Stable carbon, nitrogen and sulphur isotope analysis of permafrost preserved human hair from rescue excavations (2009, 2010) at the precontact site of Nunalleq, Alaska

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ABSTRACT

The reconstruction of diet and subsistence strategies is integral to understanding hunter-gatherer societies in the past, and is particularly of interest in high latitude environments as they can illuminate human-environmental interactions and adaptations. Until recently, very little archaeological research had been undertaken on the Bering Sea coasts of the Yukon-Kuskokwim Delta, and relatively little is known about precontact lifeways in this region. Here, we present stable carbon, nitrogen and sulphur isotope data from non-mortuary human hair excavated from Nunalleq (c. 1300 CE–1750 CE) – a precontact village site in Western Alaska. Now the focus of a major research project, excavations at Nunalleq began as a rescue excavation, as the site is eroding rapidly into the Bering Sea. Following an initial pilot study on cut strands representing a small number of individuals, a larger body of isotope data has now been generated from the first phase of the investigations of Nunalleq (2009, 2010). These new data, including sulphur isotope values, provide further evidence for the subsistence strategy at the site, including a mixed diet of marine and terrestrial foods (but likely dominated by salmonids). In addition, these new data from Nunalleq highlight some dietary variability amongst the inhabitants of the site. Analyses of additional longer hair strands suggest this variability may not be exclusively due to seasonal variation, and may evidence inter-personal dietary differences. Data from Nunalleq are compared to isotope data from previous studies of Thule-era and earlier Alaskan sites, and to isotope data from Thule sites in Canada and Greenland and the potential of ongoing and future research at the site is discussed, along with the implications for our understanding of Thule subsistence strategies and precontact lifeways on the Yukon-Kuskokwim Delta.

1. Introduction

The Yukon-Kuskokwim Delta, Alaska (Fig. 1) is the core of the Central Yup'ik culture area, which is well-documented ethnographically but poorly characterised archaeologically. This area of coastal Western Alaska, under-researched compared to the more southerly regions of Kodiak (Clark, 1966, 1998; Jordan and Knecht, 1988; Fitzhugh, 2004; West, 2011) or the Aleutian Islands (Laughlin, 1951; Knecht and Davis, 2001, 2008; Balter, 2012), has the potential to reveal important information about the peopling and precontact cultures of Alaska, and past human-environmental relationships along the Bering Sea coast. The need for (and potential of) archaeological investigations in this region has been highlighted by recent excavations at the site of Nunalleq (GDN-248), near the village of Quinhagak, Alaska. Yup'ik for 'the old village', Nunalleq was revealed along an eroding coastline, a product of recent climatic change, which has also led to a reduction in permafrost and more unpredictable weather conditions in the region, along with other impacts such as local infrastructural damage and changes in the abundance, distribution and seasonality of plant and animal resources (Callaway et al., 1999; ACIA, 2004; Hinzman et al., 2005; Moore and Huntington, 2008; Joly et al., 2011). Permafrost and waterlogged soils at Nunalleq have led to the preservation of tens of thousands of *in situ* artefacts at the precontact village site (c.1300 CE–1750 CE), and an extensive assemblage of organic ecofactual and bioarchaeological remains including animal bone, fur, and cut strands of human hair from non-mortuary contexts (Britton et al., 2013; Farrell et al., 2014; Forbes et al., 2015). Following several years of rescue excavations, the site and its landscapes, are now the subject of a major research project and field investigation.

Modern Yup'ik and Cup'ik cultures are assumed to be descended from the Western Thule Tradition which may have originated in Northern Alaska spreading down the coastal Bering Sea as far south as the Alaska Peninsula shortly after 1000 years Before Present (BP) (Dumond, 1984). Indeed, recent mtDNA analysis of human hair from

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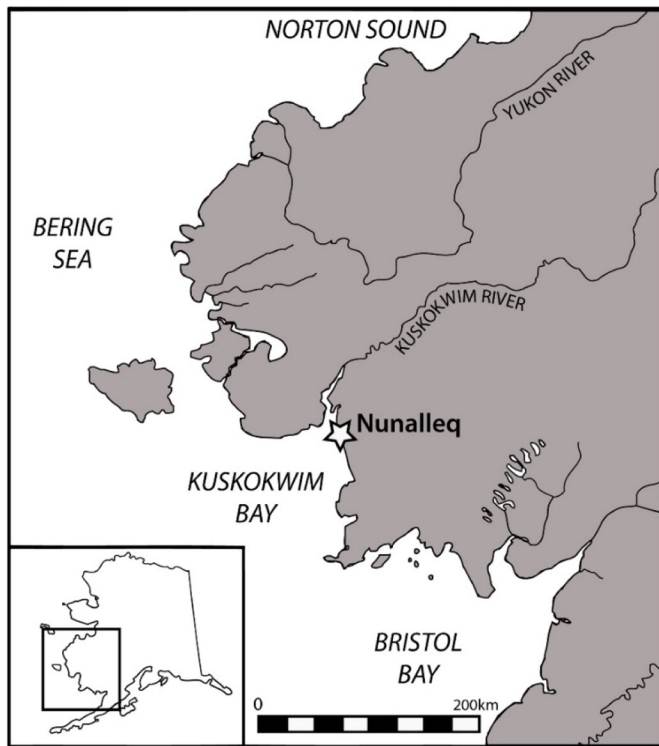E-mail address: k.britton@abdn.ac.uk (K. Britton).

Fig. 1. Map of Western Alaska showing the location of the Nunalleq archaeological site. Adapted from Britton et al. (2013: 449, Fig. 1).

the Thule-era site of Nunalleq, confirms genetic continuity between ancestral and recent populations in the area (Raghavan et al., 2014). Today, and in recent history, traditional Yup'ik subsistence diets incorporate a wide variety of resources including marine mammals, salmonids, caribou, sea birds, sea fish, small mammals and other animals, as well as berries and limited other vegetative foods (Fitzhugh and Kaplan, 1982; Barker and Barker, 1993). However, ethnographic accounts may not accurately reflect the deeper Yup'ik past. This is especially important in a region that has seen profound and dramatic climate changes over the last 1000 years (Mann et al., 1998; Hu et al., 2001), including a period of warming known as the Medieval Warm Phase (~900 CE–1350 CE) to the later cooling of the Little Ice Age (~1350 CE–1900 CE) (Mann, 2007). Inversely, understanding change and continuity in subsistence strategies and dietary habits, especially at latitudinal and climatic extremes, and during periods of environmental change, is important to understanding past human adaptations to those changes, particularly in hunter-gatherer-fisher societies.

In Alaskan archaeology, the reconstruction of past diets and subsistence strategies has relied primarily upon analysis of faunal assemblages; lithic, antler, bone and wooden artefacts; ethnographic accounts and oral histories. While these proxies provide a good picture of general subsistence activities, the stable isotope analysis of human tissues is a useful additional tool in archaeology as it can provide an estimate of foods actually consumed, offering an opportunity to reconstruct the diets of ancient humans directly from their physical remains. Stable isotope data can therefore provide unique dietary perspectives on the scale of individual people, and through intra-tissue, inter-individual and cross-site comparisons, can illuminate past dietary variability on multiple levels. However, few archaeological isotope studies have been undertaken in Alaska (e.g. Coltrain, 2010a; Byers et al., 2011), with only a single pilot study on a small number of archaeological samples in the Yukon-Kuskokwim Delta region (Britton et al., 2013).

Here we present stable carbon, nitrogen and sulphur isotope data from the analysis of permafrost preserved human hair from non-mortuary contexts from the rescue excavations at the site of Nunalleq, Alaska (2009, 2010). Following on from the pilot study, these data provide further evidence for a mixed subsistence strategy, including marine and terrestrial foods, albeit dominated by marine and riverine protein (likely salmonids). In addition to confirming preliminary interpretations of diet at Nunalleq made in a pilot study, the new data presented here also highlight some dietary variability amongst the inhabitants of the Nunalleq site. Analyses of longer hair strands suggest this variability may not be exclusively due to seasonal variation, but may highlight inter-personal dietary differences. Data from Nunalleq are compared to other Alaskan, Canadian and Greenlandic Thule-era and earlier period sites, and variability in late prehistoric diet is explored. The potential of ongoing and future research at the site is discussed, along with the implications for our understanding of Thule subsistence and precontact lifeways on the Yukon-Kuskokwim Delta.

2. Reconstructing palaeodiet using stable isotope analysis

The δ^{13}C, δ^{15}N and δ^{34}S values of bodily proteins such as keratin and bone collagen reflect the δ^{13}C, δ^{15}N and δ^{34}S values of ingested dietary protein (DeNiro and Epstein, 1978, 1981; Schoeninger and DeNiro, 1984; Richards et al., 2003), albeit with minor contributions from other dietary macronutrients (Warinner and Tuross, 2009; Froehle et al., 2010). Different tissues offer different temporal resolution: bone collagen can indicate long-term (~10 years or more) dietary isotopic averages (Ambrose and Norr, 1993; Hedges et al., 2007), whereas incrementally-developed tissues such as teeth (dentinal collagen) and keratinous tissues such as hair/fur, nails and claws offer shorter-term records of protein intake and time-series information (e.g. Roy et al., 2005; Wilson et al., 2007; Beaumont et al., 2013; Montgomery et al., 2013). Human hair grows at a rate of ~1 cm per month (Valkovic, 1977), and therefore can provide intra- and inter-annual records of an individual's dietary habits, although it should be noted a variety of environmental, hormonal, age-related, and other factors can influence this growth rate (Trüeb and Tobin, 2010).

Carbon isotope ratios (δ^{13}C) are used to discriminate between marine and terrestrial dietary protein sources, and nitrogen isotope ratios (δ^{15}N) can be used to determine the trophic level of the protein consumed. In general, δ^{15}N values increase by 3–5‰ with each step up the food chain, whereas δ^{13}C values are enriched by ~1‰. δ^{15}N values can also be used to indicate freshwater or marine dietary inputs, as aquatic ecosystems tend to have longer food chains (Richards et al., 2001b). Therefore, the measurement of δ^{13}C and δ^{15}N values of humans and different animal species can allow the reconstruction of trophic relationships within archaeological ecosystems, and allow the identification of likely sources of human dietary protein.

