

2016

# Taxonomy, location of origin, and health status of proboscideans from Western Canada investigated using stable isotope analysis

Jessica Z. Metcalfe

*The University of Western Ontario*

Fred Longstaffe

*The University of Western Ontario, flongsta@uwo.ca*

Christopher N. Jass

*Royal Alberta Museum*

Grant D. Zazula

*Government of Yukon*

Grant Keddie

*Royal British Columbia Museum*

Follow this and additional works at: <https://ir.lib.uwo.ca/earthpub>



Part of the [Geochemistry Commons](#), and the [Paleontology Commons](#)

---

## Citation of this paper:

Metcalfe, Jessica Z.; Longstaffe, Fred; Jass, Christopher N.; Zazula, Grant D.; and Keddie, Grant, "Taxonomy, location of origin, and health status of proboscideans from Western Canada investigated using stable isotope analysis" (2016). *Earth Sciences Publications*. 8. <https://ir.lib.uwo.ca/earthpub/8>

1  
2  
3  
4  
5  
6 **Taxonomy, location of origin, and health status of proboscideans from Western Canada**  
7  
8 **investigated using stable isotope analysis**  
9  
10

11  
12  
13  
14  
15 **Running head: Stable isotope analyses of Western Canada proboscideans**  
16  
17

18  
19  
20 Jessica Z. Metcalfe <sup>a\*</sup>, Fred J. Longstaffe <sup>a</sup>, Christopher N. Jass <sup>b</sup>, Grant D. Zazula <sup>c</sup>, Grant  
21  
22 Keddie <sup>d</sup>  
23  
24  
25

26  
27 <sup>a</sup> Department of Earth Sciences, The University of Western Ontario, London, Ontario, Canada  
28  
29 N6A 5B7  
30

31  
32 <sup>b</sup> Quaternary Palaeontology Program, Royal Alberta Museum, 12845 – 102 Avenue, Edmonton,  
33  
34 Alberta, Canada T5N 0M6  
35

36  
37 <sup>c</sup> Yukon Palaeontology Program, Department of Tourism & Culture, Government of Yukon, PO  
38  
39 Box 2703, Whitehorse, Yukon, Canada Y1A 2C6  
40

41  
42 <sup>d</sup> Department of Archaeology, Royal British Columbia Museum, 675 Belleville St., Victoria,  
43  
44 British Columbia, Canada V8V 1X4  
45

46  
47  
48 \* Corresponding author. Present address: Department of Anthropology, The University of British  
49  
50 Columbia, 6303 NW Marine Dr., Vancouver, BC, Canada V6T 1Z1.  
51

52  
53 jess\_metcalfe@hotmail.com  
54  
55  
56  
57  
58  
59  
60

### Abstract

We investigated the application of stable isotope analysis of proboscidean remains (collagen in bone/dentin/cementum and structural carbonate in enamel bioapatite) for genus-level identification of isolated specimens, assessment of geographic origins, and testing for nutritional stress. Mammoths (*Mammuthus* sp.) tended to have higher  $\delta^{15}\text{N}_{\text{col}}$  and lower  $\delta^{13}\text{C}_{\text{col}}$  than mastodons (*Mammuth americanum*), but differences were not significant in every location. Determining the genus of isolated specimens may be possible for locations and time periods with good isotopic baselines, but environmental changes can confound interpretations. For example, an Alberta proboscidean with a  $\delta^{15}\text{N}_{\text{col}}$  of +1.4‰ (characteristic of mastodons) ultimately proved to be a mammoth. Its surprisingly low nitrogen isotope composition is attributable to the recently deglaciated environment it inhabited. We provided a baseline for isotopic assessment of geographic origins of isolated proboscideans in Western Canada. Bioapatite  $\delta^{13}\text{C}_{\text{sc}}$  and  $\delta^{18}\text{O}_{\text{sc}}$  can be used to distinguish specimens from Alberta, Klondike, Old Crow, Herschel Island, and further south (e.g., Arizona, Great Lakes). Finally, we found that an Alberta mammoth with morphological evidence of nutritional stress experienced a change in diet, environment, or physiology prior to death, but its isotopic compositions did not suggest a link to hypothesized starvation (catabolism of proteins or reliance on lipids).

### Keywords:

Isotopes; mammoth; mastodon; collagen; carbonate

## Introduction

Stable isotope compositions of fossil vertebrate remains can be used to reconstruct the behaviour (e.g., diet, migration), physiology (e.g., starvation) and paleoenvironmental context (e.g., vegetation, temperature, aridity, pCO<sub>2</sub>, and canopy cover) of extinct organisms (Clementz, 2012; Hatch, 2012; Koch, 1998; Koch et al., 1994; Kohn and Cerling, 2002; West et al., 2006). Stable isotope compositions are also commonly used to identify or verify the origins of various modern substances (e.g., Chesson et al., 2010a, b), and can also be useful for inferring the geographical origins of humans or animals in ancient landscapes (e.g., Dupras and Schwarcz, 2001; Knudson and Price, 2007; White et al., 1998).

In this paper, we explore the use of  $\delta^{13}\text{C}$  (collagen and bioapatite),  $\delta^{15}\text{N}$  (collagen), and  $\delta^{18}\text{O}$  (structural carbonate in bioapatite) of extinct proboscideans for genus-level identification of isolated remains, determination of location of origin, and investigation of health status, using specimens from Western Canada (Alberta, British Columbia, and Yukon). We ask: (1) Are there consistent differences in the isotopic compositions of mammoths and mastodons in each location? (2) Can skeletal fragments of proboscideans that lack morphologically diagnostic features be assigned to genus (*Mammuthus* vs. *Mammut*) based on isotopic compositions? (3) Are there consistent differences in isotopic compositions of proboscidean fossils across geographic locations (Alberta, British Columbia, and Yukon – including Old Crow, Klondike, and Herschel Island), which could allow location-of-origin determination for isolated specimens from museum collections? (4) Do the isotopic compositions of an abnormal mammoth tooth support a hypothesis of nutritional stress prior to death?

1  
2  
3 To date, studies that have used stable isotope compositions to investigate proboscidean  
4 paleobiology in western Canada have focused on food web and climatic reconstruction (Fox-  
5  
6 Dobbs et al., 2008; Matheus et al., 2003a, b; Schwartz-Narbonne et al., 2015; Szpak et al., 2010)  
7  
8 or nursing/weaning behaviour (Metcalf et al., 2010) within the Yukon Territory. At a  
9  
10 fundamental level, our intent is to provide baseline isotopic data for Pleistocene proboscideans  
11  
12 from western Canada. All subsequent paleobiological research questions require an  
13  
14 understanding of the variation in isotopic compositions within these taxa across time and space.  
15  
16  
17  
18  
19  
20  
21

## 22 **Mammoth and Mastodon Ecology**

23  
24 The Mammutidae and Elephantidae diverged at the end of the Oligocene or the beginning  
25  
26 of the Miocene, ca. 27-23 Ma (Agenbroad, 2005). Morphologically, the most obvious difference  
27  
28 between mastodons (*Mammut americanum*) and mammoths (*Mammuthus* spp.) is their dentition,  
29  
30 which is adapted to browsing in the former, and grazing in the latter. Mastodon remains are  
31  
32 typically associated with relatively wet, forested regions, such as that present in the Great Lakes  
33  
34 region during the terminal Pleistocene (Dreimanis, 1967, 1968; Haynes, 1991; McAndrews and  
35  
36 Jackson, 1988; Newsom and Mithlbackler, 2006; Saunders, 1996; Yansa and Adams, 2012).  
37  
38 Studies of mastodon tooth enamel isotopic compositions, microwear, and gut/fecal contents  
39  
40 support a browsing adaptation (Green et al., 2005; Koch et al., 1998; Lepper et al., 1991;  
41  
42 MacFadden and Cerling, 1996; Newsom and Mithlbackler, 2006), although mastodon diets did  
43  
44 include some grasses (Gobet and Bozarth, 2001; Koch et al., 1998) and mastodons were able to  
45  
46 adapt to significant temporal changes in vegetation within the Great Lakes region (Metcalf and  
47  
48 Longstaffe, 2014). Woolly mammoths (*Mammuthus primigenius*) were associated with open  
49  
50 steppe-tundra environments (Guthrie, 2001; Haynes, 1991) and their southern cousins, the  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 Columbian mammoths (*Mammuthus columbi*), were most common in temperate grasslands  
4  
5 (Agenbroad, 2005). Mammoths were predominantly grazers, as shown by their tooth enamel  
6  
7 isotopic compositions, gut contents, fecal remains, and phyloliths in dental calculus, though they  
8  
9 also consumed woody plants, mosses, and a variety of shrubs and forbs (Cummings and Albert,  
10  
11 2007; Feranec, 2004; Guthrie, 2001; Haynes, 1991; Hoppe, 2004; Koch et al., 1998; Mead et al.,  
12  
13 1986; van Geel et al., 2008). Both mammoths and mastodons were widespread in North America  
14  
15 during the late Pleistocene (~125 to 10 ka), occupying most of the regions south of the  
16  
17 continental glaciers, as well as Beringia during relatively warm and wet interglacials  
18  
19 (Agenbroad, 2005; Harington, 1990; Zazula et al., 2014). Despite their different dietary  
20  
21 adaptations and dentitions, isolated or fragmentary postcranial elements of mammoths and  
22  
23 mastodons are difficult to distinguish. An independent identification method for these genera  
24  
25 would be useful and would permit re-evaluation of specimens previously restricted to higher-  
26  
27 level taxonomic identification.  
28  
29  
30  
31  
32  
33  
34  
35

### 36 **Taxonomic Identification using Stable Isotopes**

37  
38 Previous research has shown that some animal taxa have distinct isotopic compositions  
39  
40 relative to others. If isotopic differences among taxa are relatively consistent at a given location  
41  
42 and/or during a given time period, they might be useful for taxonomic identification of skeletal  
43  
44 elements that cannot be identified on the basis of discrete morphological or morphometric data.  
45  
46 This approach was applied by Thewissen et al. (2001), who used carbon isotopes in bioapatite to  
47  
48 support identification of Eocene fossils as cetacean versus artiodactyl or anthracobunid, and by  
49  
50 Clementz et al. (2009), who used  $\delta^{13}\text{C}$  (collagen and bioapatite) to identify fragmentary  
51  
52 archaeological bone samples as sea cow, baleen whale, or sperm whale. While ancient DNA or  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 ZooMS (“collagen fingerprinting”) analyses would provide more definitive independent  
4  
5 identification methods (e.g., Buckley et al., 2014; Buckley and Kansa, 2011; Debruyne et al.,  
6  
7 2008; Enk et al., 2011; Poinar et al., 1998), isotopic analysis could potentially provide an easier  
8  
9 and less expensive methodology that can also be used to address additional paleobiological  
10  
11 questions.  
12  
13

14  
15 In regions with both C<sub>3</sub> and C<sub>4</sub> plants, which have very distinct carbon isotope  
16  
17 compositions,  $\delta^{13}\text{C}$  can be used to distinguish between browsers (such as mastodons) and grazers  
18  
19 (such as mammoths) (e.g., Baumann and Crowley, 2015; Koch et al., 1998). Since cold northern  
20  
21 regions lack C<sub>4</sub> plants,  $\delta^{13}\text{C}$  is less useful for identification in these locations, although small  
22  
23 differences among herbivore taxa can result from differing plant (part) selection, climate or  
24  
25 (micro)habitat, and animal physiology (e.g., Bocherens, 2003). In general, mammoths tend to  
26  
27 have higher  $\delta^{15}\text{N}$  (and sometimes lower  $\delta^{13}\text{C}_{\text{col}}$ ) than coexisting herbivores (Bocherens, 2003;  
28  
29 Bocherens et al., 1994; Fox-Dobbs et al., 2008; Iacumin et al., 2000; Koch, 1991; Kuitens et al.,  
30  
31 2012; Metcalfe et al., 2013). Schwartz-Narbonne et al. (2015) demonstrated that the high  $\delta^{15}\text{N}$  of  
32  
33 the woolly mammoth resulted from a distinct habitat or diet rather than physiology. Mammoths  
34  
35 and mastodons may also have distinct isotopic compositions if they drank from different water  
36  
37 sources, as shown for southwestern Ontario and western New York (Metcalfe et al., 2013). These  
38  
39 results suggest that an isotopic approach has potential for distinguishing between mammoths and  
40  
41 mastodons in C<sub>3</sub>-dominated environments.  
42  
43  
44  
45  
46  
47  
48  
49

## 50 **Location-of-Origin Determination using Stable Isotopes**

51  
52  
53 The use of stable isotope compositions to evaluate location of origin also has precedents  
54  
55 in the literature. Stable C, N, O, and H isotope analyses have been extensively utilized to  
56  
57  
58  
59  
60

1  
2  
3 determine the location of origin of modern commercial animal products, including milk  
4  
5 (Chesson et al., 2010a; Rossmann et al., 1998), cheese (Camin et al., 2012), honey (Chesson et  
6  
7 al., 2011), and wool (Hedges et al., 2005). Isotopic compositions have been used to identify the  
8  
9 location of origin and life history of human remains (e.g., Corr et al., 2008; Dupras and  
10  
11 Schwarcz, 2001; Knudson and Price, 2007; Sharp et al., 2003; White et al., 1998; White et al.,  
12  
13 2004), and are increasingly also used to aid in the identification and characterization of forensic  
14  
15 remains (Cerling et al., 2003; Ehleringer et al., 2008; Fraser and Meier-Augenstein, 2007; Mays  
16  
17 et al., 2011; Meier-Augenstein et al., 2013; O'Brien and Wooller, 2007).

