirev.2012.12.001

———, AND OTHERS. 2008. Distinctive $^{13}C$ isotopic signature distinguishes a novel sea ice biomarker in Arctic sediments and sediment traps. Mar. Chem. **112:** 158–167, doi:10.1016/j.marchem.2008.09.002

BERNT, K. E., M. O. HAMMILL, AND K. M. KOVACS. 1996. Age estimation of grey seals (*Halichoerus grypus*) using incisors. Mar. Mamm. Sci. **12:** 476–482, doi:10.1111/j.1748-7692.1996.tb00602.x

BROWN, T. A., AND S. T. BELT. 2012*a*. Closely linked sea ice–pelagic coupling in the Amundsen Gulf revealed by the sea ice diatom biomarker $IP_{25}$. J. Plankton Res. **34:** 647–654, doi:10.1093/plankt/fbs045

———, AND ———. 2012*b*. Identification of the sea ice diatom biomarker $IP_{25}$ in Arctic benthic macrofauna: Direct evidence for a sea ice diatom diet in Arctic heterotrophs. Polar Biol. **35:** 131–137, doi:10.1007/s00300-011-1045-7

———, ———, A. TATAREK, AND C. J. MUNDY. 2014. Source identification of the Arctic sea ice proxy $IP_{25}$. Nature Comm. **5**, 5197, doi:10.1038/ncomms5197

———, ———, S. H. FERGUSON, D. J. YURKOWSKI, N. J. DAVISON, J. E. F. BARNETT, AND P. D. JEPSON. 2013*a*. Identification of the sea ice diatom biomarker $IP_{25}$ and related lipids in marine mammals: A potential method for investigating regional variations in dietary sources within higher trophic level marine systems. J. Exp. Mar. Biol. Ecol. **441:** 99–104, doi:10.1016/j.jembe.2013.01.020

———, ———, B. PHILIPPE, C. J. MUNDY, G. MASSÉ, M. POULIN, AND M. GOSSELIN. 2011. Temporal and vertical variations of lipid biomarkers during a bottom ice diatom bloom in the Canadian Beaufort Sea: Further evidence for the use of the $IP_{25}$ biomarker as a proxy for spring Arctic sea ice. Polar Biol. **34:** 1857–1868, doi:10.1007/s00300-010-0942-5

———, A. W. J. BICKNELL, S. C. VOTIER, AND S. T. BELT. 2013*b*. Novel molecular fingerprinting of marine avian diet provides a tool for gaining insights into feeding ecology. Environ. Chem. Lett. **11:** 283–288, doi:10.1007/s10311-013-0402-x

———, E. N. HEGSETH, AND S. T. BELT. In press *a*. A biomarker-based investigation of the mid-winter ecosystem in Rijpfjorden, Svalbard. Polar Biol., doi:10.1007/s00300-013-1352-2

———, D. J. YURKOWSKI, S. H. FERGUSON, C. ALEXANDER, AND S. T. BELT. In press *b*. H-Print: A new chemical fingerprinting approach for distinguishing primary production sources in Arctic ecosystems. Environ. Chem. Lett., doi:10.1007/s10311-014-0459-1

CRAWFORD, J., K. FROST, L. QUAKENBUSH, AND A. WHITING. 2012. Different habitat use strategies by subadult and adult ringed seals (*Phoca hispida*) in the Bering and Chukchi seas. Polar Biol. **35:** 241–255, doi:10.1007/s00300-011-1067-1

EILERTSEN, H. C., S. SANDBERG, AND H. TÖLLEFSEN. 1995. Photoperiodic control of diatom spore growth: A theory to explain the onset of phytoplankton blooms. Mar. Ecol. Prog. Ser. **116:** 303–307, doi:10.3354/meps116303

GILES, K., AND OTHERS. 2013. Impacts of climate change on Arctic sea-ice. MCCIP Sci. Rev. 2013, p. 13–19, doi:10.14465/2013.arc02.013-019