Sulphur isotope ratios (δ^{34}S) of bone collagen and other body proteins also reflect dietary protein intake and can be used to distinguish between marine and terrestrial influences in the diet. Sulphur isotope ratios in plants and animals are derived from soil sulphur, which is influenced by both the local lithology and rainfall. Terrestrial organisms normally have sulphur isotope values of 5–10‰ (Peterson and Fry, 1987), but can be closer to 0‰ (Nehlich and Richards, 2009), and freshwater organisms exhibit similar values ranging from −5 (or lower) to 10–14‰ (Nriagu et al., 1991; Nehlich and Richards, 2009). Oceanic sulphate has a mean value of ~20‰ (Rees et al., 1978), therefore marine organisms or organisms consuming marine protein are likely to have more elevated δ^{34}S values closer to this value (Peterson and Fry, 1987; Nehlich and Richards, 2009). Sulphur isotope values above 14‰ in mammalian bodily proteins are consistent with a significant marine contribution to the diet (Nehlich, 2015), although values above this 'threshold' (Nehlich, 2015: 10) have also been observed in terrestrial food-webs in coastal regions due to the sea spray effect (Richards et al., 2001a; Nehlich, 2015).

3. Materials and methods

3.1. Materials

Non-mortuary human hair, and accompanying faunal samples, were selected from the site of Nunalleq (GDN-248), close to the village of Quinhagak, Alaska (Fig. 1). Nunalleq was a densely occupied village site of semi-subterranean sod houses. Permafrost and waterlogged soils led to the preservation of tens of thousands of *in situ* artefacts on house floors, and an extensive assemblage of organic ecofactual and bioarchaeological remains, including cut strands of human hair. Cultural attributions at Nunalleq (Western Thule/early Yup'ik) have been made on the basis of the large artefactual assemblages by one of the authors (RK), and associated radiocarbon dates (Britton et al., 2013; this study).

The site has now been excavated for six field seasons (2009, 2010, 2012, 2013, 2014, 2015), and the material utilised in this study (and the previously-published pilot study) originates from the 2009 and 2010 field seasons. The 2009 and 2010 rescue excavations covered an area of 60 m^2 (fifteen 2 × 2 m squares) at the western extent of the site, an area which has subsequently been eroded away entirely – with the artefactual and ecofactual assemblage, and site records, being all that remain of this portion of the site. Although areas of the site (excavated in more recent years) have utilised single context recording (where each stratigraphic unit or 'context' is recorded and described independently, allowing a site matrix to be produced; see Branch et al., 2005: 34), this western area was excavated as levels, numbered sequentially from Level 1 (the sod and topsoil) downwards, reflecting the logistics of a rescue excavation with a small field crew and the associated time-constraints.

A total of nineteen samples (comprising plant remains and caribou [*Rangifer tarandus*] bone collagen) were submitted for AMS ^{14}C dating at Beta Analytic, Miami and SUERC, East Kilbride (Supplementary Information, SI Table 1) from the 2009–2010 field seasons. Dates were calibrated in OxCal (Bronk Ramsey, 2009) using the IntCal13 calibration curve (Reimer et al., 2013). These samples derive exclusively from the 2009 and 2010 field seasons and cover all areas and levels of the site excavated during these years. The dates ranged from 650 ± 40 BP (Beta-263581; wood; 1270–1400 cal CE [2σ]; δ^{13}C = −25.2‰) to 182 ± 37 BP (SUERC-54993; caribou bone collagen; 1490–1670 cal CE [2σ]; δ^{13}C = −17.8‰), with the majority of dates suggesting a main phase of occupation in the 15th–17th Centuries. A simple Bayesian model (Supplementary information, SI Fig. 1) constructed in OxCal (Bronk Ramsey, 2009), which constrains all of the dates within a single occupation phase, provides a similar result but also suggests a potentially longer period of site use (with occupation beginning between 1300 and 1405 cal CE and persisting until 1650–1750 cal CE). This modelled date for abandonment is consistent with the absence of Euroamerican material culture at the site. However, it is important to note that the oldest date is on wood, which can be a problematic dating material at Arctic sites such as Nunalleq where trees are absent in the immediate environment. The majority of wood from the archaeological deposits at Nunalleq is driftwood, which would have been deposited on beaches by, *inter alia*, ocean currents, melting permafrost or rivers from the interior, and therefore has an 'in-built' age (*sensu* McFadgen, 1982) reflecting the death of the tree rather than its deposition at the site. Despite these concerns, a similar date was also determined from plant material (grass) (Beta-308742; 570 ± 30 BP; 1300–1430 cal CE [2σ]; δ^{13}C = −22.3‰), a short-lived terrestrial macrofossil, suggesting that an earlier 14th Century phase at the site is plausible.

Hair samples selected for this study consisted of short discrete locks of cut human hair (2–6 cm in length) and longer locks (~8–14 cm), excavated *in situ* from preserved house-floors. Their origin is non-mortuary: the hair most likely represents domestic debris accidentally or intentionally discarded following haircuts. Numbers of samples from the different squares and levels are broadly representative of the distribution and density of human hair across the area excavated. Sub

samples of groups of hairs (30–150 hairs) from the same lock were selected and prepared for 'bulk' analysis. Samples were judged to be from different individuals on the basis of their different cut lengths, texture, colour, and strand thickness. However, as the hair growth cycle is influenced by macro- and micro-environmental; hormonal; age-related and other systematic changes (Trüeb and Tobin, 2010), changes in these features might be expected across the human scalp or during different phases of an individual's life, and duplication in sampling (without genetic screening) was therefore impossible to avoid. Human hair grows at a rate of ~1 cm per month (Valkovic, 1977), therefore these 'bulk' samples could represent a dietary history of between 2 and 6 months for the different individuals. The sequential analysis of the longer strands was undertaken in order to help explore potential seasonal dietary differences that may affect the shorter 'bulk' samples and, through this, inter-individual variation. New data in this study (n = 51, including 7 longer strands) are added to the pilot dataset published in Britton et al., 2013 (n = 6, including a single longer strand), and all data (n = 57) are analysed and discussed collectively here. Faunal data utilised in this study originate predominantly from research undertaken as part of a recent doctoral thesis by one of the co-authors (EM-F; McManus-Fry, 2015, also presented in McManus-Fry et al., 2016 [this volume]), and is combined here with previously published faunal data (Britton et al., 2013).

3.2. Methods

Human hair samples were prepared for isotope analysis at the Department of Archaeology, University of Aberdeen (Aberdeen, UK) and measured at the Department of Human Evolution, Max Planck Institute for Evolutionary Anthropology (Leipzig, Germany). Human hair samples were prepared for isotopic analysis using standard protocols to degrease and clean samples (O'Connell and Hedges, 1999; O'Connell et al., 2001; Hedges et al., 2005). Samples were initially rinsed in deionised water to remove large pieces of debris. This was followed by ultrasonication in 2:1 methanol/chloroform solution (30 min; 3×) and then 1:2 methanol/chloroform solution (30 min; 3×) to remove lipids. Finally, samples were ultrasonicated in deionised water (30 min; 3×) to remove any remaining methanol and chloroform from the hair samples. All samples were then freeze-dried for 12–24 h. The shorter 'bulk' hair samples were analysed as individual or multiple fibre segments (1–5 fibres per analysis, dependent on mass). For the longer hair locks, which were secured with aluminium foil during the cleaning process, each shaft was orientated (aligned to the cut end), and sectioned into 5 mm serial segments using a scalpel. Serial samples were then analysed as grouped-fibre 5 mm segments (number of fibres dependent on mass; where insufficient sample existed, adjacent 5 mm serial segments were combined). Faunal bone collagen was extracted using a modified Longin method, with the addition of an ultrafiltration step, as detailed in a previous publication (Britton et al., 2013).

Samples were weighed into tin capsules for carbon and nitrogen isotope analysis. Samples for sulphur isotope analysis were weighed into tin capsules with vanadium pentoxide. Carbon, nitrogen and sulphur contents, and carbon and nitrogen isotope ratios (δ^{13}C, δ^{15}N) were determined using EA/CF-IRMS (ThermoFinnigan Flash EA 22112 coupled to a Delta Plus XP isotope ratio mass spectrometer). δ^{13}C values are reported relative to the Vienna Pee Dee Belemnite (V-PDB) standard, and δ^{15}N values were measured relative to the ambient inhalable reservoir (AIR) standard. Sulphur isotope ratios (δ^{34}S) were also determined using EA/CF-IRMS (HekaTek Euro Vector coupled to a Delta V Plus isotope ratio mass spectrometer), and were measured relative to international standards (S1, S2, S3, SO-5 and NBS127). %C, %N and %S refer to the percentage of carbon, nitrogen and sulphur measured during the analysis of each sample compared to the original sample mass. Measurement error on the δ^{13}C and δ^{15}N values was calculated from the repeat measurements of internal and international standards and was ± 0.2‰ (1σ) or better, and for δ^{34}S measurements was ± 0.5‰ (1σ).

4. Results and discussion

The stable isotope data (along with the %C, %N, C:N ratio, and %S, C:S and N:S) of all 'bulk' samples are shown in Table 1. The 'bulk' δ^{13}C and δ^{15}N values of the human hair presented in this study (along with previously-published pilot data, including mean values of longer strands) are plotted in Fig. 2, where data are compared to faunal bone collagen isotope data from Nunalleq (summary data shown in Table 2, taken from Britton et al., 2013; McManus-Fry, 2015; McManus-Fry et al., 2016 [this volume]). The input data (from major prey species/ taxa groups) and results of a linear mixing model are shown in Table 3 and Fig. 3 respectively. 'Bulk' δ^{34}S data are shown in Figs. 4, and 5 (with sulphur % content). The δ^{13}C and δ^{15}N data from the longer

strands of human hair are provided in the supplementary information (SI Table 2), and are shown in Fig. 6a–h, and combined in Fig. 7a–b. Mean 'bulk' δ^{13}C and δ^{15}N data from Nunalleq are compared to previously-published archaeological stable isotope data from a range of other Alaskan and Aleutian sites in Fig. 8, and to other Thule-era Arctic/sub-Arctic sites (in Alaska, Canada and Greenland) in Fig. 9.