22 Stable isotope methods for identifying location of origin of animal products and human  
23  
24 remains are based on differences in diet and physiology (for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ) (DeNiro and Epstein,  
25  
26 1978, 1981; Koch et al., 1994), and geographical variations in drinking water (for  $\delta^{18}\text{O}$  and  $\delta^2\text{H}$ )  
27  
28 (Bowen et al., 2005; Bowen and Wilkinson, 2002; Dansgaard, 1964; Ehleringer et al., 2008;  
29  
30 Rozanski et al., 1993). In  $\text{C}_3$ -dominated environments such as western Canada, regional  
31  
32 differences in climate (e.g., temperature, rainfall) and environment (e.g., canopy cover, soil  
33  
34 salinity, water availability, nutrient content) can cause small variations in the  $\delta^{13}\text{C}$  of  $\text{C}_3$  plants,  
35  
36 which are passed on to consumers (Arens et al., 2000; Drucker et al., 2008; Kohn, 2010; Murphy  
37  
38 and Bowman, 2009; Tieszen, 1991; Van Der Merwe and Medina, 1991). Herbivores occupying  
39  
40 different ecological niches may also select plants or plant parts with different  $\delta^{13}\text{C}$  values (e.g.,  
41  
42 Brooks et al., 1997; Tieszen, 1991), leading to different tissue  $\delta^{13}\text{C}$ . Temporal variations in the  
43  
44 proportion and isotopic composition of atmospheric  $\text{CO}_2$ , or in any of the factors described  
45  
46 above, can also affect plant and consumer  $\delta^{13}\text{C}$  (Arens et al., 2000; Long et al., 2005; Schmitt et  
47  
48 al., 2012). In the present study, we expect that differences in  $\delta^{13}\text{C}$  among regions will be small  
49  
50 (e.g., around 1-3‰), with higher  $\delta^{13}\text{C}$  in more arid locations (Kohn, 2010; Stewart et al., 1995).  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60



1  
2  
3 The largest variations in nitrogen isotope compositions of animals result from trophic  
4 differences (higher  $\delta^{15}\text{N}$  at higher trophic levels), which also affect juveniles consuming their  
5 mothers' milk (e.g., Fuller et al., 2006; Jenkins et al., 2001; Metcalfe et al., 2010; Minagawa and  
6 Wada, 1984; Rountrey et al., 2007). Herbivores can also have different  $\delta^{15}\text{N}$  values because of  
7 environmental conditions (e.g., low rainfall amounts and high temperatures are related to high  
8  $\delta^{15}\text{N}$  in plants), plant selectivity (e.g., plants with mycorrhizal associations tend to have lower  
9  $\delta^{15}\text{N}$ ), or physiology (e.g., nutritional stress can increase animal  $\delta^{15}\text{N}$ ) (e.g., Amundson et al.,  
10 2003; Craine et al., 2009; Drucker et al., 2010; Hartman, 2011; Szpak, 2014; Szpak et al., 2010).  
11 If regional differences are observed in the present study, we expect that samples from wetter  
12 locations and/or those with less-developed soils (e.g., recently deglaciated landscapes) will have  
13 lower  $\delta^{15}\text{N}$  values. The magnitude of differences in  $\delta^{15}\text{N}$  among regions in the present study is  
14 likely to be around 7‰ or less, based on previous isotopic studies of large mammals in  
15 glacial/postglacial Europe (Hedges et al., 2004; Richards and Hedges, 2003; Stevens and  
16 Hedges, 2004).

17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
Oxygen isotope compositions of meteoric water decrease with increasing latitude,  
altitude, and distance from the sea, as a result of variations in temperature, rainfall amount, and  
the movement of air masses (Dansgaard, 1964; Rozanski et al., 1993; West et al., 2006). For  
example, modern mean annual meteoric water  $\delta^{18}\text{O}$  values are approximately 3.5‰ lower in the  
Klondike region (Mayo, Yukon) than in Edmonton (Alberta) (IAEA/WMO, 2010). Geographic  
variations in modern meteoric water  $\delta^{18}\text{O}$  are well-documented in global isoscape maps (Bowen,  
2010; Bowen et al., 2005; Bowen and Wilkinson, 2002; West et al., 2006). Drinking water  $\delta^{18}\text{O}$   
values are passed on to large-bodied obligate drinkers such as proboscideans, but interpretation  
of ancient animal  $\delta^{18}\text{O}$  can be complicated by climate change, mobility, and non-meteoric water

1  
2  
3 sources (e.g., ancient groundwater or glacial meltwater) (Ayliffe et al., 1992; Bryant and  
4  
5  
6 Froelich, 1995; Kohn, 1996; Kohn et al., 1996; Levin et al., 2006; Longinelli, 1984). Thus, if  
7  
8 isotopic analyses are used to infer location of origin, it is important to establish local baselines  
9  
10 for the ancient ecosystems and taxa of interest. Generally, we expect that samples from higher-  
11  
12 latitude locations like Klondike and Old Crow will have lower  $\delta^{18}\text{O}$  values than samples from  
13  
14 lower-latitude or coastal locations like Alberta and British Columbia.

15  
16  
17 Based on the variations described above, an isotopic approach could be used to  
18  
19 investigate the geographic origins of museum specimens. For example, Metcalfe et al. (2013)  
20  
21 established a baseline for mammoth and mastodon isotopic compositions in Ontario, and  
22  
23 examined three proboscidean specimens that lacked contextual records. A mastodon tooth they  
24  
25 examined had isotopic compositions consistent with an origin in southwestern Ontario, whereas  
26  
27 two mammoth teeth were extreme outliers. The authors determined that one mammoth likely  
28  
29 lived during an interglacial period or in a more southern location, and the other was likely from  
30  
31 Klondike, Yukon Territory. In some cases, a similar isotopic approach could also be used to  
32  
33 investigate migration: if depositional contexts are secure and environmental change or  
34  
35 microhabitat differences are deemed unlikely (e.g., because the same taxa are analyzed), outlying  
36  
37 isotopic compositions can indicate migration from distant locations (Hobson and Koehler, 2015;  
38  
39 Hoppe, 2004; Keenleyside et al., 2011; Toyne et al., 2014; Webb et al., 2013).  
40  
41  
42  
43  
44  
45  
46  
47

#### 48 **Nutritional Stress: Isotopic Indicators**

49

50  
51 The isotopic effects of nutritional stress are generally difficult to distinguish from other  
52  
53 factors such as diet, climate, or geographic origins, and the relationship between food stress and  
54  
55 isotopic compositions is not always straightforward, particularly among different species (Hatch,  
56  
57  
58  
59  
60

1  
2  
3 2012; McCue and Pollock, 2008; Reitsema, 2013). Nevertheless, starvation, leading to  
4 catabolism of body tissues, commonly leads to an increase in  $\delta^{15}\text{N}$  of  $<+1$  to  $+2\%$  and either no  
5 change or a  $\leq 1\%$  increase in  $\delta^{13}\text{C}$  (Hatch, 2012; McCue and Pollock, 2008; Reitsema, 2013).  
6  
7 Thus, in special circumstances it may be possible to use isotopic compositions of skeletal  
8 remains to investigate health (e.g., Olsen et al., 2014). Fossil specimens that record  
9 morphological signals of poor health provide an ideal opportunity to evaluate the link between  
10 isotopic compositions and nutritional stress.  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21

## 22 **Materials and Methods**

### 23 **Samples**

24  
25  
26  
27  
28  
29 Samples of proboscidean skeletal remains (bone, tusk dentin, and molar enamel, dentin,  
30 and cementum) from known geographic localities were obtained from museum collections from  
31 British Columbia, Alberta, and Yukon (Table 1, Fig. 1). Only adult teeth and bones were  
32 selected, to avoid isotopic variations resulting from nursing (Metcalf et al., 2010; Rountrey et  
33 al., 2007). Most specimens were identified as mammoth or mastodon based on morphology, but  
34 a few were identifiable only as proboscidean. The latter were sampled as case studies for  
35 taxonomic identification. All samples were selected opportunistically, to avoid damage to  
36 museum specimens. Several grams of bone, tooth, and dentin were removed from each specimen  
37 using a Dremel tool and/or by manual breakage. Since relatively large amounts of “bulk” bone  
38 and dentin ( $\sim 0.5$  to  $1.5$  g) were used for collagen extraction, and because bone does not grow  
39 incrementally, collagen samples represent an average of many seasons of growth. There is some  
40 potential for seasonal bias in enamel samples, since enamel grows incrementally and smaller  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 samples are required for analysis. To minimize bias, we sampled pieces of enamel that  
4  
5 represented the entire enamel thickness (~1-2 mm) and typically ~10 mm or greater of tooth  
6  
7 height, homogenizing the samples for each individual. Growth rate estimates for both mammoth  
8  
9 and mastodon molar enamel indicate that more than 1 year is required for the full thickness of  
10  
11 the enamel to develop (Metcalf and Longstaffe, 2012, 2014). Growth in the occlusal-basal  
12  
13 direction of mammoth and modern elephant teeth occurs at a rate of 13 to 22 mm/a, with the  
14  
15 faster rates only occurring near the occlusal surface (Dirks et al., 2012; Metcalf and Longstaffe,  
16  
17 2012; Uno et al., 2013). Occlusal-basal growth rates for mastodon enamel are slower, with a  
18  
19 maximum rate of 12 mm/a near the occlusal surface and a minimum of 2 mm/a near the cervix  
20  
21 (Metcalf and Longstaffe, 2014). Thus, our enamel sampling method averaged seasonal  
22  
23 variations in both directions of tooth growth.  
24  
25  
26  
27  
28

29 One specimen, AB5 (RAM P90.7.1) allowed a unique investigation into the influence of  
30  
31 nutritional stress on the isotopic compositions of an extinct megaherbivore. This specimen is a  
32  
33 morphologically-normal upper-left M6 with a malformed, supernumerary M7 united to it by  
34  
35 hypercementosis (Fig. 2). According to Burns et al. (2003:83):  
36  
37

38 “The Villeneuve mammoth [AB5 / RAM P90.7.1] was clearly in great pain due to a  
39  
40 conspicuous bulging of the buccal alveolus, and it may be argued that it was the ultimate  
41  
42 cause of death.... Normal chewing action was eventually rendered impossible. The  
43  
44 secondary occlusal facet on the buccal edge of the molar suggests that the mammoth  
45  
46 made an effort to avoid the painful passing of the muscle across the bulge... and the  
47  
48 animal likely died of malnutrition shortly after cutting the new facet, at age 46 to 48  
49  
50 years, unable to process sufficient graze to survive.”  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 To examine temporal changes in the collagen  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of this unique specimen, six distinct  
4 subsamples were obtained: (1) dentin from the anterior M6, near the occlusal surface, (2)  
5  
6 cementum from the anterior M6, near the occlusal surface, (3) M6 root (mostly dentin), (4)  
7  
8 cementum joining the M6 and M7, (5) M7 crown cementum, and (6) M7 root (mostly dentin).  
9  
10  
11 These subsamples represent different formation times (Appendix S1), and could provide insight  
12  
13 into isotopic changes in the mammoth – due to nutritional stress or other causes – prior to death.  
14  
15  
16  
17  
18  
19

## 20 Collagen

21  
22 Bone, dentin (crown, root, tusk), and cementum were used for collagen analysis.  
23  
24 Collagen extraction was modified from Longin (1971), and included demineralization with 0.25-  
25  
26 0.5 M HCl (at room temperature), rinsing to neutrality, humic removal with 0.1 M NaOH (at  
27  
28 room temperature), rinsing to neutrality, and solubilization in  $10^{-3}$  M HCl at 75-90°C. Carbon  
29  
30 and nitrogen isotope ratios were obtained using a Costech elemental combustion system  
31  
32 (ECS4010) coupled to a Thermo-Finnigan Delta<sup>Plus</sup>XL stable isotope ratio mass spectrometer  
33  
34 (IRMS) operated in continuous-flow mode. The  $\delta^{13}\text{C}_{\text{col}}$  values were calibrated to VPDB using  
35  
36 ANU-Sucrose (IAEA-CH-6) and NBS-22 (accepted values =  $-10.50\text{‰}$  and  $-30.03\text{‰}$ ,  
37  
38 respectively), yielding the accepted value of  $+1.95\text{‰}$  for NBS-19 (Coplen, 1994; Coplen et al.,  
39  
40 2006). The  $\delta^{15}\text{N}_{\text{col}}$  values were calibrated relative to AIR using IAEA N-1 and IAEA N-2  
41  
42 (accepted values =  $+0.4$  and  $+20.3\text{‰}$ , respectively). Carbon and nitrogen contents were also  
43  
44 calibrated using the accepted values for the above standards. An internal keratin standard was  
45  
46 included in each analytical session to monitor accuracy and precision (mean  $\pm$  1 SD:  $\delta^{13}\text{C}$   
47  
48 measured =  $-24.03 \pm 0.06\text{‰}$  (n=77), accepted =  $-24.04\text{‰}$ ;  $\delta^{15}\text{N}_{\text{col}}$  measured =  $+6.29 \pm 0.10\text{‰}$   
49  
50 (n=77), accepted =  $+6.36\text{‰}$ ). The average difference between duplicate analyses of collagen  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 samples was 0.06‰ for  $\delta^{13}\text{C}$  and 0.11‰ for  $\delta^{15}\text{N}_{\text{col}}$  (n=16). For six individuals, collagen was  
4 extracted from multiple tissues (e.g., bone, dentin, cementum) from the same individual. The  
5 means of all measurements obtained for each individual are presented in Table 2. The differences  
6 between tissues from the same individuals ranged from 0.3 to 0.7‰ for  $\delta^{13}\text{C}_{\text{col}}$  and 0.6 to 1.5‰  
7 for  $\delta^{15}\text{N}_{\text{col}}$  (Table 3, Table S1).  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17

### 18 **Bioapatite**

19  
20 Only enamel was used for bioapatite analysis, because it is much more resistant to  
21 alteration than bone (Ayliffe et al., 1994). Pieces of enamel were mechanically cleaned of  
22 adhering dirt, dentin and cementum, sonicated in distilled water 2-3 times (minimum), and  
23 examined under an optical microscope prior to being ground to a powder using a mortar and  
24 pestle. Aliquots of enamel powder were treated with ~2.5% NaOCl at room temperature for ~21  
25 hours, rinsed five times with de-ionized water, reacted with 0.1M acetic acid for 4 hours, rinsed  
26 as above, and freeze-dried (Garvie-Lok et al., 2004; Koch et al., 1997). Carbon and oxygen  
27 isotope compositions were obtained using a MultiPrep automated sampling device (reaction with  
28 ortho-phosphoric acid at 90°C) coupled with a VG Optima IRMS in dual-inlet mode. The  $\delta^{13}\text{C}$   
29 values were calibrated relative to VPDB using NBS-19 (+1.95‰) and Suprapur (-35.28‰), and  
30 the  $\delta^{18}\text{O}$  values were calibrated relative to VSMOW using NBS-19 (+28.6‰) and NBS-18  
31 (+7.2‰). Accuracy and precision were assessed using standards not included in the calibration  
32 curve: NBS-18 ( $\delta^{13}\text{C}$  measured =  $-4.94 \pm 0.10$  ‰, n=16, accepted =  $-5.01$  ‰), internal laboratory  
33 standard WS-1 calcite ( $\delta^{13}\text{C}$  measured =  $+0.76 \pm 0.15$  ‰, n=5, accepted =  $+0.76$  ‰;  $\delta^{18}\text{O}$   
34 measured =  $+26.36 \pm 0.18$  ‰, n=5, accepted =  $+26.23$  ‰), and Suprapur ( $\delta^{18}\text{O}$  measured =  
35  $+13.15 \pm 0.12$  ‰, n=15, accepted =  $+13.25$  ‰). The average difference between duplicate  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 analyses of enamel samples was 0.07‰ for  $\delta^{13}\text{C}$  and 0.19‰ for  $\delta^{18}\text{O}$  (n=6). Carbonate contents  
4  
5 were determined using a calibration curve based on the reading of a pressure transducer in the  
6  
7 mass spectrometer and the known carbonate contents of calcite standards. Replicate  
8  
9 measurements of tooth enamel  $\text{CO}_3$  contents were within 0.4% on average (range of offsets = 0.1  
10  
11 to 0.9%).  
12  
13  
14  
15  
16  
17