GOUTTE, A., Y. CHEREL, C. OZOUF-COSTAZ, C. ROBINEAU, J. LANSHERE, AND G. MASSÉ. 2014. Contribution of sea ice organic matter in the diet of Antarctic fishes: A diatom-specific highly branched isoprenoid approach. Polar Biol. **37:** 903–910, doi:10.1007/s00300-014-1489-7

———, AND OTHERS. 2013. Diatom-specific highly branched isoprenoids as biomarkers in Antarctic consumers. PLOS ONE **8:** e56504, doi:10.1371/journal.pone.0056504

HANNAH, C. G., F. DUPONT, AND M. DUNPHY. 2009. Polynyas and tidal currents in the Canadian Arctic Archipelago. Arctic **62:** 83–95, doi:10.14430/arctic115

KELLY, B., O. BADAJOS, M. KUNNASRANTA, J. MORAN, M. MARTINEZ-BAKKER, D. WARTZOK, AND P. BOVENG. 2010. Seasonal home ranges and fidelity to breeding sites among ringed seals. Polar Biol. **33:** 1095–1109, doi:10.1007/s00300-010-0796-x

KRAFFT, B. A., K. M. KOVACS, AND C. LYDERSEN. 2007. Distribution of sex and age groups of ringed seals *Pusa hispida* in the fast-ice breeding habitat of Kongsfjorden, Svalbard. Mar. Ecol. Prog. Ser. **335:** 199–206, doi:10.3354/meps335199

MCLAREN, I. A. 1993. Growth in pinnipeds. Biol. Rev. **68:** 1–79, doi:10.1111/j.1469-185X.1993.tb00731.x

PARKINSON, C. L., AND J. C. COMISO. 2013. On the 2012 record low Arctic sea ice cover: Combined impact of preconditioning and an August storm. Geophys. Res. Lett. **40:** 1356–1361, doi:10.1002/grl.50349

RYSGAARD, S., T. NIELSEN, AND B. HANSEN. 1999. Seasonal variation in nutrients, pelagic primary production and grazing in a high-Arctic coastal marine ecosystem, Young Sound, Northeast Greenland. Mar. Ecol. Prog. Ser. **179:** 13–25, doi:10.3354/meps179013

SMITH, T. G., AND M. O. HAMMILL. 1981. Ecology of the ringed seal, *Phoca hispida*, in its fast ice breeding habitat. Can. J. Zool. **59:** 966–981, doi:10.1139/z81-135

STEWART, R. E. A., B. E. STEWART, I. STIRLING, AND E. STREET. 1996. Counts of growth layer groups in cementum and dentine in ringed seals (*Phoca hispida*). Mar. Mamm. Sci. **12:** 383–401, doi:10.1111/j.1748-7692.1996.tb00591.x

WALSH, J. E. 2008. Climate of the Arctic marine environment. Ecol. Appl. **18:** S3–S22, doi:10.1890/06-0503.1

WEYDMANN, A., J. E. SØREIDE, S. KWAŚNIEWSKI, E. LEU, S. FALK-PETERSEN, AND J. BERGE. 2013. Ice-related seasonality in zooplankton community composition in a high Arctic fjord. J. Plankton Res. **35:** 831–842, doi:10.1093/plankt/fbt031

YOUNG, B., L. LOSETO, AND S. FERGUSON. 2010. Diet differences among age classes of Arctic seals: Evidence from stable isotope and mercury biomarkers. Polar Biol. **33:** 153–162, doi:10.1007/s00300-009-0693-3

YOUNG, B. G., AND S. H. FERGUSON. 2013. Seasons of the ringed seal: Pelagic open-water hyperphagy, benthic feeding over winter and spring fasting during molt. Wildl. Res. **40:** 52–60, doi:10.1071/WR12168

*Associate editor: Michael R. Landry*

*Received: 18 January 2014*
*Accepted: 23 April 2014*
*Amended: 08 May 2014*