4.1. 'Bulk' human hair isotope data at Nunalleq

4.1.1. 'Bulk' carbon and nitrogen isotope data

Mean δ^{13}C and δ^{15}N values for the 'bulk' hair samples from Nunalleq (including mean values calculated from the sequential analysis of the longer strands) are -15.7 ± 0.4‰ (1σ) and 16.5 ± 1.1‰ (1σ)

Table 1

Stable carbon, nitrogen and sulphur isotope data from human hair from Nunalleq, incorporating data from a previous study (Britton et al., 2013; denoted with asterisk), including %C, %N and %S, and atomic C:N, C:S and N:S ratios.

| Sample ID	Length (mm)	Square/level	δ^{13}C	δ^{15}N	%C	%N	C:N	δ^{34}S	%S	C:S	N:S
*19182	45	Sq1 L3	−16.4	14.9	45.2	15.0	3.5	14.9	4.3	29.8	8.5
*19178	45	Sq1 L4	−15.1	17.4	44.2	14.7	3.5	14.9	4.5	27.9	8.0
*19179	50–60	Sq1 L4	−15.6	16.3	43.7	14.8	3.5	15.6	5.1	24.5	7.1
*17790	105	Sq1 L4	−15.9	13.7	43.3	14.2	3.6	–	–	–	–
21387	36	Sq1 L4	−15.1	18.4	45.0	15.1	3.5	14.2	5.9	21.6	6.2
21502	24	Sq1 L4	−15.9	16.9	45.2	14.8	3.6	15.1	5.1	24.9	7.0
21509	27	Sq1 L4	−15.8	16.6	45.5	14.8	3.6	13.5	5.4	23.8	6.7
*19180	35	Sq2 L4	−15.5	17.1	44.2	14.8	3.5	13.9	5.0	25.1	7.2
*19181	35–40	Sq3 L3	−15.8	16.5	43.1	14.5	3.5	15.8	5.3	23.1	6.7
21511	40	Sq5 L5	−16.1	17.4	44.0	14.4	3.6	14.5	5.3	23.5	6.6
21523	130	Sq6 L4	−15.7	11.9	43.9	14.3	3.6	–	–	–	–
21359	27	Sq7 L2	−15.2	17.7	45.8	15.2	3.5	13.6	5.0	25.7	7.3
21513	61	Sq7 L4	−15.2	17.1	46.0	15.4	3.5	15.3	4.8	27.0	7.7
21521	113	Sq7 L4	−15.7	17.4	44.8	14.5	3.6	–	–	–	–
21371	34	Sq8 L3	−15.4	16.0	44.9	15.3	3.4	16.1	5.9	21.5	6.3
21380	22	Sq8 L3	−15.5	16.2	44.6	15.1	3.5	15.5	5.7	22.2	6.4
21516	137	Sq 8 L3	−15.7	13.6	45.4	14.5	3.7	–	–	–	–
21385	29	Sq 8 L3	−15.8	15.9	44.2	15.1	3.4	13.2	5.4	23.3	6.8
21358	49	Sq8 L4	−15.4	17.4	45.0	15.2	3.4	15.9	7.6	16.8	4.9
21514	42	Sq8 L4	−15.8	17.5	44.9	15.2	3.4	15.6	5.8	22.0	6.4
21378	32	Sq9 L3	−15.8	16.2	44.3	14.9	3.5	14.9	5.5	22.8	6.6
21365	52	Sq10 L3	−17.1	15.3	46.6	15.1	3.6	14.0	5.1	25.9	7.2
21367	19	Sq10 L3	−15.3	16.8	44.7	15.1	3.5	14.8	5.7	22.3	6.4
21370	22	Sq10 L3	−15.9	16.2	45.3	15.3	3.4	14.7	5.9	21.6	6.3
21383	51	Sq10 L3	−16.0	15.4	45.0	14.9	3.5	14.6	5.2	24.5	7.0
21384	40	Sq10 L3	−15.7	16.3	44.6	15.2	3.4	15.8	5.7	22.0	6.4
21386	31	Sq10 L4	−15.7	17.4	41.6	14.1	3.4	15.1	6.5	18.2	5.3
21503	57	Sq10 L4	−15.9	16.7	45.2	15.3	3.5	16.0	4.8	26.7	7.7
21504	35	Sq10 L4	−15.8	15.7	44.0	15.0	3.4	14.9	5.4	23.2	6.8
21505	32	Sq10 L4	−15.5	16.2	44.3	15.1	3.4	16.4	5.9	21.1	6.2
21507	34	Sq10 L4	−15.5	16.2	45.3	15.4	3.4	16.3	5.8	22.0	6.4
21508	38	Sq10 L4	−15.5	17.2	44.0	14.9	3.4	14.3	4.9	25.5	7.4
21360	32	Sq11 L2	−15.7	15.5	46.5	15.6	3.5	15.5	4.9	27.0	7.8
21361	42	Sq11 L2	−15.9	15.5	46.9	15.6	3.5	15.7	5.0	26.5	7.5
21377	52	Sq11 L3	−15.1	17.3	42.7	14.5	3.4	14.5	5.2	23.2	6.8
21379	31	Sq11 L3	−15.3	16.2	44.0	15.0	3.4	14.2	5.7	22.0	6.4
21368	18	Sq11 L3	−15.5	16.3	46.3	15.1	3.6	16.2	4.8	27.3	7.6
21363	28	Sq11 L3	−15.6	17.2	45.6	15.3	3.5	14.5	4.9	26.6	7.6
21366	59	Sq11 L3	−15.7	16.3	44.0	14.9	3.4	15.3	4.9	25.7	7.4
21372	25	Sq11 L3	−15.5	16.7	45.6	15.4	3.5	14.4	4.8	26.7	7.7
21374	55	Sq11 L3	−15.2	17.6	44.9	15.2	3.4	14.5	5.3	24.1	7.0
21375	28	Sq11 L3	−15.4	17.0	44.5	15.1	3.4	15.4	5.3	23.6	6.9
21381	39	Sq11 L3	−15.7	17.0	45.0	15.2	3.5	12.9	5.2	24.6	7.1
21382	35	Sq11 L3	−15.7	15.8	44.5	15.1	3.4	13.5	5.2	24.2	7.1
21518	73	Sq11 L3	−15.1	16.3	45.5	15.3	3.5	–	–	–	–
21519	89	Sq11 L3	−15.1	17.7	45.1	14.7	3.6	–	–	–	–
21506	50	Sq11 L4	−15.4	16.1	44.6	15.1	3.4	15.6	5.1	24.9	7.2
21510	32	Sq11 L4	−15.5	16.7	44.0	14.7	3.5	14.8	5.6	22.2	6.4
21369	34	Sq12 L3	−15.7	17.9	44.8	14.6	3.6	14.7	5.1	24.9	6.9
21373	30	Sq12 L3	−15.9	16.5	45.2	15.0	3.5	15.1	4.8	26.9	7.7
21517	76	Sq12 L3	−15.4	14.5	45.1	14.9	3.5	–	–	–	–
21520	78	Sq12 L4	−16.1	16.1	45.2	15.2	3.5	16.0	5.0	25.7	7.4
21364	31	Sq14 L3	−16.1	17.3	46.2	15.1	3.6	14.0	4.9	26.9	7.5
21515	36	Sq14 L3	−15.7	18.2	46.1	14.7	3.6	14.9	5.5	23.5	6.5
21376	31	Sq14 L3	−15.8	17.0	44.2	14.7	3.5	14.4	5.2	24.2	6.9
21522	130	Sq15 L3	−16.8	17.0	43.6	14.6	3.5	–	–	–	–
21362	47	Sq16 L3	−15.4	17.2	38.1	13.1	3.4	14.3	5.2	20.8	6.1
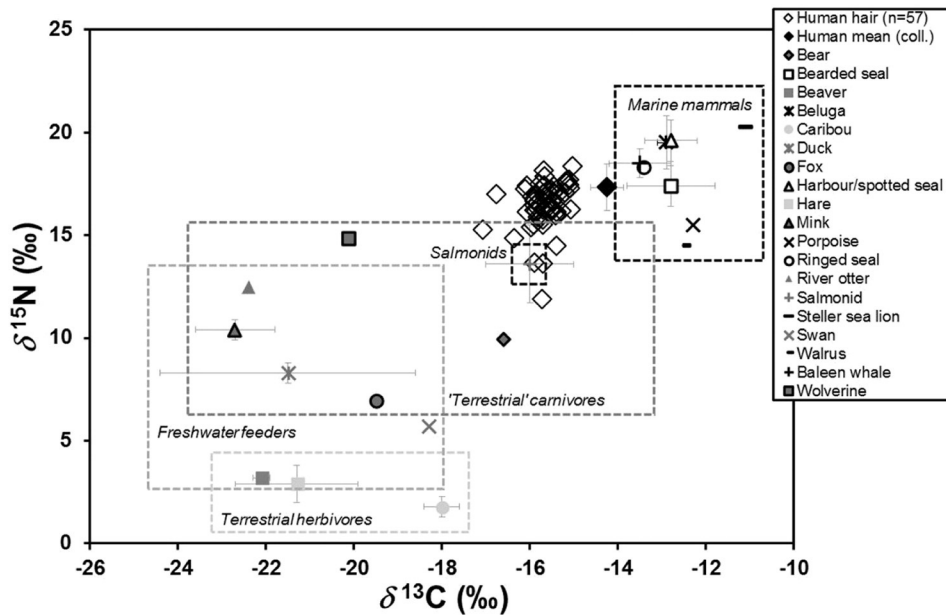
Fig. 2. Human hair (Britton et al., 2013, and this study) and faunal bone collagen (Britton et al., 2013; McManus-Fry, 2015; McManus-Fry et al., 2016 [this volume]) stable carbon and nitrogen isotope data from Nunalleq. In addition to measured human hair values, a *predicted* collagen datapoint (mean) is also shown for humans. Measurement error was ±0.2‰ (1σ) or better. Error bars show ±1σ for mean values.

respectively. These averages, based on the analysis of 57 samples (incorporating the six strands from the previously pilot study), are very similar to those previously reported from the site (−15.7 ± 04‰ and 16.0 ± 1.4‰, $n = 6$; Britton et al., 2013). $\delta^{13}C$ values range from −17.1‰ to −15.1‰, and $\delta^{15}N$ values range from 11.9‰ to 18.4‰. All samples have C:N ratios within the range of modern hair (3.0 to 3.8; O'Connell and Hedges, 1999), and can therefore be considered to have a good level of preservation.

Human hair carbon and nitrogen isotope data, and that from local fauna, are compared in Fig. 2.