## 18 **Statistics**

19  
20 For isotopic comparisons of mammoths and mastodons within a region (Alberta,  
21  
22 Klondike, or Old Crow), an F-test was used to determine whether variances of the two groups  
23  
24 were equal, followed by a t-test assuming either equal or unequal variance, as appropriate. For  
25  
26 isotopic comparisons among locations (Alberta, Klondike, Old Crow, Herschel Island, and  
27  
28 British Columbia), a Kruskal-Wallis non-parametric analysis of variance was used to determine  
29  
30 whether there were differences among groups, and a Dunn's multiple comparison test was used  
31  
32 to determine which groups differed. For comparisons among locations, British Columbia (BC)  
33  
34 and Herschel Island proboscideans specimens that were not identified to genus were compared  
35  
36 separately to mammoths and mastodons from other locations. If the unknowns had isotopic  
37  
38 compositions more similar to either mammoths or mastodons from the other locations, a case  
39  
40 could be made for their inclusion in that genus. An alpha of 0.05 was used for all statistical tests.  
41  
42  
43  
44  
45  
46  
47

## 48 **Results**

## 49 50 51 52 53 **Diagenesis** 54 55 56 57 58 59 60

1  
2  
3 The C/N ratios, carbon and nitrogen contents, and extraction yields indicate that the  
4 collagen samples were well-preserved (Table 2) (Ambrose, 1990; DeNiro, 1985; van Klinken,  
5 1999). Three samples [dentin from AB5 (P90.7.1), AB11 (P00.2.1), and AB6 (P94.4.3)] had C  
6 and N contents (ca. 25-30% and 10%, respectively) that were slightly lower than is usual for  
7 well-preserved collagen, but their C/N ratios were normal. Powder X-ray diffraction (pXRD)  
8 analysis of the collagen showed that inorganic salts (brushite and halite) were present in these  
9 (but not other) samples. Since brushite and halite contain no carbon or nitrogen, the low C and N  
10 contents were likely caused by the presence of these salts. We note that a variation in the  
11 collagen extraction procedure for these samples (i.e., demineralization of large intact chunks of  
12 dentin rather than small pieces) rather than poor preservation was likely responsible for the  
13 formation of the salts, since the salts only formed in samples extracted using the former method.  
14 Based on the results above, the  $\delta^{13}\text{C}_{\text{col}}$  and  $\delta^{15}\text{N}_{\text{col}}$  results obtained for these specimens are  
15 included in the discussion below.  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33

34 The carbonate contents of enamel samples ( $4.9 \pm 0.8\%$ , Table 2) are similar to those of  
35 mature enamel (2.7 to 5.0%) (Hillson, 1996). Fourier-transform infrared spectroscopy and pXRD  
36 analysis of selected pretreated samples indicated no secondary mineral contamination. Thin  
37 section analysis of one enamel specimen (AB6 / P94.4.3) revealed clearly visible incremental  
38 growth features (e.g., Striae of Retzius, cross-striations). All indicators suggest good preservation  
39 of enamel bioapatite.  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50

### 51 **Taxonomic Identification (Mammoth vs. Mastodon)**

52  
53  
54  
55  
56  
57  
58  
59  
60



1  
2  
3 The mean mammoth  $\delta^{15}\text{N}_{\text{col}}$  was higher than that of mastodons in all regions (Table 2,  
4 Fig. 3). The difference was significant for Klondike (mean  $\delta^{15}\text{N}_{\text{col}} = +7.5$  vs  $+4.2\text{‰}$ ;  $t = 7.8$ ,  $df =$   
5 24,  $p < 0.001$ ) and Old Crow (mean  $\delta^{15}\text{N}_{\text{col}} = +9.1$  vs  $+3.1\text{‰}$ ;  $t = 9.9$ ,  $df = 13$ ,  $p < 0.001$ ) but not for  
6 Alberta (mean  $\delta^{15}\text{N}_{\text{col}} = +7.6$  vs  $+4.7\text{‰}$ ;  $t = 1.7$ ,  $df = 5$ ,  $p = 0.15$ ). The mean mammoth  $\delta^{13}\text{C}_{\text{col}}$  was  
7 lower than that of mastodons in all regions, but the difference was only significant for Old Crow  
8 (mean  $\delta^{13}\text{C}_{\text{col}} = -21.4$  vs  $-20.1\text{‰}$ ;  $t = 6.6$ ,  $df = 13$ ,  $p < 0.001$ ). Neither  $\delta^{13}\text{C}_{\text{sc}}$  nor  $\delta^{18}\text{O}_{\text{sc}}$  differed  
9 for mammoths and mastodons (note: only Alberta and Klondike had sufficient samples for  
10 comparison).  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21

22 The  $\delta^{13}\text{C}_{\text{col}}$  and  $\delta^{15}\text{N}_{\text{col}}$  of BC proboscideans (unidentified to genus) were intermediate  
23 between those of mammoths and mastodons from other locations, and did not differ significantly  
24 from any other group. Herschel Island unknown proboscideans (unidentified to genus) had  
25 significantly different  $\delta^{13}\text{C}_{\text{col}}$  and/or  $\delta^{15}\text{N}_{\text{col}}$  from Yukon mastodons (Old Crow and Klondike)  
26 (Kruskall-Wallis and Dunn's tests,  $p < 0.05$  and  $p < 0.001$ , for  $\delta^{13}\text{C}_{\text{col}}$  and  $\delta^{15}\text{N}_{\text{col}}$ , respectively) but  
27 did not differ significantly from Yukon mammoths.  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38

## 39 **Location of Origin**

## 40 **Collagen Analyses**

41 Because mammoths and mastodons had significantly different  $\delta^{15}\text{N}_{\text{col}}$  and  $\delta^{13}\text{C}_{\text{col}}$  in Old  
42 Crow and Klondike, statistical analyses considered the taxa separately for these locations.  
43 Mammoth, mastodon, and unknown proboscidean collagen data from Alberta were combined  
44 because they did not have significantly different means. BC and Herschel Island samples that  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 were not identified to genus were included as separate groups in the statistical analyses, and  
4  
5 compared to both mammoths and mastodons from other regions, as described above.  
6  
7

8 For mastodons, there were no significant differences among locations for  $\delta^{13}\text{C}_{\text{col}}$  or  
9  
10  $\delta^{15}\text{N}_{\text{col}}$ . For mammoths, there were clear differences among locations for  $\delta^{13}\text{C}_{\text{col}}$  (Kruskall-Wallis  
11  
12 test,  $p < 0.001$ ). Dunn's multiple comparison tests showed that differences in mammoth  $\delta^{13}\text{C}_{\text{col}}$   
13  
14 existed between Alberta and Old Crow (means of  $-20.5$  vs.  $-21.4\text{‰}$ ), Alberta and Herschel  
15  
16 Island (means of  $-20.5$  vs.  $-21.6\text{‰}$ ), and Klondike and Old Crow (means of  $-20.7$  vs.  $-21.4\text{‰}$ ).  
17  
18 The Kruskal-Wallis test suggested differences for mammoth  $\delta^{15}\text{N}_{\text{col}}$  in different locations  
19  
20 ( $p < 0.05$ ), but Dunn's multiple comparison tests showed no differences among groups. To be  
21  
22 conservative, we assume this contradictory result indicates no reliable significant differences.  
23  
24  
25  
26  
27  
28

### 29 **Bioapatite Carbonate Analyses**

30  
31 Bioapatite isotopic compositions for mammoths and mastodons did not differ  
32  
33 significantly in any of the regions studied, and were combined within each region in the  
34  
35 following comparisons. Results for tooth enamel are available only for Alberta, Klondike, Old  
36  
37 Crow, and Herschel Island. The Kruskal-Wallis test showed significant differences among  
38  
39 locations for  $\delta^{13}\text{C}_{\text{sc}}$  and  $\delta^{18}\text{O}_{\text{sc}}$  ( $p < 0.001$  in both cases). Dunn's multiple comparison tests showed  
40  
41 that differences for  $\delta^{13}\text{C}_{\text{sc}}$  existed between Old Crow and Klondike (means of  $-9.9$  vs.  $-11.5\text{‰}$ ),  
42  
43 and Old Crow and Herschel Island (means of  $-9.9$  vs.  $-13.7\text{‰}$ ), whereas differences for  $\delta^{18}\text{O}$   
44  
45 existed between Alberta and Old Crow (means of  $+14.2$  vs.  $+10.7\text{‰}$ ), and Alberta and Klondike  
46  
47 (means of  $+14.2$  vs.  $+11.0\text{‰}$ ).  
48  
49  
50  
51  
52  
53  
54

### 55 **Health Status**

1  
2  
3  
4  
5  
6 Two of the AB5 (P90.7.1) mammoth subsamples lacked collagen (M6-M7 cementum and  
7  
8 M7 root), but data were obtained for the remaining four subsamples (Table 3). Overall, AB5 had  
9  
10 the second-highest mean  $\delta^{15}\text{N}_{\text{col}}$  and the lowest  $\delta^{13}\text{C}_{\text{col}}$  of any Alberta proboscidean (Table 2).  
11  
12 The  $\delta^{13}\text{C}_{\text{col}}$  values of all AB5 subsamples were similar, with a range of only 0.4‰ (Table 3). The  
13  
14  $\delta^{15}\text{N}_{\text{col}}$  values of the three M6 subsamples were very similar, but that of the M7 was considerably  
15  
16 lower (by 1.2 to 1.5 ‰) (Table 3). Enamel was obtained only from the M6 crown. Its  $\delta^{13}\text{C}_{\text{sc}}$  was  
17  
18 the lowest of any Alberta proboscidean, but its  $\delta^{18}\text{O}$  was not an outlier (Table 2).  
19  
20  
21  
22  
23

## 24 Discussion

### 25 26 27 28 29 Taxonomic Identification

30  
31 The data suggest that  $\delta^{15}\text{N}_{\text{col}}$  might be useful for assigning preliminary taxonomic  
32  
33 designations (mammoth vs. mastodon) to isolated postcranial remains. Differences in mammoth  
34  
35 and mastodon  $\delta^{15}\text{N}_{\text{col}}$  were generally large, as shown previously for other regions (e.g., France et  
36  
37 al., 2007; Koch, 1991; Metcalfe et al., 2013), and there were no significant differences among  
38  
39 locations in the  $\delta^{15}\text{N}_{\text{col}}$  of either mammoths or mastodons.  
40  
41  
42  
43  
44  
45

### 46 Case Study 1: Alberta (individual specimens)

47  
48 The unidentified proboscidean bones from Alberta had a range for  $\delta^{15}\text{N}_{\text{col}}$  that exceeded  
49  
50 the range for the securely-designated mammoths and mastodons from the same region (Table 2,  
51  
52 Fig. 3). Despite the lack of statistical significance for the Alberta mammoth vs. mastodon  $\delta^{15}\text{N}_{\text{col}}$   
53  
54 comparison, we might reasonably hypothesize taxonomic designations for the unknown  
55  
56  
57  
58  
59  
60

1  
2  
3 proboscidean bones with the highest and lowest  $\delta^{15}\text{N}_{\text{col}}$  values. We therefore hypothesized that  
4  
5 AB3 (P95.1.427) (+10.9‰) and AB7 (P91.11.9) (+9.0‰) were mammoth, and AB12 (P96.12.1)  
6  
7 (+1.4‰) was mastodon.  
8  
9

10 To test our isotopically-derived taxonomic prediction for AB12 (P96.12.1), which had the  
11  
12 lowest  $\delta^{15}\text{N}_{\text{col}}$  of any specimen in the present study, we submitted a segment of unprocessed bone  
13  
14 (a remainder of the same piece used for isotopic analysis) for ancient DNA analysis. Contrary to  
15  
16 expectations, the DNA results suggested that the bone belongs to *Mammuthus* (Jake Enk and  
17  
18 Hendrik Poinar, personal communication, Jan. 30, 2014). To our knowledge, such a low  $\delta^{15}\text{N}_{\text{col}}$   
19  
20 has never been previously reported for *Mammuthus*. Interestingly, AB12 (P96.12.1) was the only  
21  
22 post-glacial specimen from Alberta (directly dated to  $10,743 \pm 100$   $^{14}\text{C}$  a BP, BGS 2131) (Burns,  
23  
24 2010), and was recovered further south than the other Alberta individuals analyzed in this study  
25  
26 (Table 1, Fig. 1). Thus, it may have inhabited a very different environment than the other (more  
27  
28 northern, glacial-aged) specimens. The very low  $\delta^{15}\text{N}_{\text{col}}$  of AB12 (P96.12.1), as well as the  
29  
30 greater variability in Alberta mammoth  $\delta^{15}\text{N}_{\text{col}}$  in general (SD = 2.4 for Alberta vs. 1.2 and 1.1‰  
31  
32 for Klondike and Old Crow, respectively) is likely related to Alberta's geographical position at  
33  
34 the point of convergence of the Cordilleran and Laurentide Ice Sheets, whose waxing and  
35  
36 waning would have had significant effects on the local nitrogen cycle. Both the Alberta and  
37  
38 Yukon proboscidean remains were from time-averaged deposits, but only Alberta was repeatedly  
39  
40 covered by glacial ice. The so-called ice-free corridor in Alberta not only facilitated gene flow  
41  
42 between southern and northern faunal populations (Burns, 1996; Shapiro et al., 2004; Wilson,  
43  
44 1996), but also underwent significant environmental transformations as the ice sheets advanced  
45  
46 and receded (Beaudoin and Oetelaar, 2003; Jass et al., 2011), drastically altering the soil-plant  
47  
48 nitrogen cycle. Plants and animals in recently deglaciated areas tend to have low  $\delta^{15}\text{N}$  as a result  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 of low temperatures, soil immaturity (low nitrogen availability), plant type, and mycorrhizal  
4 associations (Drucker et al., 2003; Drucker et al., 2012; Drucker et al., 2011; Hobbie et al., 1999;  
5 Hobbie et al., 2000; Szpak, 2014). The first proboscideans to enter Alberta after deglaciation  
6 (e.g., AB12 / P96.12.1) would therefore be expected to have had low  $\delta^{15}\text{N}$  values. In contrast, the  
7 Yukon locations were never covered by glacial ice and may not have experienced such dramatic  
8 environmental shifts, though we note that very low  $\delta^{15}\text{N}$  values have been observed for  
9 herbivores in unglaciated northwestern Europe during the Last Glacial Maximum (Dyke et al.,  
10 2002; Gautney and Holliday, 2015; Hedges et al., 2004; Richards and Hedges, 2003; Stevens and  
11 Hedges, 2004).

12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

Recent research supports the idea that the environmental niches of various proboscidean taxa were broader than traditionally envisioned (Baumann and Crowley, 2015; Metcalfe and Longstaffe, 2014; Saunders et al., 2010). The example of AB12 (P96.12.1) demonstrates that mammoths in Alberta occupied a range of environments, and suggests that taxonomic designations based on stable isotope analyses should ideally be attempted for specimens from known locations and time periods, where baseline isotopic compositions for the taxa in question have already been determined.