Owing to differences in the amino acid composition between keratin and collagen (specifically the higher proportion of the amino acid glycine in bone collagen, which is enriched in ^{13}C relative to other amino acids), a stable isotope ratio offset is expected between these two analytes. Although paired analyses have demonstrated that bone

Table 2
Summary stable carbon and nitrogen isotope data from faunal bone collagen from Nunalleq (taken from McManus-Fry, 2015, McManus-Fry et al., 2016 [this volume], and incorporating data from Britton et al., 2013). Species identifications were made by one of the authors (EM-F).

Taxa	Species	n	$\delta^{13}C$ (±1σ)	$\delta^{15}N$ (±1σ)
Bear	*Ursus* sp.	1	−16.6	9.9
Beaver	*Castor canadensis*	7	−22.1 (±0.2)	3.2 (±0.2)
Caribou	*Rangifer tarandus*	18	−18.0 (±0.4)	1.8 (±0.5)
Fox	*Vulpes vulpes*	2	−19.5 (±0.0)	6.9 (±0.3)
Hare	*Lepus othus*	4	−21.3 (±1.4)	2.9 (±0.9)
American mink	*Neovison vison*	4	−22.7 (±0.9)	10.4 (±0.5)
River otter	*Lontra canadensis*	1	−22.4	12.5
Wolverine	*Gulo gulo*	1	−20.1	14.8
Baleen whale	*Mysticeti* sp.	4	−13.5 (±0.7)	18.5 (±0.7)
Bearded seal	*Erignathus barbatus*	7	−12.8 (±1.0)	17.4 (±1.0)
Beluga whale	*Delphinapterus leucas*	3	−12.9 (±0.2)	19.5 (±1.3)
Duck	*Anatidae* sp.	2	−21.5 (±2.9)	8.3 (±0.5)
Harbour/spotted seal	*Phoca vitulina/largha*	4	−12.8 (±0.6)	19.6 (±1.0)
Porpoise	*Phocoena phocoena*	1	−12.3	15.5
Ringed seal	*Pusa hispida*	8	−13.4 (±1.0)	18.3 (±0.9)
Salmonid	*Oncorhynchus* sp.	2	−16.0 (±1.0)	13.6 (±1.9)
Steller sea lion	*Eumetopias jubatus*	1	−11.1	20.3
Phocid seal	*Phoca* sp.	3	−13.4 (±1.0)	18.3 (±1.9)
Tundra swan	*Cygnus columbianus*	1	−18.3	5.7
Walrus	*Odobenus rosmarus*	3	−12.5 (±0.4)	14.5 (±0.2)

collagen-hair keratin differences are minimal (0–2‰ in both $\delta^{13}C$ and $\delta^{15}N$; O'Connell and Hedges, 1999; O'Connell et al., 2001), and not likely to alter the interpretation of broad dietary trends based on cross-tissue comparisons (i.e. faunal collagen to human keratin), keratin values can be adjusted to predicted collagen values for like-to-like (faunal collagen to human collagen) comparisons. Therefore, a predicted mean collagen value (±1σ) is also shown (see Fig. 2), calculated from the mean keratin value of all individuals ($n = 57$) based on the bone-keratin offsets reported for paired-tissue analyses in a modern population reported by O'Connell et al. (2001; +1.41‰ and +0.86‰ for carbon and nitrogen respectively). When compared to the accompanying terrestrial and marine faunal data, it is likely that the late prehistoric diet at Nunalleq included high trophic level marine protein (such as pinnipeds), but also incorporated significant contributions from lower trophic level marine protein sources such as sea fish or shellfish, or migratory fish such as salmonids, and/or terrestrial protein such as caribou meat. Nearby rivers host significant runs of anadromous fish today and historically (Fitzhugh and Kaplan, 1982; Barker and Barker, 1993), and mixed but marine-/salmonid-dominated economy is consistent with historical and ethnographic accounts of Yup'ik diet in the Yukon-Kuskokwim Delta, with caribou hunting and salmon fishing in particular remaining economic activities in the region today (Fitzhugh and Kaplan, 1982; Barker and Barker, 1993; Callaway et al., 1999).

While the standard deviations (1σ) for $\delta^{13}C$ are low at Nunalleq, suggesting dietary homogeneity amongst individuals, the $\delta^{15}N$ data demonstrate a little more spread as shown in Fig. 2 and there are some outliers, with lower or more elevated values relative to the majority of the dataset. Given the broad chronological grouping the samples included in this study likely fall into, a more refined assessment of diachronic variation is not possible at this time. It is hoped further analysis of more recently excavated materials from other areas of the Nunalleq site (recorded and provenienced using single context recording, matrix analyses and age-depth modelling), will allow a targeted investigation of any possible diachronic dietary shifts before and during the Little Ice Age at Nunalleq. Similarly, the determining of biological sex in samples would provide greater insight into inter-individual variation and past diet at the site. Although the hair from Nunalleq is cut (lacking roots), previous research has demonstrated it is possible to obtain the whole genome, including the sex chromosomes, from hair shafts (Rasmussen et al.,

Table 3
Mean stable isotope data for potential food-source taxa/major prey-groups; adjusted IsoSource input values (see text for details); and summary IsoSource mass balance data for Nunalleq. Mean faunal data was compiled from Britton et al., 2013, McManus-Fry, 2015, McManus-Fry et al., 2016 [this volume], and including additional salmonid and marine fish values from Byers et al., 2011. IsoSource increments were set at 5% and the tolerance at 0.7%.

Taxa/group	n	Measured mean values		Adjusted IsoSource input values		Proportional contribution to diet		
		$\delta^{13}C$ (‰)	$\delta^{15}N$ (‰)	$\delta^{13}C$ (‰)	$\delta^{15}N$ (‰)	Min (%)	Mean (%)	Max (%)
Nunalleq Humans	57	−15.7	16.5	−14.2	17.3	–	–	–
Pinnipeds (seals)	22	−13.1	18.2	−12.1	21.2	0.0	18.6	60.0
Cetaceans	8	−13.1	18.5	−12.1	21.5	10.0	22.8	65.0
Salmonids	7	−15.3	12.2	−14.3	15.2	0.0	48.3	75.0
Marine fish	29	−11.6	15.6	−10.6	18.6	0.0	1.3	5.0
Caribou	18	−18.0	1.8	−17.0	4.8	0.0	9.0	25.0

2010, 2011), particularly in cases of good preservation (such as permafrost).

4.1.2. A linear mixing model at Nunalleq

While the main sources of dietary protein can be estimated from 'bulk' isotopic data, and inferred from the range of fauna found in the assemblage at the site (McManus-Fry, 2015; McManus-Fry et al., 2016 [this volume]), the application of a linear mixing model (IsoSource 1.3.1; Phillips and Gregg, 2003) can be useful in further evaluating trends observed. This can also be useful in comparing data here to other Thule-era studies in Canada and the Aleutians (see Section 4.3), where such approaches are common (e.g. Coltrain, 2009; Byers et al., 2011). The IsoSource model examines combinations of potential source inputs from different prey types in order to identify possible contributions from each source to the human diet. It should be noted that the mixing model undertaken here does not take into account other dietary macronutrients, such as carbohydrates or fats, but instead seeks to estimate the composition of dietary protein. This is done by comparing the isotope values of protein of possible animal foods (modified to account for trophic level enrichment) to human protein isotope values (which largely reflect the isotopic values of proteins consumed) to determine a relative contribution (%). This is of course not likely to reflect whole dietary composition: while starchy plant foods (and thus carbohydrate) are likely to have been scarce in the region, dietary fat content was probably high (as with contemporary traditional Arctic diets) and very high protein diets are unsustainable (Speth and Spielmann, 1983; Cordain et al., 2000). Summary mean isotopic values for a range of taxa types/ groups (Table 3) were determined from site faunal isotope data

(Britton et al., 2013; McManus-Fry, 2015; McManus-Fry et al., 2016 [this volume]), with marine fish (near-shore and open water species) and additional salmonid values being contributed from another archaeological (Aleutian) study (Byers et al., 2011). All mean food source $\delta^{15}N$ values were adjusted by +3‰ (Coltrain, 2009: 770) and by +1‰ for $\delta^{13}C$ (Byers et al., 2011) to account for trophic level enrichment. In order for animal bone collagen isotopic values and human isotope data to be directly comparable, the predicted mean bone collagen human value was used in the mixing model (see above). IsoSource increments were set at 5% and tolerance at 0.7%, and a summary of the results is shown in Fig. 3.

Based on data inputted, the programme calculated 114 mass balance diets for Nunalleq. The mean diet includes contributions from all the input prey taxa/groups, and is dominated by salmonids (48.3%), cetaceans (22.8%), and seal (18.6%). Caribou meat and marine fish likely made less of a contribution (9.0% and 1.3% respectively). These data provide further support to primary isotope data interpretation, that diet at Nunalleq incorporates a mixture of different animal resources, including lower trophic level marine resources such as salmonids, marine mammals, with a minor contribution of terrestrial protein. It should be noted, however, that while useful in helping to quantify relative dietary inputs, the results of mixing models must always be treated with caution, particularly when applied to archaeological sites, and results should always be interpreted within the broader framework of evidence. In ecology, such modelling approaches are common and useful, but not unproblematic (Phillips and Gregg, 2003; Bond and Diamond, 2011; Phillips et al., 2014). In archaeological case studies the use of mixing models is perhaps yet more problematic, given that many
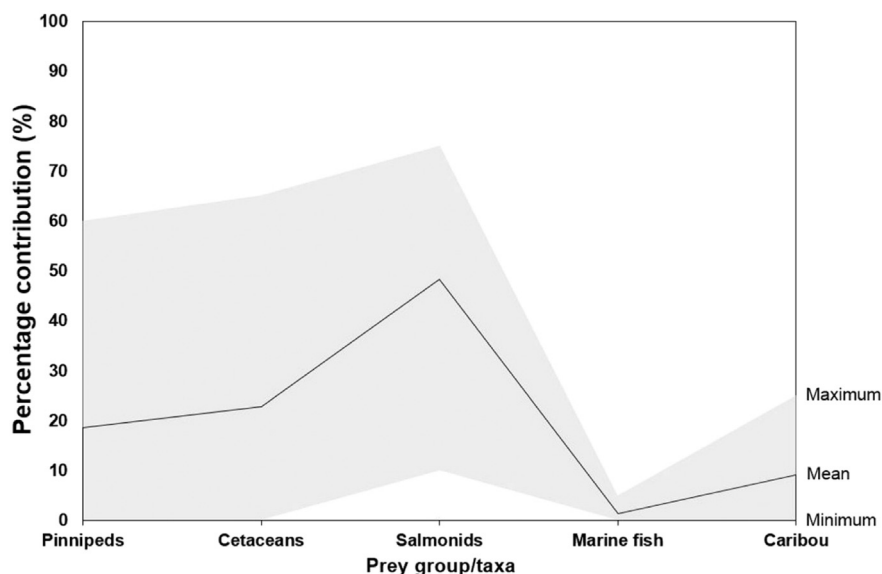

Fig. 3. Summary IsoSource mass balance data for Nunalleq, showing minimum, maximum and mean proportional contributions to the diet of major prey taxa groups.
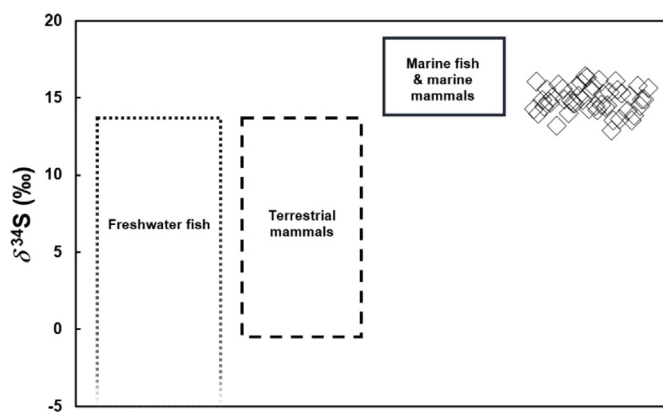
Fig. 4. Human sulphur isotope data from Nunalleq, plotted with general baseline approximations for freshwater, terrestrial and marine ecosystems (based on Nehlich and Richards, 2009; Nehlich, 2015). Measurement error was ±0.5‰ (1σ).

factors such as precise trophic relationships, seasonality of resources/resource density, and the full range of potential foods ("end members") are unlikely to be determinable (Britton et al., 2013: 456).

4.1.3. 'Bulk' sulphur isotope data

The sulphur isotope data from human hair from Nunalleq range from 12.9‰ to 16.4‰, with a mean value of 14.9 ± 0.8‰ ($n = 49$; Table 1). While the determination of sulphur isotope values for fauna from the site is still ongoing, $\delta^{34}S$ values are known to vary considerably between freshwater, terrestrial and marine ecosystems and previously-published data from bone collagen studies of organisms from different ecosystems (e.g. Nehlich and Richards, 2009; Nehlich, 2015) can be used as a guideline for interpreting the human hair values from Nunalleq. Although bone collagen and hair keratin differ in their amino acid composition, and the concentration of sulphur in hair keratin is far higher than bone collagen (Eastoe, 1955; Valkovic, 1977), no *consistent* off-sets or physiological discrepancies might be expected by comparing bone collagen and hair $\delta^{34}S$ values in animals, and there is little diet-tissue fractionation offset (Nehlich, 2015), permitting these broad comparisons to be made. Sulphur isotope ratios measured in bone collagen from modern marine mammals range from +14‰ to +19‰, which are similar to marine fish (Nehlich and Richards, 2009). Such values, if observed in humans, can therefore be consistent with a

major marine contribution to the diet (Nehlich, 2015). It should be noted, however, that 'baseline' terrestrial values in coastal regions can also be high (>14‰) due to the sea-spray effect (Richards et al., 2001a; Nehlich, 2015). Freshwater animals display a wide range of values, from −20‰ to +14‰, with terrestrial mammals exhibiting values between approximately 0‰, and 10‰, (Nehlich and Richards, 2009). When compared to these general ecosystem sulphur baselines (Fig. 4), the human hair sulphur isotope data from Nunalleq are in agreement with the carbon and nitrogen isotope data (see Sections 4.1.1 and 4.1.2) supporting the hypothesis that protein in the diet at Nunalleq was predominantly marine-based, although with some possible freshwater and/or terrestrial inputs.

These sulphur data provide further evidence for subsistence strategies at Nunalleq, suggesting that protein in the diet was dominated by marine/anadromous sources, but likely incorporating other sources. This is consistent with the carbon and nitrogen isotope data presented here, and in the initial pilot study (Britton et al., 2013). It should be noted, however, that there is also a known sea-spray influence in environmental sulphur, which can lead to enriched values even in purely terrestrial ecosystems along coastlines (see review in Nehlich, 2015). The ongoing sulphur isotope analyses of the faunal assemblage from Nunalleq will be helpful in better understanding sulphur isotope systematics in this region, and the possible influence of marine sulphur in terrestrial ecosystems and the influence of freshwater in the marine foodwebs in the Kuskokwim Bay, and ultimately in the interpretation of sulphur isotope data from human hair at the site. In light of the evidence from other isotopes presented here and the faunal assemblage at the site, even if a sea-spray influence was demonstrated it would be unlikely to alter the dietary interpretation significantly. It is also important to highlight that there are currently no accepted criteria for establishing the preservation of human hair for sulphur isotope analysis. Sulphur content of mammalian keratinous tissues is variable, although previously measured values from human hair (and the amino acid composition of human hair) indicate the sulphur content of hair should be no more than 5% (Nehlich, 2015). As shown in Table 1, around two-thirds of the dataset generated from Nunalleq have sulphur contents >5%, and two of the samples have compositions exceeding 6%. This may suggest the addition of sulphur-containing contaminants from the burial matrix that have not been removed during the pretreatment of hair samples (which focused on the removal of lipids), or possible degradation of the samples (i.e. loss of integral organic material, but preservation of the sulphur bonds, leading to its slight over-
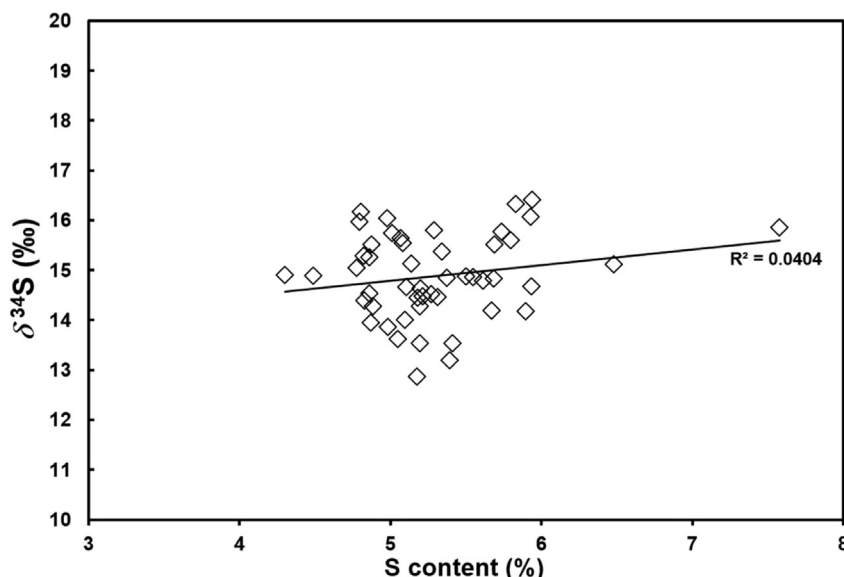