### **Case Study 2: British Columbia and Herschel Island (groups of specimens)**

Unfortunately, the paleontological record is incomplete and it is sometimes not possible to develop baseline isotopic data for all taxa at any given location, such as British Columbia and Herschel Island. BC proboscideans had  $\delta^{13}\text{C}_{\text{col}}$  and  $\delta^{15}\text{N}_{\text{col}}$  that were intermediate between those of mammoths and mastodons from other locations, and therefore did not differ significantly from either taxon. Although it is possible that the BC group includes both mammoths and mastodons, the very small range of  $\delta^{15}\text{N}_{\text{col}}$  for BC specimens (difference of 1.1‰ between maximum and

1  
2  
3 minimum values) suggests, rather, that the animals were all of the same taxon. In support of this  
4  
5 inference, ancient DNA analysis of BC1 (EH2009.008.0001), BC2 (EH1994.003.0041), BC4,  
6  
7 and BC5 (EH1994.003.0040) showed that nine genus-discriminating sites within two species-  
8  
9 delineable regions of the mitochondrial genome of these individuals matched those of  
10  
11 *Mammuthus* (Jake Enk and Hendrik Poinar, personal communication, Jan. 30, 2014).  
12  
13

14  
15 Herschel Island specimens not identified to genus had  $\delta^{13}\text{C}_{\text{col}}$  and  $\delta^{15}\text{N}_{\text{col}}$  that differed  
16  
17 significantly from Old Crow and Klondike mastodons, but were indistinguishable from Old  
18  
19 Crow or Klondike mammoths. This suggests that the specimens from Herschel Island were  
20  
21 predominantly *Mammuthus*, an interpretation consistent with the preponderance of mammoth  
22  
23 remains, and rarity of mastodon remains, in Alaska and Yukon (Harington, 2003; Zazula et al.,  
24  
25 2014).  
26  
27  
28  
29  
30  
31

### 32 **Location of Origin**

33  
34  
35

36 Bioapatite  $\delta^{13}\text{C}_{\text{sc}}$  and  $\delta^{18}\text{O}_{\text{sc}}$  could be useful for determining locations of origin, as they  
37  
38 are quite distinct among several locations (Fig. 4). The oxygen isotope compositions of Old  
39  
40 Crow and Klondike specimens are lower than those of Alberta specimens, as expected based on  
41  
42 latitudinal variations in meteoric water  $\delta^{18}\text{O}$  (Dansgaard, 1964; Rozanski et al., 1993). Contrary  
43  
44 to latitude-based expectations, the highest-latitude location (Herschel Island) had relatively high  
45  
46  $\delta^{18}\text{O}_{\text{sc}}$  values. This is likely a result of Herschel's close proximity to the sea, where local  
47  
48 evaporation of seawater would result in precipitation with a relatively high  $\delta^{18}\text{O}$  (Dansgaard,  
49  
50 1964; Rozanski et al., 1993). The similarity in  $\delta^{18}\text{O}_{\text{sc}}$  for Old Crow and Klondike specimens is  
51  
52 unexpected since they are at different latitudes, but could reflect different atmospheric moisture  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 or surface water source regions, and/or temporal differences among the specimens. The carbon  
4  
5 isotope compositions of the Herschel Island specimens are strikingly low, which may reflect  
6  
7 significantly greater water availability in proximity to the coast (Kohn, 2010). Likewise, the  
8  
9 highest  $\delta^{13}\text{C}$  values in Old Crow may indicate that the driest conditions were present in that  
10  
11 location.  
12  
13

14  
15       Regardless of the causes of variation, the data presented here provide a baseline for  
16  
17 investigating the origins of museum specimens lacking collection data. Alberta specimens can  
18  
19 clearly be distinguished from Yukon specimens using  $\delta^{13}\text{C}_{\text{sc}}$  and  $\delta^{18}\text{O}_{\text{sc}}$  (Fig. 4). Specimens from  
20  
21 different regions of Yukon (Klondike, Old Crow, Herschel Island) may also be distinguishable  
22  
23 using a combination of  $\delta^{18}\text{O}_{\text{sc}}$  and  $\delta^{13}\text{C}_{\text{sc}}$  (and possibly  $\delta^{13}\text{C}_{\text{col}}$ ), though there is some overlap  
24  
25 between Old Crow and Klondike specimens (Fig. 4). Further, because of the predictable change  
26  
27 in meteoric water  $\delta^{18}\text{O}$  with latitude,  $\delta^{18}\text{O}_{\text{sc}}$  could be used to distinguish between regions  
28  
29 included in the present study and areas further south. For example, mammoths and mastodons  
30  
31 from Ontario and western New York had  $\delta^{18}\text{O}_{\text{sc}}$  values between  $\sim +19$  and  $+23\text{‰}$  (Metcalfé et  
32  
33 al., 2013), and mammoths from Arizona had  $\delta^{18}\text{O}_{\text{sc}}$  values between  $\sim +22$  and  $+28\text{‰}$  (Metcalfé  
34  
35 et al., 2011), in contrast to values of  $\sim +10$  to  $+15\text{‰}$  presented here (all locations combined). Of  
36  
37 course, temporal differences among regions can complicate such comparisons, since animals  
38  
39 living in colder intervals would have lower  $\delta^{18}\text{O}_{\text{sc}}$  than those living in warmer intervals, all else  
40  
41 being equal (Ayliffe et al., 1992; Koch, 1998).  
42  
43  
44  
45  
46  
47

48  
49       Unlike the situation presented here, mammoth and mastodon  $\delta^{18}\text{O}$  values may not be the  
50  
51 same in every location. For example, in the Great Lakes region there was no overlap between  
52  
53  $\delta^{18}\text{O}_{\text{sc}}$  of mammoths and mastodons (Metcalfé et al., 2013). This demonstrates the need for  
54  
55 baseline studies in the particular areas and time periods of interest, to determine the range of  
56  
57  
58  
59  
60

1  
2  
3 values and degree of overlap among taxa. Only after these are established can comparisons with  
4  
5 other regions and inferences about geographic origins be made.  
6  
7

8 Since knowing the proboscidean taxon was deemed unnecessary for  $\delta^{13}\text{C}_{\text{sc}}$  and  $\delta^{18}\text{O}_{\text{sc}}$   
9  
10 comparisons, these proxies could be applied to isolated postcranial elements that lack taxonomic  
11  
12 designation. However, bone bioapatite is much more susceptible to diagenetic alteration of its  
13  
14 isotopic composition, and its analysis (not attempted here) is not recommended (Ayliffe et al.,  
15  
16 1994; Koch et al., 1997).  
17  
18  
19

20 Collagen isotopic compositions were not useful for distinguishing location of origin for  
21  
22 mastodons, and were of limited use for mammoths. Although the  $\delta^{13}\text{C}_{\text{col}}$  of mammoths in several  
23  
24 locations differed significantly, the magnitude of the differences was very small ( $\sim 1\%$  or less).  
25  
26 Compared to these differences among means, variability within a single location was relatively  
27  
28 large ( $\sim 1\text{-}2\%$ ). As such, it would not be possible to match an isolated specimen to any particular  
29  
30 location based on  $\delta^{13}\text{C}_{\text{col}}$  alone. However,  $\delta^{13}\text{C}_{\text{col}}$  and/or  $\Delta^{13}\text{C}_{\text{sc-col}}$  (the difference between  $\delta^{13}\text{C}_{\text{sc}}$   
31  
32 and  $\delta^{13}\text{C}_{\text{col}}$ , which can be calculated for specimens with isotopic data for both collagen and  
33  
34 bioapatite) might provide secondary lines of evidence for location of origin in certain cases. For  
35  
36 example, Old Crow mammoths had  $\Delta^{13}\text{C}_{\text{sc-col}}$  values between 10.1 and 13.1‰, whereas  
37  
38 mammoths and mastodons from other locations had  $\Delta^{13}\text{C}_{\text{sc-col}}$  between 8.0 and 10.1‰.  
39  
40  
41  
42  
43  
44  
45

## 46 **Health Status**

47  
48  
49  
50

51 The abnormal M6/M7 (AB5 / P90.7.1) mammoth specimen provides a clear example of a  
52  
53 skeletal anomaly that directly impacted the health of the individual, and a unique opportunity for  
54  
55 investigating the links between isotopic compositions and nutritional stress in an individual  
56  
57  
58  
59  
60



1  
2  
3 extinct animal. The subsamples of the AB5 specimen likely developed in the following order: (1)  
4 anterior M6 dentin, (2) anterior M6 cementum, (3) M6 root dentin, (4) M7 crown cementum, (5)  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

extinct animal. The subsamples of the AB5 specimen likely developed in the following order: (1) anterior M6 dentin, (2) anterior M6 cementum, (3) M6 root dentin, (4) M7 crown cementum, (5) cementum between the M6 and M7, (6) M7 root dentin (Appendix S1) (Haynes, 1991; Hillson, 2005; Laws, 1966; Maschenko, 2002; Metcalfe et al., 2010). Unfortunately the two latest-developing subsamples lacked collagen, depriving us of data that would have represented the time closest to death. The fact that the M6 root and M7 crown had very distinct  $\delta^{15}\text{N}_{\text{col}}$  values suggests that they formed at different times, with the M7 forming after the M6 was fully developed (Appendix S1). This is in agreement with Burns et al.'s (2003:82) inference that "the M6 was fully erupted when the M7 was forced into place." The lack of space for the M7 caused the still-developing lamellae to deform as they impacted against the fully-formed M6. Based on this evidence, in combination with elephant tooth development times and the estimated age-at-death for this specimen, we can surmise that the M7 formed primarily during the last 4-5 years of the animal's life (Appendix S1). It is currently not possible to make a more precise estimate of how much time elapsed between formation of the M7 crown cementum (our latest-developing sample that yielded results) and the animal's death (Appendix S1).

The  $\delta^{15}\text{N}_{\text{col}}$  values of AB5 (P90.7.1) are relatively high, as expected for nutritional stress leading to the catabolism of body tissues, which tends to cause increases of  $<+1$  to  $+2\%$  (Hatch, 2012; McCue and Pollock, 2008; Reitsema, 2013). However, contrary to expectations for starvation later in life, the M7 had a lower  $\delta^{15}\text{N}_{\text{col}}$  than the M6 tissues (Table 3). This suggests that body proteins were not being catabolized late in life, during the development of the M7 crown. Nutritional stress leading to reliance on body fats (which would not affect  $\delta^{15}\text{N}$ ) remains a possibility (Hatch, 2012). It is also possible, however, that the relatively high  $\delta^{15}\text{N}$  values reflect the environment in which the animal was living, and are unrelated to nutritional stress.

1  
2  
3 The low  $\delta^{13}\text{C}$  values of AB5 (P90.7.1) relative to other Alberta mammoths also do not  
4 support expectations for severe starvation and fasting (which tend to have either no effect on  
5 tissue  $\delta^{13}\text{C}$ , or cause a slight increase) (Hatch, 2012; McCue and Pollock, 2008). Although the  
6  
7  
8  
9 low  $\delta^{13}\text{C}$  values could reflect increased fat metabolism (Bocherens, 2003; Bocherens et al., 1996;  
10 Hatch, 2012; Polischuk et al., 2001), there is no difference in the  $\delta^{13}\text{C}_{\text{col}}$  of the M6 root and M7  
11  
12  
13  
14 crown, suggesting no change in the last several years prior to death. Together, the differences in  
15  
16  
17  
18  
19  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  among subsamples of AB5 suggest a change in diet, environment, or physiology  
20 in the years leading up to death, but there is no isotopic evidence for starvation (neither  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

## Conclusions

Stable isotopes can be of use in designating isolated proboscidean skeletal remains as mammoth or mastodon, but such designations remain tentative in the absence of morphological or molecular data. Mammoths tend to have higher  $\delta^{15}\text{N}_{\text{col}}$  and lower  $\delta^{13}\text{C}_{\text{col}}$  than mastodons, but the specific range of isotopic compositions differs among geographic locations and time periods, and may not be significantly different in every region. Mammoths, in particular, can have a very large range of  $\delta^{15}\text{N}$  values, as shown for specimens from Alberta. Ideally, taxonomic designations based on stable isotopes should only be attempted for specimens from known locations and time periods, where baseline (preferably non-overlapping) isotopic compositions for the taxa in question have been determined previously.

1  
2  
3 We have shown that extraordinary exceptions to general trends in taxonomic differences  
4 can exist (i.e., a mammoth with a  $\delta^{15}\text{N}_{\text{col}}$  of +1.4‰). This extremely low value may be typical for  
5 fauna inhabiting recently-deglaciated late Pleistocene landscapes of Alberta. Further research  
6 into the isotopic compositions of plants and animals present at that time and location would be  
7 useful to support this hypothesis. This example shows that isotopic analyses may be useful in  
8 determining outliers that may be selected for additional research (e.g., DNA, morphology) and  
9 may inspire broader research questions. These data also demonstrate the environmental  
10 adaptability of *Mammuthus*.  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21

22 Our results also demonstrate that stable isotope analysis can be helpful in assigning  
23 geographical provenance to isolated museum samples that lack collection data. Bioapatite  $\delta^{13}\text{C}_{\text{sc}}$   
24 and  $\delta^{18}\text{O}_{\text{sc}}$  are most useful for determining locations of origin, and could be used to distinguish  
25 proboscideans from Alberta, Old Crow, Klondike, Herschel Island, and locations further south  
26 (e.g., Arizona and the Great Lakes). Collagen  $\delta^{13}\text{C}$  could provide a secondary line of evidence  
27 for determining the location of origin of mammoths, but  $\delta^{15}\text{N}_{\text{col}}$  was not useful for this  
28 application. The isotopic data we have provided can be used as a baseline for future studies of  
29 specimens lacking provenance, and are particularly useful since the Yukon is the most prolific  
30 source of proboscidean fossils in Canada. It is highly probable that many skeletal remains  
31 without provenance in local museum collections are from Yukon, and isotopic determination of  
32 their location of origin may be possible. Furthermore, the isotopic baseline we have provided  
33 could be used to support inferences of migration, but only if climate-related variations can be  
34 ruled out.  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51

52 The AB5 (P90.7.1) mammoth possessed a dental anomaly that caused impaired chewing,  
53 which was hypothesized to have caused nutritional stress and ultimately the animal's death  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 (Burns et al., 2003). Our data indicate that in the years prior to its death, the animal experienced  
4 a change in diet, environment, or physiology – but not one consistent with nutritional stress. This  
5  
6 does not necessarily contradict the interpretation that this animal starved to death (Burns et al.,  
7  
8  
9 2003), since the precise amount of time that elapsed between the formation of the analyzed  
10  
11 tissues and death remains unknown.  
12  
13  
14  
15  
16

### 17 **Acknowledgements**

18  
19 The authors thank Kim Law, Li Huang, Reba Macdonald, and the staff and students of  
20  
21 the Laboratory for Stable Isotope Science (LSIS) for laboratory assistance and support, and Ryan  
22  
23 Manickchand for photographing the mammoth teeth. Funding for this study was provided by the  
24  
25 Natural Sciences and Engineering Research Council of Canada (NSERC Canada Government  
26  
27 Scholarship- Doctoral to J.Z.M.; NSERC Discovery Grant 38940 to F.J.L.), the Canada  
28  
29 Foundation for Innovation (Grant 2732 to F.J.L.) and the Ontario Research Fund (and its  
30  
31 precursors (Grant 2732 to F.J.L.). Research time provided through the Canada Research Chairs  
32  
33 program (Grant 950-226925 to F.J.L.) and a Killam Postdoctoral Research Fellowship at UBC  
34  
35 (J.Z.M.) is also gratefully acknowledged. This is LSIS Contribution #331.  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

## List of Tables

Table 1. Geographic and temporal context and sampling information for specimens used in this study.