Fig. 5. Sulphur isotope ratios and sulphur content of human hair from the Nunalleq site. Measurement error was ±0.5‰ (1σ).
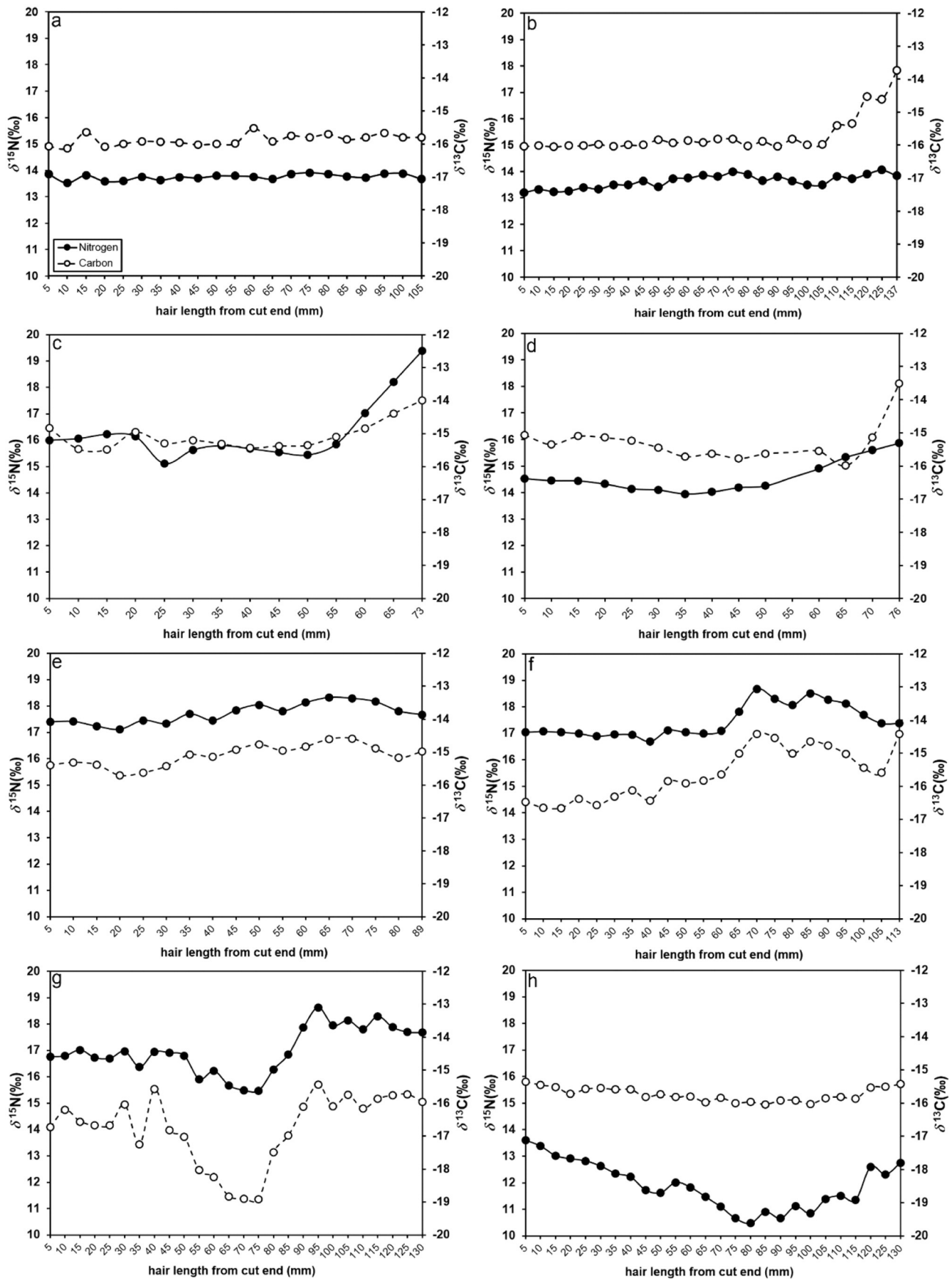
Fig. 6. a–h: Carbon and nitrogen isotope values of sequentially-sampled human hair from the Nunalleq site (a = 17790.1–17790.21; b = 21516.1–21516.26; c = 21518.1–21518.14; d = 21517.1–21517.15; e = 21519.1–21519.17; f = 21521.1–21521.22; g = 21522.1–21522.26; h = 21523.1–21523.26). Measurement error was 0.2‰ (1σ) or better. Please note, hair length is distance (mm) from the cut end. The cut end represents a more recent period in the individual's life than the distal end.
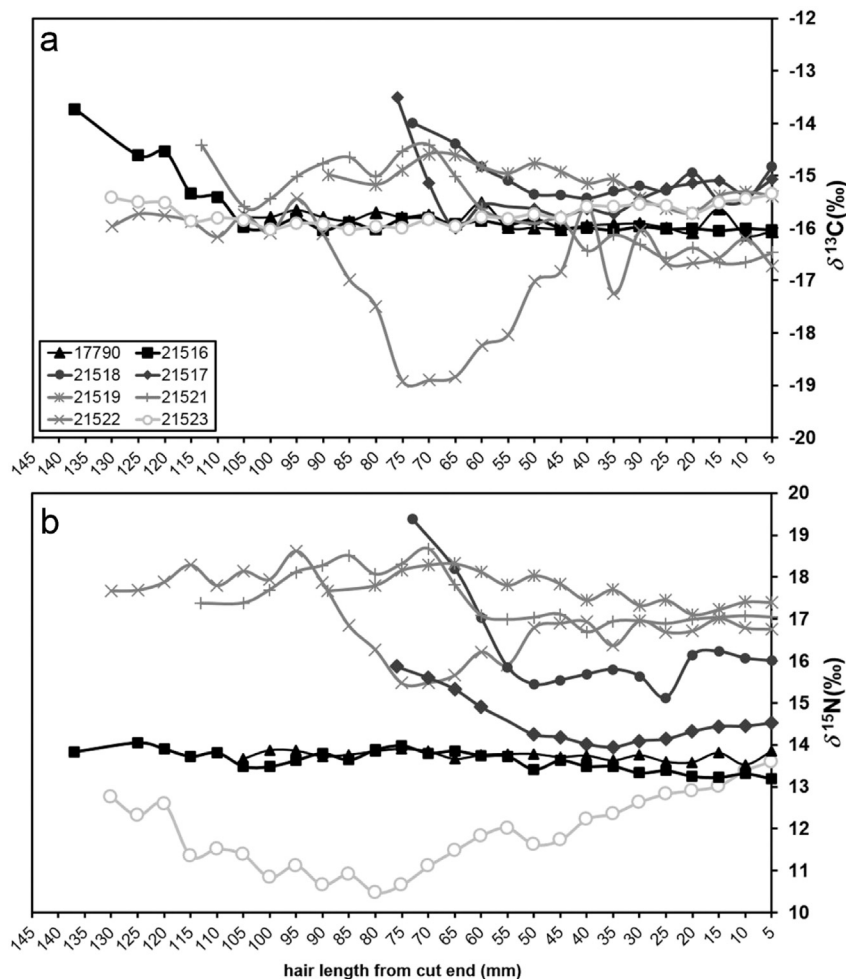
Fig. 7. a–b: Carbon (a) and nitrogen (b) isotope values of sequentially-sampled human hair from the Nunalleq site. Measurement error was 0.2‰ (1σ) or better.

representation). As shown in Fig. 5, samples with the highest sulphur contents have $\delta^{34}S$ values that fall within the range of $\delta^{34}S$ values from samples with contents ≤5% and there is no strong correlation between $\delta^{34}S$ value and sulphur content (%).

It should also be noted that the atomic C:S and N:S values (Table 1) are largely within the range expected for human hair (based on carbon, nitrogen and sulphur content), and ratios are similar to those that can be calculated from previously-published data from archaeological
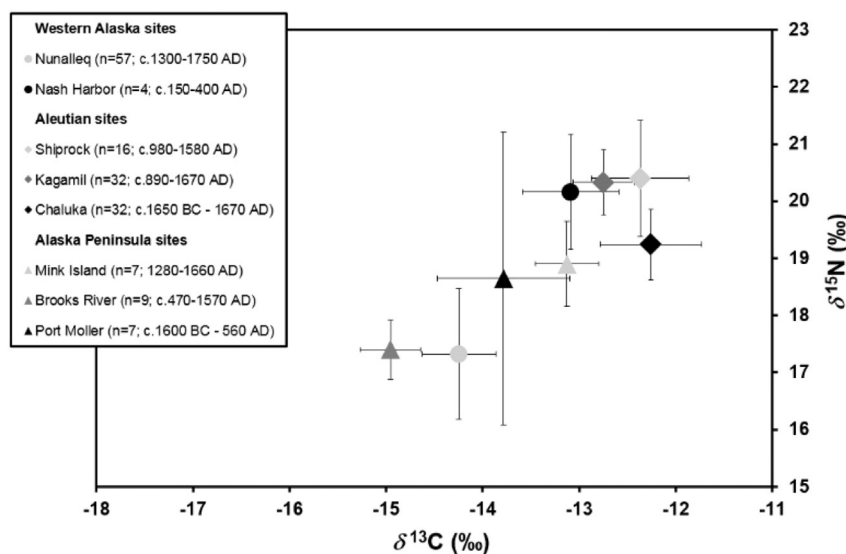
Fig. 8. Mean stable carbon and nitrogen isotope data (±1σ) of bone collagen from a range of Aleutian and Alaskan sites/assemblages from previous publications (Coltrain, 2010a, b; Byers et al., 2011). Values from human hair from Nash Harbor (Britton et al., 2013) and Nunalleq (Britton et al., 2013, and this study) have been adjusted to a predicted mean bone collagen value (based on O'Connell et al., 2001) for comparative purposes.
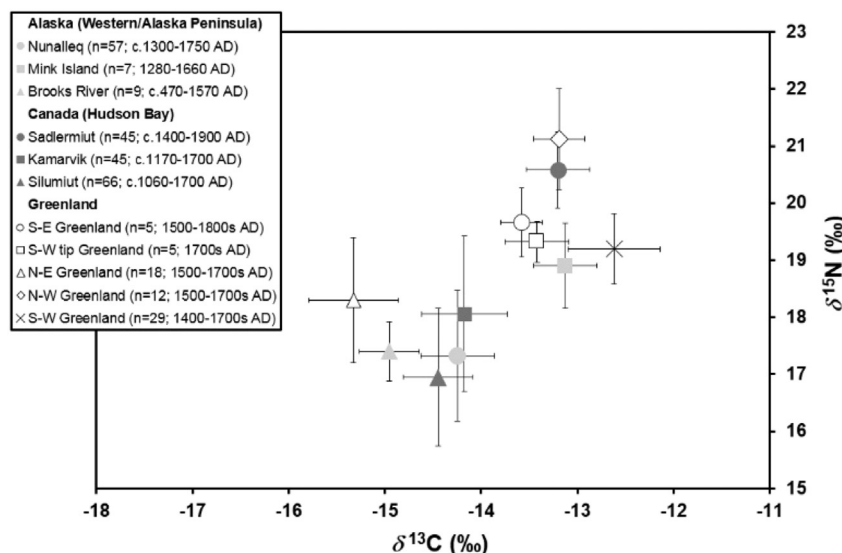
Fig. 9. Mean stable carbon and nitrogen isotope data ($\pm 1\sigma$) of bone collagen from a range of Thule-era Alaskan, Canadian and Greenlandic sites/assemblages from previous publications (Coltrain et al., 2004; Coltrain, 2009, 2010a; Nelson et al., 2012). Values from human hair from Nunalleq (Britton et al., 2013, and this study) have been adjusted to a predicted mean bone collagen value (based on O'Connell et al., 2001) for comparative purposes.

human hair from Egypt (Touzeau et al., 2014: 118; atomic C:S range from 25.9–31.2 and atomic N:S range from 6.9–8.1, when corrected for atomic mass). Based on these data, and the ones presented in this study, a guideline of C:S 30 ± 15 and N:S 10 ± 5 can perhaps be tentatively estimated for human hair, however carbon, nitrogen and sulphur compositional data on modern human hair is required to validate this. Although large bodies of $\delta^{13}C$, $\delta^{15}N$ and $\delta^{34}S$ data from modern hair have been published (e.g. Thompson et al., 2010; Valenzuela et al., 2011), accompanying content values are unfortunately rarely reported.

Beyond sample integrity, physiological and metabolic influences may also play a role in the sulphur content of hair, for example, during dietary stress human hair follicles synthesise protein low in cysteine which could lead to an overall decrease in sulphur (Valkovic, 1977: 24). While the sulphur isotope data presented here are in agreement with the carbon and nitrogen isotope data (in terms of dietary reconstruction), until quality assessment criteria for hair sulphur data (based on a large modern dataset) are established, such data should be viewed with due caution. In addition to the determining of quality control indices, more work is also required on the influences of diet and location (and proximity to the coast) on sulphur in human hair, and the effect of *in vivo*, taphonomic and diagenetic processes.

4.2. Stable isotope data from sequentially-sampled strands

Carbon and nitrogen isotope analysis of a single longer lock of hair in the original pilot study at Nunalleq suggested no intra-annual dietary variation (Britton et al., 2013). This was perhaps surprising, in light of the highly seasonal nature of resource availability in the region. Sequential analysis of a single hair from one of the Qilakitsoq mummies from Greenland provides the only other comparable Thule-era data published, and these did reveal intra-hair variability in $\delta^{13}C$ (~3‰) and $\delta^{15}N$ (~1‰) values, evidencing seasonal differences (Wilson, 2005: 326, Fig. 14.3). Therefore, additional longer samples were incorporated into the current study in order to better understand and characterise (possible) seasonal dietary variability at Nunalleq. Furthermore, these data could help to explain the variability observed between some of the 'bulk' samples in Fig. 2. Seven additional longer locks (ranging from 75 to 145 mm) were selected for sequential isotope analysis, and data from these – alongside the single strand from the pilot study – are shown in Fig. 6a–h, and grouped together in Fig. 7a (carbon) and b (nitrogen). The total isotopic range of the eight sequentially-sampled

longer locks of hair from Nunalleq varies considerably, from −18.9‰ to −13.7‰ for $\delta^{13}C$, and 10.5‰ to 19.4‰ for $\delta^{15}N$.

Fig. 6a presents data from the first longer hair strand analysed as part of the previously-published pilot study (Britton et al., 2013). This sample (17790.1–17790.21; Square 1, Level 4) demonstrates very little intra-hair variation (only 0.6‰ and 0.4‰ in $\delta^{13}C$ and $\delta^{15}N$), reflecting an isotopically-homogenous diet over a period of 10–11 months. Similar values and a similar trend is observed in sample 21516.1–21516.26 (Fig. 6b; Square 8, Level 3), which demonstrates very little intra-hair variation in $\delta^{13}C$ or $\delta^{15}N$ for a period of 10-11 months (105 mm), until the final ~30 mm when the $\delta^{13}C$ rises from consistent values of ~16‰ to a peak of −13.7‰. These consistent $\delta^{15}N$ values, coupled with higher $\delta^{13}C$, perhaps indicate the inclusion of more (lower trophic level) marine protein in the diet during the period represented by that section of hair (~2 to 3 months, in the ontogenetically 'oldest' portion of the hair). The isotopic homogeneity demonstrated through much of both these longer hair strands come in contrast to the highly seasonal nature of resource availability in this high-latitude region, perhaps reflecting the storage of highly seasonal resources and the consumption of them throughout the year (Britton et al., 2013). These strands are also notable, along with strands 21517.1–21517.15 (Fig. 6d) and 21523.1–21523.26 (Fig. 6h), for having generally lower $\delta^{15}N$ values than the other four longer strands.

Samples 21518.1–21518.14 (Fig. 6c), demonstrate higher values than those shown in Fig. 6a and b but, like 21516.1–21516.26 (Fig. 6b), demonstrate relatively homogenous values throughout the hair section until the final ~20 mm, when both the $\delta^{13}C$ and $\delta^{15}N$ values rise. This co-variance, and higher overall values, are typical of the inclusion of a greater proportion of higher trophic level marine protein in the diet. A similar trend is observed in samples 21517.1–21517.15 (Fig. 6d), with co-variance and an increase in values in both isotope systems in approximately the last 20 mm of hair analysed (albeit with lower overall $\delta^{15}N$ values, suggesting a lower trophic level marine input to the diet). It should be noted however that both these strands are shorter than some of the others, 70–80 mm in length, representing approximately 7–8 months of hair growth, which may be insufficient to reveal seasonal variations in their entirety.

As shown in Fig. 6e, f, and g, samples 21519.1–21519.17, 21521.1–21521.22 and 21522.1–21522.26 also demonstrate intra-hair co-variation in $\delta^{13}C$ and $\delta^{15}N$. While this variation is very slight in 21519.1–21519.17 (Fig. 6e), with $\delta^{13}C$ values ranging from −15.7‰ to

— 14.6‰, and δ^{15}N values ranging from 17.1‰ to 18.3‰ throughout the entire length of the hair, trends in both isotope systems co-occur. These elevated values indicate the consumption of higher trophic level marine-based proteins. Co-variance is also observable in samples 21521.1–21521.22 (Fig. 6f), albeit with a greater range, as δ^{13}C values vary by ~2‰ throughout the growth period represented in this strand (approximately 1 year), and δ^{15}N values vary by around 2.5‰, potentially highlighting seasonal dietary variation including the consumption of higher trophic level marine protein. Substantial intra-hair variation is observed in samples 21522.1–21522.26 (Fig. 6g), again with δ^{13}C and δ^{15}N co-varying, although by >3‰ over the growth period of this long strand, demonstrating substantial variation. What is remarkable about 21522.1–21522.26 is that, while δ^{13}C and δ^{15}N do co-vary, this individual demonstrates some of the most elevated δ^{15}N values and some of the lowest (most 'terrestrial') δ^{13}C values. This substantial intra-hair variation provides the best evidence for seasonal (or supra-seasonal/inter-annual) dietary variation at Nunalleq, at least in this individual. The lowest δ^{15}N values observed (~15.5‰), correspond with low δ^{13}C values of ~— 19‰. This could suggest the consumption of high trophic level terrestrial foods, such as carnivores, or the consumption of freshwater fish or waterfowl. Seasonal exploitation of waterfowl and waterfowl eggs is ethnographically well-documented in the Yukon-Kuskokwim Delta, along with the consumption of freshwater fish species such as black fish and lamprey eels (Fitzhugh and Kaplan, 1982: 51). The carbon values of this individual are so different from any of the others determined at the site so far that this could suggest a distinctive life history, possibly reflecting a different mobility history of this individual, with a period of time spent inland. Interestingly, the sections of hair representing the most recent (5–40 mm) and oldest periods of growth (100–130 mm) are similar, with elevated δ^{13}C and (especially) δ^{15}N indicating the inclusion of high trophic level marine protein in the diet at these times. As well as seasonal dietary variability, this could be indicative of seasonal mobility/seasonal rounds and (possibly) seasonally-focused hunting activities.

Hair samples 21523.1–21523.26 (Fig. 6h) also demonstrate an unusual overall trend. While δ^{13}C values vary little throughout the period of growth of this hair (~6 months), δ^{15}N values vary substantially and are far lower than observed in other individuals, ranging from around 13.5‰ to as low as 10.5‰. While the consistent and relatively elevated δ^{13}C values appear to suggest a steady marine influence in the diet, the nitrogen values are far lower than those observed alongside similar δ^{13}C values in other individuals. This suggests the consumption of very low trophic level marine or estuarine foods, such as bivalves or other shellfish.

Compared alongside one another in Fig. 7a and b, it is clear that – aside from the case of sample 21522 – there is substantially more variation in δ^{15}N than in δ^{13}C values between the individuals at the site. The range of values observed span from 10.5‰ (sample 21523) to 19.4‰ (sample 21518). These data, while suggesting an ongoing marine influence in the diet (as reflected in the δ^{13}C values), highlight a variety of different trophic level resources consumed at different points by different individuals. While δ^{13}C and δ^{15}N values commonly co-vary, trends are not necessarily seasonally predictable – with some of the longest strands demonstrating very little intra-hair variability in either isotope system (e.g. sample 21516), and some evidencing substantial variation in one or both isotopes (e.g. samples 21522, and 21523). Instead, these data may highlight substantial inter-annual or inter-personal dietary differences, or – perhaps in some cases – differences in the life history and mobility of individuals. Involvement in specialised and seasonal hunting activities, for example of marine mammals, and/or the consumption of the products of these activities, could account for some of the very elevated δ^{15}N in some individuals (e.g. in individuals 21518, 21521 and 21522). In addition to dietary inputs, physiological factors could also play a role in inter-individual/inter-annual isotopic variability in growing hair. For example, δ^{15}N values have been shown to increase in growing human hair during periods of nutritional stress (Fuller et al.,

2005; Mekota et al., 2006). Carbon isotope values in hair have also been demonstrated to decrease substantially when the supply of protein in the diet is severely compromised (Mekota et al., 2006: 1609). However, given that carbon and nitrogen isotopes largely co-vary in the intra-hair data presented here, the patterns observed are unlikely to be accounted for by severe dietary stress. Instead, these are likely to reflect dietary differences, particularly in the proportion of high trophic level marine protein in the diet and the varying trends revealed in the sequentially-sampled hairs suggest this is not the product of straight-forward ('natural'/predictable) seasonal variability. Instead these differences either reflect supra-annual/temporal resource changes, or (perhaps more likely) reflect differences in individual dietary habits, access to resources, or behaviour (for example, mobility) during the period of hair growth.

The isotope data from sequentially-sampled hair presented in this study demonstrate the ability of this approach to reveal relatively short-term dietary histories of individuals and create a more nuanced picture of dietary variability than bone collagen, which provides an averaged record of diet over decades rather than months (e.g. Hedges et al., 2007). Conversely, this short-term record can serve as a limitation, revealing variation that is difficult to understand and contextualise without more detailed information concerning the individuals involved (for example, precise dates, biological sex or age). Some of this information may be discernible in future studies, but other aspects are impossible to ever account for (e.g. age, status): For example, if hair strands originated from infants and young children who were still being breastfed, intra-hair nitrogen and carbon variability would be attributable to changes in maternal diet (albeit further enriched in ^{15}N) and also be dependent on the relative contribution of breast milk and complementary foods to the pre-weaning diet, and also the time at which weaning was finally complete (e.g. Fuller et al., 2006). However, despite these inevitable unknowns, the analysis of the longer strands is certainly effective in better understanding some of the inter-personal variations noted in the 'bulk' studies (or for illuminating lifetime variations, where samples may originate from the same person), and appears to confirm the variability observed in the shorter strands is unlikely to reflect predictable seasonal variation in diet, but instead may be the result of other temporal or inter-personal differences in diet or even life history/mobility.