Table 2. The isotopic compositions of collagen (various tissues) and structural carbonate in bioapatite (enamel) for mammoths, mastodons, and unknown proboscideans analyzed in this study and by Zazula et al. (2014).

Table 3. Isotopic results for collagen extracted from different areas on AB5 / P90.7.1 (mammoth M6 and M7).

## List of Figures

Figure 1. Locations mentioned in the text. Red dots indicate provenance for specimens used to establish baseline data for location of origin studies. VI = Vancouver Island, C = Cochrane, E/V = Edmonton/Villeneuve, B = Bezanson, K = Klondike, OC = Old Crow, H = Herschel Island.

Adapted from <http://jan.ucc.nau.edu>

Figure 2. (A) Lingual view of the sampled left M6/M7 *Mammuthus* teeth (AB5 / P90.7.1) as compared to (B) the normal condition in a left M6 *Mammuthus* tooth (P84.5.1). Scale bars = 10 cm.

1  
2  
3 Figure 3. Carbon and nitrogen isotope compositions of collagen for (A) *Mammut*, (B) unknown  
4 proboscideans, and (C) *Mammuthus* from Western Canada locations.  
5  
6  
7  
8  
9

10 Figure 4. Carbon and oxygen isotope results obtained for structural carbonate in enamel  
11 bioapatite from *Mammuthus* (open symbols and stars) and *Mammut* (closed symbols). Polygons  
12 enclose specimens from the different regions. AB = Alberta, OC = Old Crow, K = Klondike, H =  
13 Herschel Island.  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23

## 24 **Supplementary Information**

25  
26  
27  
28

29 Table S1. Isotopic results for collagen extracted from different tissues within the same  
30 individuals (excluding AB5).  
31  
32  
33  
34  
35

36 Appendix S1. Developmental timing of the AB5 (P90.7.1) mammoth M6/M7  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

## References Cited

- 1  
2  
3  
4  
5  
6 Agenbroad LD. 2005. North American proboscideans: Mammoths - the state of knowledge,  
7 2003. *Quaternary International* **126-28**: 73-92.
- 8 Ambrose SH. 1990. Preparation and characterization of bone and tooth collagen for isotopic  
9 analysis. *Journal of Archaeological Science* **17**: 431-451.
- 10 Amundson R, Austin AT, Schuur EAG, Yoo K, Matzek V, Kendall C, Uebersax A, Brenner D,  
11 Baisden WT. 2003. Global patterns of the isotopic composition of soil and plant nitrogen.  
12 *Global Biogeochemical Cycles* **17**: 1031, doi:10.1029/2002GB001903.
- 13 Arens NC, Jahren AH, Amundson R. 2000. Can C<sub>3</sub> plants faithfully record the carbon isotopic  
14 composition of atmospheric carbon dioxide? *Paleobiology* **26**: 137-164.
- 15 Ayliffe LK, Chivas AR, Leakey MG. 1994. The retention of primary oxygen isotope  
16 compositions of fossil elephant skeletal phosphate. *Geochimica et Cosmochimica Acta*  
17 **58**: 5291-5298.
- 18 Ayliffe LK, Lister AM, Chivas AR. 1992. The preservation of glacial-interglacial climatic  
19 signatures in the oxygen isotopes of elephant skeletal phosphate. *Palaeogeography*  
20 *Palaeoclimatology Palaeoecology* **99**: 179-191.
- 21 Baumann EJ, Jr., Crowley BE. 2015. Stable isotopes reveal ecological differences amongst now-  
22 extinct proboscideans from the Cincinnati region, USA. *Boreas* **44**: 240-254.
- 23 Beaudoin AB, Oetelaar GA. 2003. The changing ecophysical landscape of southern Alberta  
24 during the late Pleistocene and early Holocene. *Plains Anthropologist* **48**: 187-207.
- 25 Bocherens H. 2003. Isotopic biogeochemistry and the paleoecology of the mammoth steppe  
26 fauna. *Deinsea* **9**: 57-71.
- 27 Bocherens H, Fizet M, Mariotti A, Gangloff RA, Burns JA. 1994. Contribution of isotopic  
28 biogeochemistry (<sup>13</sup>C, <sup>15</sup>N, <sup>18</sup>O) to the paleoecology of mammoths (*Mammuthus*  
29 *primigenius*). *Historical Biology* **7**: 187-202.
- 30 Bocherens H, Pacaud G, Lazarev PA, Mariotti A. 1996. Stable isotope abundances (<sup>13</sup>C, <sup>15</sup>N) in  
31 collagen and soft tissues from Pleistocene mammals from Yakutia: Implications for the  
32 palaeobiology of the Mammoth Steppe. *Palaeogeography Palaeoclimatology*  
33 *Palaeoecology* **126**: 31-44.
- 34 Bowen GJ. 2010. Isoscapes: spatial pattern in isotopic biogeochemistry. *Annual Review of Earth*  
35 *and Planetary Sciences* **38**: 161-187.
- 36 Bowen GJ, Wassenaar LI, Hobson KA. 2005. Global application of stable hydrogen and oxygen  
37 isotopes to wildlife forensics. *Oecologia* **143**: 337-348.
- 38 Bowen GJ, Wilkinson B. 2002. Spatial distribution of δ<sup>18</sup>O in meteoric precipitation. *Geology*  
39 **30**: 315-318.
- 40 Brooks JR, Flanagan LB, Buchmann N, Ehleringer JR. 1997. Carbon isotope composition of  
41 boreal plants: Functional grouping of life forms. *Oecologia* **110**: 301-311.
- 42 Bryant JD, Froelich PN. 1995. A model of oxygen-isotope fractionation in body-water of large  
43 mammals. *Geochimica et Cosmochimica Acta* **59**: 4523-4537.
- 44 Buckley M, Fraser S, Herman J, Melton ND, Mulville J, Palsdottir AH. 2014. Species  
45 identification of archaeological marine mammals using collagen fingerprinting. *Journal*  
46 *of Archaeological Science* **41**: 631-641.
- 47 Buckley M, Kansa SW. 2011. Collagen fingerprinting of archaeological bone and teeth remains  
48 from Domuztepe, South Eastern Turkey. *Archaeological and Anthropological Sciences* **3**:  
49 271-280.
- 50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 Burns JA. 1996. Vertebrate paleontology and the alleged ice-free corridor: the meat of the  
4 matter. *Quaternary International* **32**: 107-112.
- 5 Burns JA. 2010. Mammalian faunal dynamics in Late Pleistocene Alberta, Canada. *Quaternary*  
6 *International* **217**: 37-42.
- 7 Burns JA, G. BC, Mol D. 2003. An extraordinary woolly mammoth molar from Alberta, Canada.  
8 *Deinsea* **9**: 77-85.
- 9 Camin F, Wehrens R, Bertoldi D, Bontempo L, Ziller L, Perini M, Nicolini G, Nocetti M,  
10 Larcher R. 2012. H, C, N and S stable isotopes and mineral profiles to objectively  
11 guarantee the authenticity of grated hard cheeses. *Anal. Chim. Acta* **711**: 54-59.
- 12 Cerling TE, Ehleringer JR, West A, Stange E, Dorigan J. 2003. Forensic applications of stable  
13 isotopes in hair. *Forensic Science International* **136**: 172-172.
- 14 Chesson LA, Tipple BJ, Erkkila BR, Cerling TE, Ehleringer JR. 2011. B-HIVE: Beeswax  
15 hydrogen isotopes as validation of environment. Part I: Bulk honey and honeycomb  
16 stable isotope analysis. *Food Chemistry* **125**: 576-581.
- 17 Chesson LA, Valenzuela LO, O'Grady SP, Cerling TE, Ehleringer JR. 2010a. Hydrogen and  
18 oxygen stable isotope ratios of milk in the United States. *Journal of Agricultural and*  
19 *Food Chemistry* **58**: 2358-2363.
- 20 Chesson LA, Valenzuela LO, O'Grady SP, Cerling TE, Ehleringer JR. 2010b. Links between  
21 purchase location and stable isotope ratios of bottled water, soda, and beer in the United  
22 States. *Journal of Agricultural and Food Chemistry* **58**: 7311-7316.
- 23 Clementz MT. 2012. New insight from old bones: stable isotope analysis of fossil mammals.  
24 *Journal of Mammalogy* **93**: 368-380.
- 25 Clementz MT, Fox-Dobbs K, Wheatley PV, Koch PL, Doak DF. 2009. Revisiting old bones:  
26 coupled carbon isotope analysis of bioapatite and collagen as an ecological and  
27 palaeoecological tool. *Geological Journal* **44**: 605-620.
- 28 Coplen TB. 1994. Reporting stable hydrogen, carbon, and oxygen isotopic abundances. *Pure and*  
29 *Applied Chemistry* **66**: 271-276.
- 30 Coplen TB, Brand WA, Gehre M, Groning M, Meijer HAJ, Toman B, Verkouteren RM. 2006.  
31 New guidelines for  $\delta^{13}\text{C}$  measurements. *Analytical Chemistry* **78**: 2439-2441.
- 32 Corr LT, Richards MP, Jim S, Ambrose SH, Mackie A, Beattie O, Evershed RP. 2008. Probing  
33 dietary change of the Kwaday Dan Ts'inchi individual, an ancient glacier body from  
34 British Columbia: I. Complementary use of marine lipid biomarker and carbon isotope  
35 signatures as novel indicators of a marine diet. *Journal of Archaeological Science* **35**:  
36 2102-2110.
- 37 Craine JM, Elmore AJ, Aidar MPM, Bustamante M, Dawson TE, Hobbie EA, Kahmen A, Mack  
38 MC, McLauchlan KK, Michelsen A, Nardoto GB, Pardo LH, Penuelas J, Reich PB,  
39 Schuur EAG, Stock WD, Templer PH, Virginia RA, Welker JM, Wright IJ. 2009. Global  
40 patterns of foliar nitrogen isotopes and their relationships with climate, mycorrhizal  
41 fungi, foliar nutrient concentrations, and nitrogen availability. *New Phytol.* **183**: 980-992.
- 42 Cummings LS, Albert RM. 2007. Phytolith and starch analysis of Dent site mammoth teeth  
43 calculus: new evidence for Late Pleistocene mammoth diets and environments, In  
44 *Frontiers in Colorado Paleoindian Archaeology: From the Dent Site to the Rocky*  
45 *Mountains*. Brunswig, R.H., Pitblado, B.L. (eds.) University Press of Colorado: Boulder;  
46 185-192.
- 47 Dansgaard W. 1964. Stable isotopes in precipitation. *Tellus* **16**: 436-468.
- 48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60



- 1  
2  
3 Debruyne R, Chu G, King CE, Bos K, Kuch M, Schwarz C, Szpak P, Grocke DR, Matheus P,  
4 Zazula G, Guthrie D, Froese D, Buigues B, de Marliave C, Flemming C, Poinar D, Fisher  
5 D, Southon J, Tikhonov AN, MacPhee RDE, Poinar HN. 2008. Out of America: Ancient  
6 DNA evidence for a New World origin of Late Quaternary woolly mammoths. *Current*  
7 *Biology* **18**: 1320-1326.
- 8  
9 DeNiro MJ. 1985. Post-mortem preservation and alteration of "in vivo" bone collagen ratios:  
10 implications for paleodietary analysis. *Nature* **317**: 806-809.
- 11 DeNiro MJ, Epstein S. 1978. Influence of diet on the distribution of carbon isotopes in animals.  
12 *Geochimica et Cosmochimica Acta* **42**: 495-406.
- 13 DeNiro MJ, Epstein S. 1981. Influence of diet on the distribution of nitrogen isotopes in animals.  
14 *Geochimica et Cosmochimica Acta* **45**: 341-351.
- 15 Dirks W, Bromage TG, Agenbroad LD. 2012. The duration and rate of molar plate formation in  
16 *Palaeoloxodon cypristes* and *Mammuthus columbi* from dental histology. *Quaternary*  
17 *International* **255**: 79-85.
- 18  
19 Dreimanis A. 1967. Mastodons, their geologic age and extinctions in Ontario, Canada. *Canadian*  
20 *Journal of Earth Sciences* **4**: 663-675.
- 21 Dreimanis A. 1968. Extinction of mastodons in eastern North America: testing a new climatic-  
22 environmental hypothesis. *The Ohio Journal of Science* **68**: 337-352.
- 23 Drucker DG, Bocherens H, Billiou D. 2003. Evidence for shifting environmental conditions in  
24 Southwestern France from 33 000 to 15 000 years ago derived from carbon-13 and  
25 nitrogen-15 natural abundances in collagen of large herbivores. *Earth and Planetary*  
26 *Science Letters* **216**: 163-173.
- 27 Drucker DG, Bridault A, Cupillard C. 2012. Environmental context of the Magdalenian  
28 settlement in the Jura Mountains using stable isotope tracking ( $^{13}\text{C}$ ,  $^{15}\text{N}$ ,  $^{34}\text{S}$ ) of bone  
29 collagen from reindeer (*Rangifer tarandus*). *Quaternary International* **272**: 322-332.
- 30 Drucker DG, Bridault A, Hobson KA, Szuma E, Bocherens H. 2008. Can carbon-13 in large  
31 herbivores reflect the canopy effect in temperate and boreal ecosystems? Evidence from  
32 modern and ancient ungulates. *Palaeogeography Palaeoclimatology Palaeoecology* **266**:  
33 69-82.
- 34 Drucker DG, Hobson KA, Ouellet JP, Courtois R. 2010. Influence of forage preferences and  
35 habitat use on  $^{13}\text{C}$  and  $^{15}\text{N}$  abundance in wild caribou (*Rangifer tarandus caribou*) and  
36 moose (*Alces alces*) from Canada. *Isotopes in Environmental and Health Studies* **46**: 107-  
37 121.
- 38 Drucker DG, Kind CJ, Stephan E. 2011. Chronological and ecological information on Late-  
39 glacial and early Holocene reindeer from northwest Europe using radiocarbon (C-14) and  
40 stable isotope (C-13, N-15) analysis of bone collagen: Case study in southwestern  
41 Germany. *Quaternary International* **245**: 218-224.
- 42 Dupras TL, Schwarcz HP. 2001. Strangers in a strange land: Stable isotope evidence for human  
43 migration in the Dakhleh Oasis, Egypt. *Journal of Archaeological Science* **28**: 1199-  
44 1208.
- 45 Dyke AS, Andrews JT, Clark PU, England JH, Miller GH, Shaw J, Veillette JJ. 2002. The  
46 Laurentide and Inuitian ice sheets during the Last Glacial Maximum. *Quaternary*  
47 *Science Reviews* **21**: 9-31.
- 48 Ehleringer JR, Bowen GJ, Chesson LA, West AG, Podlesak DW, Cerling TE. 2008. Hydrogen  
49 and oxygen isotope ratios in human hair are related to geography. *Proceedings of the*  
50 *National Academy of Sciences of the United States of America* **105**: 2788-2793.
- 51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 Enk J, Devault A, Debruyne R, King CE, Treangen T, O'Rourke D, Salzberg SL, Fisher D,  
4 MacPhee R, Poinar H. 2011. Complete Columbian mammoth mitogenome suggests  
5 interbreeding with woolly mammoths. *Genome Biology* **12**: R51.  
6  
7 Feranec RS. 2004. Geographic variation in the diet of hypsodont herbivores from the  
8 Rancholabrean of Florida. *Palaeogeography Palaeoclimatology Palaeoecology* **207**: 359-  
9 369.  
10  
11 Fox-Dobbs K, Leonard JA, Koch PL. 2008. Pleistocene megafauna from eastern Beringia:  
12 Paleoecological and paleoenvironmental interpretations of stable carbon and nitrogen  
13 isotope and radiocarbon records. *Palaeogeography Palaeoclimatology Palaeoecology*  
14 **261**: 30-46.  
15  
16 France CAM, Zelanko PM, Kaufman AJ, Holtz TR. 2007. Carbon and nitrogen isotopic analysis  
17 of Pleistocene mammals from the Saltville Quarry (Virginia, USA): Implications for  
18 trophic relationships. *Palaeogeography Palaeoclimatology Palaeoecology* **249**: 271-282.  
19  
20 Fraser I, Meier-Augenstein W. 2007. Stable H-2 isotope analysis of modern-day human hair and  
21 nails can aid forensic human identification. *Rapid Communications in Mass Spectrometry*  
22 **21**: 3279-3285.  
23  
24 Fuller BT, Fuller JL, Harris DA, Hedges REM. 2006. Detection of breastfeeding and weaning in  
25 modern human infants with carbon and nitrogen stable isotope ratios. *American Journal*  
26 *of Physical Anthropology* **129**: 279-293.  
27  
28 Garvie-Lok SJ, Varney TL, Katzenberg MA. 2004. Preparation of bone carbonate for stable  
29 isotope analysis: the effects of treatment time and acid concentration. *Journal of*  
30 *Archaeological Science* **31**: 763-776.  
31  
32 Gautney JR, Holliday TW. 2015. New estimations of habitable land area and human population  
33 size at the Last Glacial Maximum. *Journal of Archaeological Science* **58**: 103-112.  
34  
35 Gobetz KE, Bozarth SR. 2001. Implications for late Pleistocene mastodon diet from opal  
36 phytoliths in tooth calculus. *Quaternary Research* **55**: 115-122.  
37  
38 Green JL, Semperebon GM, Solounias N. 2005. Reconstructing the palaeodiet of Florida *Mammot*  
39 *americanum* via low-magnification stereomicroscopy. *Palaeogeography*  
40 *Palaeoclimatology Palaeoecology* **223**: 34-48.  
41  
42 Guthrie RD. 2001. Origin and causes of the mammoth steppe: a story of cloud cover, woolly  
43 mammal tooth pits, buckles, and inside-out Beringia. *Quaternary Science Reviews* **20**:  
44 549-574.  
45  
46 Harington CR. 1990. Vertebrates of the last interglaciation in Canada: a review, with new data.  
47 *Geographie Physique et Quaternaire* **44**: 375-387.  
48  
49 Harington CR. 2003. *Annotated bibliography of Quaternary vertebrates of northern North*  
50 *America, with radiocarbon dates*. University of Toronto Press: Toronto.  
51  
52 Hartman G. 2011. Are elevated  $\delta^{15}\text{N}$  values in herbivores in hot and arid environments caused by  
53 diet or animal physiology? *Functional Ecology* **25**: 122-131.  
54  
55 Hatch KA. 2012. The use and application of stable isotope analysis to the study of starvation,  
56 fasting, and nutritional stress in animals, In *Comparative Physiology of Fasting,*  
57 *Starvation, and Food Limitation*. McCue, M.D. (Ed.) Springer Berlin Heidelberg; 337-  
58 364.  
59  
60 Haynes G. 1991. *Mammoths, mastodonts, and elephants: biology, behavior, and the fossil*  
*record*. Cambridge University Press: Cambridge New York.  
Hedges REM, Stevens RE, Richards MP. 2004. Bone as a stable isotope archive for local  
climatic information. *Quaternary Science Reviews* **23**: 959-965.

- 1  
2  
3 Hedges REM, Thompson JMA, Hull BD. 2005. Stable isotope variation in wool as a means to  
4 establish Turkish carpet provenance. *Rapid Communications in Mass Spectrometry* **19**:  
5 3187-3191.  
6  
7 Hillson S. 1996. *Dental Anthropology*. Cambridge University Press: Cambridge.  
8 Hillson S. 2005. *Teeth*. In Cambridge manuals in archaeology (New York, Cambridge University  
9 Press).  
10 Hobbie EA, Macko SA, Shugart HH. 1999. Insights into nitrogen and carbon dynamics of  
11 ectomycorrhizal and saprotrophic fungi from isotopic evidence. *Oecologia* **118**: 353-360.  
12 Hobbie EA, Macko SA, Williams M. 2000. Correlations between foliar  $\delta^{15}\text{N}$  and nitrogen  
13 concentrations may indicate plant-mycorrhizal interactions. *Oecologia* **122**: 273-283.  
14 Hobson KA, Koehler G. 2015. On the use of stable oxygen isotope ( $\delta^{18}\text{O}$ ) measurements for  
15 tracking avian movements in North America. *Ecology and Evolution* **5**: 799-806.  
16 Hoppe KA. 2004. Late Pleistocene mammoth herd structure, migration patterns, and Clovis  
17 hunting strategies inferred from isotopic analyses of multiple death assemblages.  
18 *Paleobiology* **30**: 129-145.  
19  
20 Iacumin P, Nikolaev V, Ramigni M. 2000. C and N stable isotope measurements on Eurasian  
21 fossil mammals, 40 000 to 10 000 years BP: Herbivore physiologies and  
22 palaeoenvironmental reconstruction. *Palaeogeography Palaeoclimatology Palaeoecology*  
23 **163**: 33-47.  
24  
25 IAEA/WMO. 2010. *Global Network of Isotopes in Precipitation. The GNIP Database*.  
26 Accessible at: <http://www.iaea.org/water>.  
27  
28 Jass CN, Burns JA, Milot PJ. 2011. Description of fossil muskoxen and relative abundance of  
29 Pleistocene megafauna in central Alberta. *Canadian Journal of Earth Sciences* **48**: 793-  
30 800.  
31  
32 Jenkins SG, Partridge ST, Stephenson TR, Farley SD, Robbins CT. 2001. Nitrogen and carbon  
33 isotope fractionation between mothers, neonates, and nursing offspring. *Oecologia* **129**:  
34 336-341.  
35  
36 Keenleyside A, Schwarcz HP, Panayotova K. 2011. Oxygen isotopic evidence of residence and  
37 migration in a Greek colonial population on the Black Sea. *Journal of Archaeological*  
38 *Science* **38**: 2658-2666.  
39  
40 Knudson KJ, Price TD. 2007. Utility of multiple chemical techniques in archaeological  
41 residential mobility studies: Case studies from Tiwanaku- and Chiribaya-affiliated sites in  
42 the Andes. *American Journal of Physical Anthropology* **132**: 25-39.  
43  
44 Koch PL. 1991. The isotopic ecology of Pleisocene proboscideans. *Journal of Vertebrate*  
45 *Paleontology* **11**: 40A.  
46  
47 Koch PL. 1998. Isotopic reconstruction of past continental environments. *Annual Review of*  
48 *Earth and Planetary Sciences* **26**: 573-613.  
49  
50 Koch PL, Fogel ML, Tuross N. 1994. Tracing the diets of fossil animals using stable isotopes, In  
51 *Stable Isotopes in Ecology and Environmental Science*. Lathja, K., Michener, R.H. (eds.)  
52 Blackwell Scientific Publications: Oxford; 63-92.  
53  
54 Koch PL, Hoppe KA, Webb SD. 1998. The isotopic ecology of late Pleistocene mammals in  
55 North America - Part 1. Florida. *Chemical Geology* **152**: 119-138.  
56  
57 Koch PL, Tuross N, Fogel ML. 1997. The effects of sample treatment and diagenesis on the  
58 isotopic integrity of carbonate in biogenic hydroxylapatite. *Journal of Archaeological*  
59 *Science* **24**: 417-429.  
60