4.3. Comparison with other Alaskan, Canadian and Greenlandic sites

While intra-group variability can provide a more nuanced picture of individual dietary histories, as with most bone collagen studies, the cross-comparison of trends with other comparable datasets is of clear interest to archaeologists to better contextualise new data and to gain insight into broader trends. Stable isotope data, largely from bone collagen, have been previously-published from a number of Aleutian, Alaskan, Canadian and Greenlandic sites. These include datasets with multiple individuals from earlier sites (paleo-Aleut/-Eskimo or pre-Thule/-Inuit) as well as Thule-era assemblages (e.g. Coltrain et al., 2004; Coltrain, 2009, 2010a, b; Byers et al., 2011; Nelson et al., 2012; Britton et al., 2013), and reported carbon and nitrogen isotope values from single individuals (e.g. Qilakitsoq Mummy 3, Wilson, 2005; Saqqaq man, Rasmussen et al., 2010; the Umingmak human mandible (including sulphur), Bocherens et al., 2016).

In order to compare the data from Nunalleq to other previously-published datasets incorporating multiple individuals (which are largely based on bone collagen), it is necessary to use the adjusted predicted bone collagen values (see above). In Fig. 8, the mean predicted bone collagen value from Nunalleq is compared to archaeological stable isotope data from a range of other Alaskan prehistoric and precontact sites/components of sites of various cultural attributions/date, including Norton, Paleo- and Neo-Aleut, Thule-era, and earlier sites (data taken from: Coltrain, 2010a, b; Byers et al., 2011; Britton et al., 2013). Samples from Norton components of the Western Alaska site of Nash Harbor, Nunivak Island, and the Paleo- and (predominantly) Neo-Aleut sites of Kagamil

and Shiprock, demonstrate similar, elevated $\delta^{15}N$ and $\delta^{13}C$ values and thus a focus on higher trophic level marine foods, such as pinnipeds and cetaceans (Byers et al., 2011; Britton et al., 2013). While values from the (predominantly) Paleo-Aleut burials at Chaluka midden are also elevated, data may suggest differences in the trophic level and foraging location of resources targeted (near-shore vs off-shore, Byers et al., 2011). The oldest, and most westerly of the Alaska Peninsula sites, Port Moller (3547–1388 cal BP [2σ]), also demonstrates relatively high values (Coltrain, 2010a) as do burials from the Alaska Peninsula site of Mink Island, which dates to a similar period to the Nunalleq site (666–292 cal BP [2σ]). Although high trophic level marine prey clearly make a significant contribution at both of these sites, the data are not as isotopically-enriched as at the Aleutian or Nunivak Island sites, likely reflecting the inclusion of salmon, lower trophic level marine invertebrates or caribou (Coltrain, 2010a). In contrast, stable isotope analyses of bone collagen from Thule-era burials from Alaska Peninsula site of Brooks River (1484–381 cal BP [2σ]) imply a more mixed economy, with significant reliance on caribou and salmon, similar to Nunalleq. While there is no clear temporal pattern, the most elevated values are found at the more westerly Aleutian and island sites, suggesting that prehistoric subsistence strategies in the Western Arctic were tuned to local ecological conditions, albeit with influences from other climatic, cultural, and social factors (Coltrain, 2010b). Despite being geographically distant, the Thule-era sites (Nunalleq, and Brooks River) display similar, more mixed (and less marine mammal focused) dietary inputs, perhaps attesting to a generalist Thule subsistence strategy and reflecting their locations close to major rivers/tributaries and salmon runs. The burials from Mink Island (which are near-contemporary to the occupation at Nunalleq), are significantly enriched compared to data from both Nunalleq and Brooks River, suggesting this more mixed subsistence strategy (dominated by marine sources and salmonid protein, but also incorporating terrestrial resources), however, was not universal.

A similar pattern of dietary diversity emerges when comparing Thule-era datasets from Alaska (Alaska Peninsula/Western Alaska: Coltrain, 2010a; Britton et al., 2013 and this study) to those from Canada (Hudson Bay: Coltrain et al., 2004; Coltrain, 2009) and Greenland (Nelson et al., 2012) (Fig. 9). Data from the Hudson Bay Thule-era Silimiut and Kamarvik sites demonstrate mixed economies, dominated by seal and bowhead whale, but also including caribou (Coltrain et al., 2004; Coltrain, 2009). Analysis of the later Native Point Sadlermiut burials (also Hudson Bay) demonstrated a far more intensely marine-focused economy, with a heavy reliance on ringed seal and piscivorous sea birds (Coltrain et al., 2004; Coltrain, 2009). Thule Greenlandic sites also demonstrate considerable variability. Bone collagen analysed from human burials at north-eastern Greenlandic sites, such as Suess Land and Dødemandsbugten, demonstrate lower carbon and nitrogen isotope values and therefore suggest much more generalist/mixed economies (or economies based on lower trophic level marine foods), contrasting with sites such as those to the south and north-west which indicate dietary protein was almost exclusively sourced from high trophic level marine mammals (Nelson et al., 2012). While there are broad geographical trends (in that the Greenlandic sites tend to be more marine-mammal focused than the Canadian and Alaskan sites), there are marine-mammal dominated and more generalist subsistence strategies evidenced at sites in all three of the broad regions over a period of only a few centuries. This diversity of procurement strategies mirrors what has been evidenced by zooarchaeological data in some regions (e.g. in the Mackenzie Delta in the Western Canadian Arctic, see Betts, 2008).

The variability in Thule-era diet is remarkable, and perhaps attests to the diversity and flexibility of Thule-era cultures, and the precontact neo-Eskimo and neo-Inuit world. While there is no consistent trend with latitude or longitude (or with time), there would undoubtedly be geographical (thus environmental) bases for some of the observed variability (e.g. proximity to major salmon runs or bird nesting sites; and

extent of sea ice coverage). Furthermore, there were pronounced climate changes in the last millennia, as the Neoglacial expansion has been marked by alternating periods of cooling and warming, including the Medieval Warm Period (~900–1350 CE) and the Little Ice Age (~1350 to 1900 CE) (Mann et al., 1998). These periods would have been overlain by more local annual and seasonal variations, which are very difficult to reconstruct and may have influenced species availability considerably. These climatic variations (and climatically-related geographical factors), could account for the some of the intra- and inter-site variations observed. Indeed, the influence of prey species biogeography (rather than cultural identity grouping) has been demonstrated to be the primary influencing factor in the types of fauna harvested for subsistence hunting and fishing in a recent study of contemporary Alaskan populations engaging in traditional subsistence activities (Renner and Huntington, 2014). Changing contemporary climates are known to influence prey species abundance and biogeography (Callaway et al., 1999; ACIA, 2004; Hinzman et al., 2005; Moore and Huntington, 2008; Joly et al., 2011), and the influence of climate change and its impact on the distribution of subsistence resources is often hypothesized to be the primary cause of change or variability in hunter-gatherer subsistence systems. For example, it has been speculated that changes in sea ice coverage during the Little Ice Age in the central Canadian Arctic, leading to changes in the distribution of bowhead whales, influenced Thule hunting patterns from the 1400s CE onwards (see discussion in Friesen, 2013: 37). Changes are likely to have occurred on decadal timescales, as well as longer periods, and climatic shifts may have necessitated a flexibility in subsistence strategies over relatively short periods, facilitated by a broader generalist technological and cultural toolset. The spread and prevalence of Thule cultures across the Arctic, and the success of descendant Eskimo and Inuit cultures, may attest to the flexibility afforded by such adaptive subsistence approaches, along with efficient transportation systems (dog sleds, umiaks and kayaks) and domestic structures. To better understand the relationship between climate change, ecosystem change and changes in human subsistence strategies in the Arctic over the last two millennia, multi-disciplinary studies on archaeological sites are required – incorporating localised palaeoclimate reconstruction, ecological modelling, zooarchaeology, material techno-culture analysis and palaeodietary studies.

5. Conclusion

The 'bulk' carbon, nitrogen and sulphur isotope data from human hair from Nunalleq infer a mixed economy at this coastal site, including marine mammals and terrestrial protein, but likely dominated by lower trophic level marine foods or estuarine/freshwater protein sources (particularly salmonids). The Kuskokwim and Yukon rivers, and other tributaries, host significant runs of anadromous fish today and historically (Fitzhugh and Kaplan, 1982; Barker and Barker, 1993). A mixed but marine-/anadromous-protein-dominated economy is consistent with historical and ethnographic accounts of Yup'ik diet in the Yukon-Kuskokwim Delta, with caribou hunting and salmon fishing in particular remaining important cultural and economic activities in the region today (Fitzhugh and Kaplan, 1982; Barker and Barker, 1993; Callaway et al., 1999). The positioning of the Nunalleq site itself likely reflects the mixed and flexible subsistence strategies of its early inhabitants: on the coast, but close to significant river tributaries and salmon runs, and with access to inland resources such as caribou. The intra-hair data presented here brings additional insights into the nature of the subsistence economy at Nunalleq. Some individuals demonstrate little or no dietary variability, and indicate the consumption of a mixed diet all year round (possibly evidencing a reliance on stored foods). Other individuals demonstrate considerable intra-hair variability, suggesting a diet incorporating proportionally more high-trophic level marine protein (such as cetaceans and pinnipeds), protein from anadromous species (such as salmon), or terrestrial protein at different times. Whether this reflects the dietary differences of individuals living

at the site/social access to food resources, or suggests differences in seasonal mobility/activity patterns (seasonal rounds) can only be speculated. Furthermore, in light of the broad chronological grouping of the individuals and likely climatic variability at the time, supra-annual/temporal resource availability differences are difficult to take into account.

Data from Nunalleq and a range of Thule-era sites in Alaska, Canada and Greenland, highlight substantial variability in Thule subsistence strategies – from the more mixed economies evidenced by studies at Nunalleq (Alaska), the north-east Greenland sites, and (to an extent) Silumiut and Kamarvik (Canada), to the marine-mammal dominated subsistence strategies at Mink Island (Alaska Peninsula) and many of the Greenlandic sites. The cause of this variability is likely complex, incorporating climatic, geographical, and cultural parameters, but may attest to resource flexibility during the Thule expansion and in response to the climatic shifts associated with the Medieval Warm Period and the beginning of the Little Ice Age. It is hoped that the analysis of further non-mortuary human hair samples from Nunalleq, incorporating the greater temporal constraints permitted by recent research excavations and site-specific palaeoclimatic proxy research (e.g. pollen, beetles), may serve as a test case to explore the impact of this late Holocene climatic variability on resource availability and diet at a single location. In combination with other lines of evidence, such as genetic sexing of hair samples or the comparison of hair samples from different structures at the site, it is also hoped that future analyses will also further illuminate the socio-cultural, as well as ecological, aspects of subsistence in the precontact Yukon-Kuskokwim Delta.

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