- 1  
2  
3 Kohn MJ. 1996. Predicting animal  $\delta^{18}\text{O}$ : Accounting for diet and physiological adaptation.  
4 *Geochimica et Cosmochimica Acta* **60**: 4811-4829.
- 5 Kohn MJ. 2010. Carbon isotope compositions of terrestrial  $\text{C}_3$  plants as indicators of  
6 (paleo)ecology and (paleo)climate. *Proceedings of the National Academy of Sciences of*  
7 *the United States of America* **107**: 19691-19695.
- 8 Kohn MJ, Cerling TE. 2002. Stable isotope compositions of biological apatite, In *Phosphates:*  
9 *Geochemical, Geobiological, and Materials Importance*. Kohn, M.J., Rakovan, J.,  
10 Hughes, J.M. (eds.) Mineralogical Society of America: Washington, D.C.; 455-488.
- 11 Kohn MJ, Schoeninger MJ, Valley JW. 1996. Herbivore tooth oxygen isotope compositions:  
12 Effects of diet and physiology. *Geochimica et Cosmochimica Acta* **60**: 3889-3896.
- 13 Kuitens M, Kolfschoten T, Plicht J. 2012. Elevated  $\delta^{15}\text{N}$  values in mammoths: a comparison  
14 with modern elephants. *Archaeological and Anthropological Sciences*: 1-7.
- 15 Laws RM. 1966. Age criteria for the African elephant, *Loxodonta a. africana*. *East African*  
16 *Wildlife Journal* **4**: 1-37.
- 17 Lepper BT, Frolking TA, Fisher DC, Goldstein G, Sanger JE, Wymer DA, Ogden JG, Hooge PE.  
18 1991. Intestinal contents of a Late Pleistocene mastodont from midcontinental North  
19 America. *Quaternary Research* **36**: 120-125.
- 20 Levin NE, Cerling TE, Passey BH, Harris JM, Ehleringer JR. 2006. A stable isotope aridity  
21 index for terrestrial environments. *Proceedings of the National Academy of Sciences of*  
22 *the United States of America* **103**: 11201-11205.
- 23 Long ES, Sweitzer RA, Diefenbach DR, Ben-David M. 2005. Controlling for anthropogenically  
24 induced atmospheric variation in stable carbon isotope studies. *Oecologia* **146**: 148-156.
- 25 Longin R. 1971. New method of collagen extraction for radiocarbon dating. *Nature* **230**: 241-  
26 242.
- 27 Longinelli A. 1984. Oxygen isotopes in mammal bone phosphate: a new tool for  
28 paleohydrological and paleoclimatological research? *Geochimica et Cosmochimica Acta*  
29 **48**: 385-390.
- 30 MacFadden BJ, Cerling TE. 1996. Mammalian herbivore communities, ancient feeding ecology,  
31 and carbon isotopes: A 10 million-year sequence from the Neogene of Florida. *Journal of*  
32 *Vertebrate Paleontology* **16**: 103-115.
- 33 Maschenko E. 2002. Individual development, biology and evolution of the woolly mammoth.  
34 *Cranium* **19**: 4-120.
- 35 Matheus P, Guthrie RD, Kunz ML. 2003a. *Isotope ecology of late Quaternary megafauna in*  
36 *eastern Beringia*. In: 3rd International Mammoth Conference, Dawson City and  
37 Whitehorse, Yukon, pp. 80-83.
- 38 Matheus P, Guthrie RD, Kunz ML. 2003b. *Predator-prey links in Pleistocene east Beringia:*  
39 *evidence from stable isotopes*. In: 3rd International Mammoth Conference, Dawson City  
40 and Whitehorse, Yukon, p. 80.
- 41 Mays S, Ogden A, Montgomery J, Vincent S, Battersby W, Taylor GM. 2011. New light on the  
42 personal identification of a skeleton of a member of Sir John Franklin's last expedition to  
43 the Arctic, 1845. *Journal of Archaeological Science* **38**: 1571-1582.
- 44 McAndrews JH, Jackson LJ. 1988. Age and environment of Late Pleistocene mastodont and  
45 mammoth in southern Ontario, In *Late Pleistocene and Early Holocene Paleoecology*  
46 *and Archeology of the Eastern Great Lakes Region: Proceedings of the Smith*  
47 *Symposium*. Laub, R.S., Miller, N.G., Steadman, D.W. (eds.) Buffalo Society of Natural  
48 Sciences: Buffalo; 161-172.
- 49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 McCue MD, Pollock ED. 2008. Stable isotopes may provide evidence for starvation in reptiles.  
4 *Rapid Communications in Mass Spectrometry* **22**: 2307-2314.
- 5 Mead JI, Agenbroad LD, Davis OK, Martin PS. 1986. Dung of *Mammuthus* in the arid  
6 southwest, North America. *Quaternary Research* **25**: 121-127.
- 7 Meier-Augenstein W, Hobson KA, Wassenaar LI. 2013. Critique: measuring hydrogen stable  
8 isotope abundance of proteins to infer origins of wildlife, food and people. *Bioanalysis* **5**:  
9 751-767.
- 10 Metcalfe JZ, Longstaffe FJ. 2012. Mammoth tooth enamel growth rates inferred from stable  
11 isotope analysis and histology. *Quaternary Research* **77**: 424-432.
- 12 Metcalfe JZ, Longstaffe FJ. 2014. Environmental change and seasonal behavior of mastodons in  
13 the Great Lakes region inferred from stable isotope analysis. *Quaternary Research* **82**:  
14 366-377.
- 15 Metcalfe JZ, Longstaffe FJ, Ballenger JAM, Haynes CV. 2011. Isotopic paleoecology of Clovis  
16 mammoths from Arizona. *Proceedings of the National Academy of Sciences* **108**: 17916-  
17 17920.
- 18 Metcalfe JZ, Longstaffe FJ, Hodgins G. 2013. Proboscideans and paleoenvironments of the  
19 Pleistocene Great Lakes: landscape, vegetation, and stable isotopes. *Quaternary Science*  
20 *Reviews* **76**: 102-113.
- 21 Metcalfe JZ, Longstaffe FJ, Zazula GD. 2010. Nursing, weaning, and tooth development in  
22 woolly mammoths from Old Crow, Yukon, Canada: Implications for Pleistocene  
23 extinctions. *Palaeogeography Palaeoclimatology Palaeoecology* **298**: 257-270.
- 24 Minagawa M, Wada E. 1984. Stepwise enrichment of  $^{15}\text{N}$  along food chains: Further evidence  
25 and the relation between  $\delta^{15}\text{N}$  and animal age. *Geochimica et Cosmochimica Acta* **48**:  
26 1135-1140.
- 27 Murphy BP, Bowman D. 2009. The carbon and nitrogen isotope composition of Australian  
28 grasses in relation to climate. *Functional Ecology* **23**: 1040-1049.
- 29 Newsom LA, Muhlbachler MC. 2006. Mastodons (*Mammot americanum*) diet foraging patterns  
30 based on analysis of dung deposits, In *First Floridians and Last Mastodons: The Page-*  
31 *Ladson site in the Aucilla River*. Webb, S.D. (Ed.) Springer; 263-331.
- 32 O'Brien DM, Wooller MJ. 2007. Tracking human travel using stable oxygen and hydrogen  
33 isotope analyses of hair and urine. *Rapid Communications in Mass Spectrometry* **21**:  
34 2422-2430.
- 35 Olsen KC, White CD, Longstaffe FJ, von Heyking K, McGlynn G, Grupe G, Ruhli FJ. 2014.  
36 Intraskelatal isotopic compositions ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of bone collagen: Nonpathological and  
37 pathological variation. *American Journal of Physical Anthropology* **153**: 598-604.
- 38 Poinar HN, Hofreiter M, Spaulding WG, Martin PS, Stankiewicz BA, Bland H, Evershed RP,  
39 Possnert G, Paabo S. 1998. Molecular coproscopy: Dung and diet of the extinct ground  
40 sloth *Nothrotheriops shastensis*. *Science* **281**: 402-406.
- 41 Polischuk SC, Hobson KA, Ramsay MA. 2001. Use of stable-carbon and -nitrogen isotopes to  
42 assess weaning and fasting in female polar bears and their cubs. *Canadian Journal of*  
43 *Zoology-Revue Canadienne De Zoologie* **79**: 499-511.
- 44 Reitsemma LJ. 2013. Beyond diet reconstruction: Stable isotope applications to human physiology,  
45 health, and nutrition. *American Journal of Human Biology* **25**: 445-456.
- 46 Richards MP, Hedges REM. 2003. Variations in bone collagen  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values of fauna  
47 from Northwest Europe over the last 40 000 years. *Palaeogeography Palaeoclimatology*  
48 *Palaeoecology* **193**: 261-267.
- 49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 Rossmann A, Kornexl B, Versini G, Pichlmayer F, Lamprecht G. 1998. Origin assignment of  
4 milk from alpine regions by multielement stable isotope ratio analysis (SIRA). *Rivista di*  
5 *Scienza dell'Alimentazione* **27**: 9-21.
- 6  
7 Rountrey AN, Fisher DC, Vartanyan S, Fox DL. 2007. Carbon and nitrogen isotope analyses of a  
8 juvenile woolly mammoth tusk: Evidence of weaning. *Quaternary International* **169**:  
9 166-173.
- 10  
11 Rozanski K, Araguas-Araguas L, Gonfiantini R. 1993. Isotopic patterns in modern global  
12 precipitation, In *Climate Change in Continental Isotope Records*. Swart, P.K., McKenzie,  
13 J., Lohmann, K.C., Savin, S. (eds.) American Geophysical Union: Washington, DC; 1-36.
- 14  
15 Saunders JJ. 1996. North American Mammutidae, In *The Proboscidea: evolution and*  
16 *palaeoecology of elephants and their relatives*. Shoshani, J., Tassy, P. (eds.) Oxford  
17 University Press: New York; 271-279.
- 18  
19 Saunders JJ, Grimm EC, Widga CC, Campbell GD, Curry BB, Grimley DA, Hanson PR,  
20 McCullum JP, Oliver JS, Treworgy JD. 2010. Paradigms and proboscideans in the  
21 southern Great Lakes region, USA. *Quaternary International* **217**: 175-187.
- 22  
23 Schmitt J, Schneider R, Elsig J, Leuenberger D, Lourantou A, Chappellaz J, Koehler P, Joos F,  
24 Stocker TF, Leuenberger M, Fischer H. 2012. Carbon isotope constraints on the deglacial  
25 CO<sub>2</sub> rise from ice cores. *Science* **336**: 711-714.
- 26  
27 Schwartz-Narbonne R, Longstaffe FJ, Metcalfe JZ, Zazula G. 2015. Solving the woolly  
28 mammoth conundrum: amino acid <sup>15</sup>N-enrichment suggests a distinct forage or habitat.  
29 *Scientific Reports* **5**.
- 30  
31 Shapiro B, Drummond AJ, Rambaut A, Wilson MC, Matheus PE, Sher AV, Pybus OG, Gilbert  
32 MTP, Barnes I, Binladen J, Willerslev E, Hansen AJ, Baryshnikov GF, Burns JA,  
33 Davydov S, Driver JC, Froese DG, Harington CR, Keddie G, Kosintsev P, Kunz ML,  
34 Martin LD, Stephenson RO, Storer J, Tedford R, Zimov S, Cooper A. 2004. Rise and fall  
35 of the Beringian steppe bison. *Science* **306**: 1561-1565.
- 36  
37 Sharp ZD, Atudorei V, Panarello HO, Fernández J, Douthitt C. 2003. Hydrogen isotope  
38 systematics of hair: archeological and forensic applications. *Journal of Archaeological*  
39 *Science* **30**: 1709-1716.
- 40  
41 Stevens RE, Hedges REM. 2004. Carbon and nitrogen stable isotope analysis of northwest  
42 European horse bone and tooth collagen, 40,000 BP-present: Palaeoclimatic  
43 interpretations. *Quaternary Science Reviews* **23**: 977-991.
- 44  
45 Stewart GR, Turnbull MH, Schmidt S, Erskine PD. 1995. <sup>13</sup>C natural abundance in plant  
46 communities along a rainfall gradient: A biological integrator of water availability  
47 *Australian Journal of Plant Physiology* **22**: 51-55.
- 48  
49 Szpak P. 2014. Complexities of nitrogen isotope biogeochemistry in plant-soil systems:  
50 implications for the study of ancient agricultural and animal management practices.  
51 *Frontiers in Plant Science* **5**: 1-19.
- 52  
53 Szpak P, Grocke DR, Debryne R, MacPhee RDE, Guthrie RD, Froese D, Zazula GD, Patterson  
54 WP, Poinar HN. 2010. Regional differences in bone collagen  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of  
55 Pleistocene mammoths: Implications for paleoecology of the mammoth steppe.  
56 *Palaeogeography Palaeoclimatology Palaeoecology* **286**: 88-96.
- 57  
58 Thewissen JGM, Williams EM, Roe LJ, Hussain ST. 2001. Skeletons of terrestrial cetaceans and  
59 the relationship of whales to artiodactyls. *Nature* **413**: 277-281.
- 60  
61 Tieszen LL. 1991. Natural variations in the carbon isotope values of plants: Implications for  
62 archaeology, ecology, and paleoecology. *Journal of Archaeological Science* **18**: 227-248.

- 1  
2  
3 Toyne JM, White CD, Verano JW, Uceda Castillo S, Millaire JF, Longstaffe FJ. 2014.  
4 Residential histories of elites and sacrificial victims at Huacas de Moche, Peru, as  
5 reconstructed from oxygen isotopes. *Journal of Archaeological Science* **42**: 15-28.  
6  
7 Uno KT, Quade J, Fisher DC, Wittemyer G, Douglas-Hamilton I, Andanje S, Omondi P, Litoroh  
8 M, Cerling TE. 2013. Bomb-curve radiocarbon measurement of recent biologic tissues  
9 and applications to wildlife forensics and stable isotope (paleo)ecology. *Proceedings of*  
10 *the National Academy of Sciences of the United States of America* **110**: 11736-11741.  
11  
12 Van Der Merwe NJ, Medina E. 1991. The canopy effect, carbon isotope ratios and foodwebs in  
13 Amazonia. *Journal of Archaeological Science* **18**: 249-260.  
14  
15 van Geel B, Aptroot A, Baittinger C, Birks HH, Bull ID, Cross HB, Evershed RP, Gravendeel B,  
16 Kompanje EJO, Kuperus P, Mol D, Nierop KGJ, Pals JP, Tikhonov AN, van Reenen G,  
17 van Tienderen PH. 2008. The ecological implications of a Yakutian mammoth's last  
18 meal. *Quaternary Research* **69**: 361-376.  
19  
20 van Klinken GJ. 1999. Bone collagen quality indicators for paleodietary and radiocarbon  
21 measurements. *Journal of Archaeological Science* **26**.  
22  
23 Webb EC, White CD, Longstaffe FJ. 2013. Exploring geographic origins at Cahuachi using  
24 stable isotopic analysis of archaeological human tissues and modern environmental  
25 waters. *International Journal of Osteoarchaeology* **23**: 698-715.  
26  
27 West JB, Bowen GJ, Cerling TE, Ehleringer JR. 2006. Stable isotopes as one of nature's  
28 ecological recorders. *Trends in Ecology & Evolution* **21**: 408-414.  
29  
30 White CD, Spence MW, Stuart-Williams HL, Schwarcz HP. 1998. Oxygen isotopes and the  
31 identification of geographical origins: the Valley of Oaxaca versus the Valley of Mexico.  
32 *Journal of Archaeological Science* **25**: 643-655.  
33  
34 White CD, Storey R, Longstaffe FJ, Spence MW. 2004. Immigration, assimilation, and status in  
35 the ancient city of Teotihuacan: stable isotope evidence from Tlajinga 33. *Latin American*  
36 *Antiquity* **15**: 176-198.  
37  
38 Wilson MC. 1996. Late Quaternary vertebrates and the opening of the ice-free corridor, with  
39 special reference to the genus *Bison*. *Quaternary International* **32**: 97-105.  
40  
41 Yansa CH, Adams KM. 2012. Mastodons and mammoths in the Great Lakes region, USA and  
42 Canada: New insights into their diets as they neared extinction. *Geography Compass* **6**:  
43 175-188.  
44  
45 Zazula GD, MacPhee RDE, Metcalfe JZ, Reyes AV, Brock F, Druckenmiller PS, Groves P,  
46 Harington CR, Hodgins GWL, Kunz ML, Longstaffe FJ, Mann DH, McDonald HG,  
47 Nalawade-Chavan S, Southon JR. 2014. American mastodon extirpation in the Arctic and  
48 Subarctic predates human colonization and terminal Pleistocene climate change.  
49 *Proceedings of the National Academy of Sciences* **111**: 18460-18465.  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

Table 1 Geographic and temporal context and sampling information for specimens used in this study

Specimen	LSIS #	Source	Museum #	Taxon	Area	Site	Tissues	Bone element	Tooth	Radiocarbon		Ref
										Date ( <sup>14</sup> C a BP)	Lab #	
<b>British Columbia</b>												
BC1	RBCM	EH2009.008.0001	Proboscidea	Vanc. Island	Island View Beach	T		tusk				1
BC2	RBCM	EH1994.003.0041	Proboscidea	Vanc. Island	Cordova Bay	T*		tusk				1
BC3-1	RBCM	EH1994.003.0039	Proboscidea	Vanc. Island	Cordova Bay	T		tusk				1
BC4	UWO		Proboscidea	Northern BC		T		tusk				
BC5	RBCM	EH1994.003.0040	Proboscidea	Vanc. Island	Cordova Bay	B		humerus		17,000 ± 240	GSC-2829	1
<b>Alberta</b>												
AB1	RAM	P97.11.1B	<i>Mammuthus</i>	Edmonton	Riverview Pit	B, D, C, E		mandibular symphysis	LLM6	43,300 ± 3000		2
AB2	RAM	P94.1.698	<i>Mammuthus</i>	Edmonton	Consolidated Concrete Pit 48	D, C, E			URM5	>37,700	AA84996	2, 3
AB3	RAM	P94.1.427	Proboscidea	Edmonton	Consolidated Concrete Pit 48	B		left innominate				2, 3
AB4	RAM	P94.16.1B	<i>Mammuthus</i>	Edmonton	Apex Galloway Pit	B, D, E		mandible (ascending ramus)	LRM5			3
AB5	RAM	P90.7.1	<i>Mammuthus</i>	Villeneuve	Alberta Concrete Products Ltd.	D, C, E			ULM6+M7			4
AB6	RAM	P94.4.3	Proboscidea	Villeneuve	Consolidated Concrete Pit 46	D, C, E			LLM6			3
AB7	RAM	P91.11.9	Proboscidea	Villeneuve	Consolidated Concrete Pit 46	B		cortical				3
AB8	RAM	P94.4.1	Proboscidea	Villeneuve	Consolidated Concrete Pit 46	B		right scapula				3
AB9	RAM	P94.4.2	<i>Mammuthus</i>	Villeneuve	Consolidated Concrete Pit 46	C*			URM6			3
AB10	RAM	P97.7.1	<i>Mammuthus</i>	Villeneuve	Consolidated Concrete Pit 46	D, E			LLM6	>41,100	AA84997	3
AB11	RAM	P00.2.1	<i>Mammuthus</i>	Bezanson	Smoky River	D, C, E			LRM4/M5	40,000 ± 3500	AA84979	
AB12	RAM	P96.12.1	Proboscidea	Cochrane	Burnco Pit	B		humerus (deltoid tuberosity)		10,743 ± 100	BGS 2141	5
<b>Yukon</b>												
YT1	YG	291.1	<i>Mammuthus</i>	Old Crow	Old Crow River	RD, E			LRM6	>41,100	AA84987	6
YT2	YG	122.2	<i>Mammuthus</i>	Old Crow	CRH 94	RD, E			ULM6	>41,100	AA84992	6
YT3	YG	173.5	<i>Mammuthus</i>	Old Crow	Ch'jee's Bluff	D, E			U M6			6
YT4	YG	285.1	<i>Mammuthus</i>	Old Crow	OCR, REM 78-1	B, E			LRM6	>39,100	AA85002	6
YT5	YG	60.2	<i>Mammuthus</i>	Old Crow	OCR, Bluffs-R bank	RD, E			LRM5/M6			6
YT6	YG	57.1	<i>Mammuthus</i>	Old Crow	OCR, Bluffs-R bank	C, E			LRM6			6
YT7	YG	325.22	<i>Mammuthus</i>	Old Crow	HH-68-10	B, E			ULM6	>40,100	AA84984	6
YT8	YG	357.1	<i>Mammuthus</i>	Old Crow	Old Crow River	D, E			M5/M6	>41,100	AA84995	6
YT9	YG	284.4	<i>Mammuthus</i>	Old Crow	CRH 11	C, E			ULM6			6
YT10	YG	252.2	<i>Mammuthus</i>	Old Crow	CRH 20	RD, E			LLM6	>40,000	AA85001	6
YT11	YG	173.1	<i>Mammuthus</i>	Old Crow	Ch'jee's Bluff	RD, C, E			U M6			6
YT24	YG	78.1	<i>Mammuthus</i>	Klondike	Last Chance Creek	RD, E			M6	>48,800	UCIAMS41491	7
YT25	YG	2.16	<i>Mammuthus</i>	Klondike	Hester Creek	C, E						
YT26	YG	2.14	<i>Mammuthus</i>	Klondike	Hester Creek	C, E		adult		35,500 ± 2000	AA84986	
YT27	YG	29.248	<i>Mammuthus</i>	Klondike	Hunker Creek	B		mandible		43,500 ± 1900	UCIAMS41493	7
YT28a,b	YG	6.51, 6.49	<i>Mammuthus</i>	Klondike	Gold Run Creek	B, E		mandible	LLM6,LRM6	17,950 ± 120	Beta 70099	
YT29	YG	358.1	<i>Mammuthus</i>	Klondike		B, E		mandible	M6			
YT30	YG	302.26	<i>Mammuthus</i>	Klondike	Skookum Gulch	B, E		mandible	M5/M6	>39,200	AA84983	
YT31	YG	2.4	<i>Mammuthus</i>	Klondike	Hester Creek	B, E		mandible	M6?	27,380 ± 730	AA85000	
YT32	YG	84.1	<i>Mammuthus</i>	Klondike	Hunker Creek	C, E		adult				
YT33	YG	159.83	<i>Mammuthus</i>	Klondike	Quartz Creek	B		fibula				
YT34	YG	43.2	<i>Mammuthus</i>	Klondike	Upper Gold Run	RD, E			M6	>49,200	UCIAMS75320	8
YT35	YG	26.1	<i>Mammuthus</i>	Klondike	Gold Run Creek	RD, E			M6	>50,300; >51,700	UCIAMS 78705, 78704	8
YT36	YG	50.1	<i>Mammuthus</i>	Klondike	Thistle Creek	RD, E			M5?	>41,100	AA84994	
YT37	YG	139.5	<i>Mammuthus</i>	Klondike	Unknown?	B, E		mandible	M6	>41,100	AA84985	
YT38	YG	33.3	<i>Mammuthus</i>	Herschel	Simpson Point	B		cuboid		>45 130	Beta 189291	9
YT39	YG	68.2	<i>Mammuthus</i>	Herschel	Simpson Point	D, E			fragment			
YT40	YG	68.4	<i>Mammuthus</i>	Herschel	Simpson Point	C, E			fragment	>35,900	AA84988	
YT41	YG	154.2	Proboscidea	Herschel	South Shore	T		tusk				
YT42	YG	12.9	Proboscidea	Herschel	Simpson Point	T		tusk		34,100 ± 1,700	AA84999	
YT43	YG	206.29	Proboscidea	Herschel	Simpson Point	T		tusk				
YT44	YG	33.7	Proboscidea	Herschel	Simpson Point	T		tusk				
YT45	YG	68.1	Proboscidea	Herschel	Simpson Point	T		tusk		>39,400	AA84982	
YT46	YG	271.93	<i>Mammuthus</i>	Herschel	Simpson Point	B		thoracic vertebra				
YT47	YG	2.7	<i>Mammuthus</i>	Klondike	Hester Creek	B		fibula		27,540 ± 270	UCIAMS 41488	7
YT48	YG	133.21	<i>Mammuthus</i>	Klondike	Whitman Gulch	B		right tibia		34,180 ± 590	UCIAMS41489	7
YT49	YG	5.46	<i>Mammuthus</i>	Klondike	Hunker Creek	B		rib		22,430 ± 140	UCIAMS41487	7
YT50	YG	219.3	<i>Mammuthus</i>	Stewart River	Ash Bend	B		long bone fragments				
YT51	YG	317.51	<i>Mammuthus</i>	Old Crow	CRH 11	T				MIS5, ~140,000		

LSIS #: Sample ID used for Laboratory for Stable Isotope Science analyses, and referred to in this study;

Source of skeletal materials: RBCM = Royal British Columbia Museum, RAM = Royal Alberta Museum, YG = Yukon Government, CMN = Canadian Museum of Nature

Tissues: T = tusk, B = bone, D = dentin (crown), RD = root dentin, C = cementum, E = enamel

Radiocarbon dates in bold were obtained as part of the present study

\* Insufficient collagen preservation; no data

#### References

- Keddie, 1979
- Burns and Young, 1994
- Young et al., 1994
- Burns et al., 2003
- Burns, 2010
- Metcalfe et al., 2010
- Debruyne et al., 2008
- Zazula et al., 2014
- Zazula et al., 2009



Table 2. The isotopic compositions of collagen (various tissues) and structural carbonate in bioapatite (enamel) for mammoths, mastodons, and unknown proboscideans analyzed in this study and by Zazula et al. (2014).

Region	LSIS #	Collagen								Structural carbonate			
		Tissue used	C (%)	$\delta^{13}\text{C}$ (‰)	N (%)	$\delta^{15}\text{N}$ (‰)	C/N (Atomic)	Yield (%)	Lab*	Tissue used	$\delta^{13}\text{C}$ (‰)	$\delta^{18}\text{O}$ (‰)	CO <sub>3</sub> (%)
<b>British Columbia</b>													
	<i>Proboscidea</i>												
	BC1	T	38.4	-19.9	14.2	+7.0	3.2	4.3	LSIS				
	BC3-1	T	42.4	-20.7	15.8	+6.9	3.1	6.3	LSIS				
	BC4	T	47.0	-21.3	17.4	+6.9	3.1	22.9	LSIS				
	BC5	B	41.3	-21.3	15.2	+5.9	3.2	11.0	LSIS				
	Mean		42.3	-20.8	15.7	+6.7	3.1	11.1					
	SD		3.6	0.7	1.3	0.5	0.0	8.3					
<b>Alberta</b>													
	<i>Proboscidea</i>												
	AB3	B	<b>42.0</b>	<b>-20.5</b>	<b>15.8</b>	<b>+10.9</b>	<b>3.1</b>	11.0	LSIS				
	AB7	B	41.5	-20.4	16.3	+9.0	3.0	10.9	LSIS				
	AB8	B	<b>39.9</b>	<b>-19.7</b>	<b>14.8</b>	<b>+4.4</b>	<b>3.1</b>	7.5	LSIS				
	AB12	B	32.8	-21.0	12.1	+1.4	3.2	4.3	LSIS				
	Mean		39.0	-20.4	14.8	+6.4	3.1	8.5					
	SD		4.3	0.5	1.8	4.3	0.1	3.2					
	<i>Mammuthus</i>												
	AB1	B,C,D	<b>40.9</b>	<b>-20.6</b>	<b>15.6</b>	<b>+7.8</b>	<b>3.1</b>	<b>&gt;5.9 to 16.0</b>	LSIS	E	<b>-10.7</b>	<b>+14.2</b>	<b>4.0</b>
	AB2	C, D	<b>36.1</b>	<b>-21.1</b>	<b>13.3</b>	<b>+9.7</b>	<b>3.2</b>	14.4 to >15.5	AMS/LSIS	E	<b>-11.4</b>	<b>+15.0</b>	<b>6.8</b>
	AB5	C, D	<b>36.1</b>	<b>-21.4</b>	<b>13.0</b>	<b>+10.0</b>	<b>3.2</b>	<b>5.3</b>	LSIS	E	-11.5	+13.6	4.9
	AB6	D	28.0	-20.1	10.2	+5.1	3.2	>14.7	LSIS	E	-10.7	+15.2	5.6
	AB11	C, D	<b>34.9</b>	<b>-20.6</b>	<b>13.0</b>	<b>+5.5</b>	<b>3.1</b>	<b>&gt;4.3 to 16.1</b>	AMS/LSIS	E	-10.5	+13.1	5.9
	Mean		35.2	-20.8	13.0	+7.6	3.2				-10.9	+14.2	5.4
	SD		4.6	0.5	1.9	2.3	0.1				0.5	0.9	1.1
	<i>Mammuth</i>												
	AB4	B, D	<b>40.3</b>	<b>-20.8</b>	<b>15.0</b>	<b>+4.5</b>	<b>3.1</b>	<b>&gt;11.9 to 13.6</b>	LSIS	E	-11.6	+14.9	3.6
	AB10	D	41.1	-19.5	15.4	+5.0	3.1	12.4	AMS	E	-10.3	+13.5	4.0
	Mean		40.7	-20.1	15.2	+4.7	3.1				-10.9	+14.2	3.8

1													
2													
3													
4		SD	0.6	0.9	0.3	0.4	0.0			0.9	1.0	0.3	
5	<b>Yukon - Klondike</b>												
6	<i>Mammuthus</i>												
7	YT24	RD	45.7	-20.8	16.9	+8.2	3.2	20.2	LSIS	E	-11.7	+10.1	5.4
8	YT25	C	42.0	-21.2	15.5	+8.0	3.2	7.3	LSIS	E	-11.8	+11.4	2.8
9	YT26	C	43.3	-20.5	16.0	+7.4	3.2	15.3	AMS	E	<b>-12.5</b>	<b>+12.3</b>	<b>5.3</b>
10	YT27	B	<b>44.4</b>	<b>-21.1</b>	<b>16.3</b>	<b>+7.0</b>	<b>3.2</b>	13.5	LSIS				
11	YT28	B	47.5	-20.7	18.0	+10.5	3.1	19.1	LSIS	<b>E</b>	<b>-11.5</b>	<b>+9.9</b>	<b>5.6</b>
12	YT29	B	42.0	-20.6	15.2	+7.2	3.2	12.7	LSIS	E	-11.47	+12.0	5.1
13	YT30	B	41.5	-20.3	15.6	+5.8	3.1	18.0	AMS	E	-12.1	+10.5	5.1
14	YT31	B	37.8	-20.5	14.1	+7.0	3.1	13.0	AMS				
15	YT32	C	45.9	-20.7	16.9	+7.7	3.2	12.4	LSIS	E	-11.1	+11.7	5.0
16	YT47	B	<b>46.8</b>	<b>-20.6</b>	<b>17.6</b>	<b>+6.5</b>	<b>3.1</b>	<b>16.8</b>	LSIS				
17	YT48	B	46.6	-20.7	17.3	+6.8	3.1	21.0	LSIS				
18	YT49	B	45.0	-21.3	16.3	+8.7	3.2	20.6	LSIS				
19	YT50	B	35.9	-20.8	12.9	+7.2	3.3	1.2	LSIS				
20	Mean		43.4	-20.7	16.0	+7.5	3.2	14.7			-11.7	+11.1	4.9
21	SD		3.5	0.3	1.4	1.2	0.0	5.7			0.4	0.9	1.0
22	<i>Mammut</i>												
23	YT33	B	43.2	-20.9	15.8	+6.9	3.2	18.5	LSIS				
24	YT34	RD	45.7	-20.0	16.9	+4.5	3.2	18.4	LSIS	E	-11.34	+10.8	4.9
25	YT35	RD	42.4	-20.6	16.0	+4.4	3.1	19.9	AMS	E	-11.3	+10.4	4.0
26	YT36	RD	41.7	-21.2	15.6	+4.3	3.1	12.6	AMS	E	-11.3	+10.9	4.4
27	YT37	B	42.0	-20.6	15.0	+3.5	3.3	14.5	AMS	E	-10.6	+11.4	3.7
28													
29	<sup>1</sup> CMN333	D/RD	<b>43.6</b>	<b>-20.6</b>	<b>15.4</b>	<b>+4.1</b>	<b>3.3</b>	<b>6.1</b>	UCI				
30	<sup>1</sup> CMN33897	D/RD	42.7	-19.9	15.7	+4.9	3.2	7.6	UCI				
31	<sup>1</sup> CMN 8707	D/RD	42.9	-20.8	15.6	+4.3	3.2	8.4	UCI				
32	<sup>1</sup> CMN 11697	D/RD	42.2	-20.5	15.4	+4.6	3.2	8.5	UCI				
33	<sup>1</sup> CMN 42551	D/RD	42.2	-20.1	15.1	+3.0	3.4	4.0	UCI				
34	<sup>1</sup> CMN 42552	D/RD	39.1	-19.9	14.0	0.0	3.3	0.6	UCI				
35	<sup>1</sup> F:AM: 104842	?	43.5	-21.4	15.3	+3.6	3.3	4.1	UCI				
36	<sup>1</sup> YG 361.9	?	41	-20.7	14.5	+3.2	3.3	8.9	UCI				
37													
38	Mean		42.5	-20.6	15.4	4.2	3.2	10.2			-11.1	+10.9	4.2
39	SD		1.5	0.5	0.7	1.0	0.1	6.2			0.4	0.4	0.5
40	<b>Yukon- Old Crow</b>												
41	<i>Mammuthus</i>												
42													
43													
44													
45													
46													
47													
48													
49													

1													
2													
3													
4													
5													
6													
7													
8													
9													
10													
11													
12													
13													
14													
15													
16													
17													
18													
19													
20													
21													
22													
23													
24													
25													
26													
27													
28													
29													
30													
31													
32													
33													
34													
35													
36													
37													
38													
39													
40													
41													
42													
43													
44													
45													
46													
47													
48													
49													

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49

Values in **bold** are the means of duplicate analyses or analyses of different tissues from the same individual (see Supplementary Table S1).  
Tissue: RD = root dentin, D = crown dentin, T = tusk dentin, B = bone, C = cementum, E = enamel  
\* Lab in which collagen extraction was performed: LSIS (University of Western Ontario), AMS (University of Arizona), UCI (University of California, Irvine)  
1 Data are from Zazula et al. (2014). Yields are >30kDa fraction of collagen.

Table 3. Isotopic results for collagen extracted from different areas on AB5 / P90.7.1 (mammoth M6 and M7), in order of inferred development time (top is earliest, bottom is latest).

LSIS #	Tooth	Location	Tissue	Collagen						
				C (%)	$\delta^{13}\text{C}$ (‰)	N (%)	$\delta^{15}\text{N}$ (‰)	C/N (Atomic)	Yield (%)	Lab*
AB5	M6	anterior crown	D	29.2	-21.6	10.6	+10.2	3.2	3.6	LSIS
AB5	M6	anterior crown	C	41.9	-21.4	15.3	+10.4	3.2	8.6	LSIS
AB5	M6	root	D	36.5	-21.2	13.2	+10.5	3.2	4.4	LSIS
AB5	M7	crown	C	37.0	-21.2	13.4	+9.0	3.2	4.7	LSIS
AB5	M6-M7	between the 2 teeth	C				<i>not preserved</i>			
AB5	M7	root	D				<i>not preserved</i>			
<i>AB5 mean</i>	<i>M6, M7</i>	<i>all</i>	<i>C, D</i>	<i>36.1</i>	<i>-21.4</i>	<i>13.1</i>	<i>+10.0</i>	<i>3.2</i>	<i>5.3</i>	<i>LSIS</i>

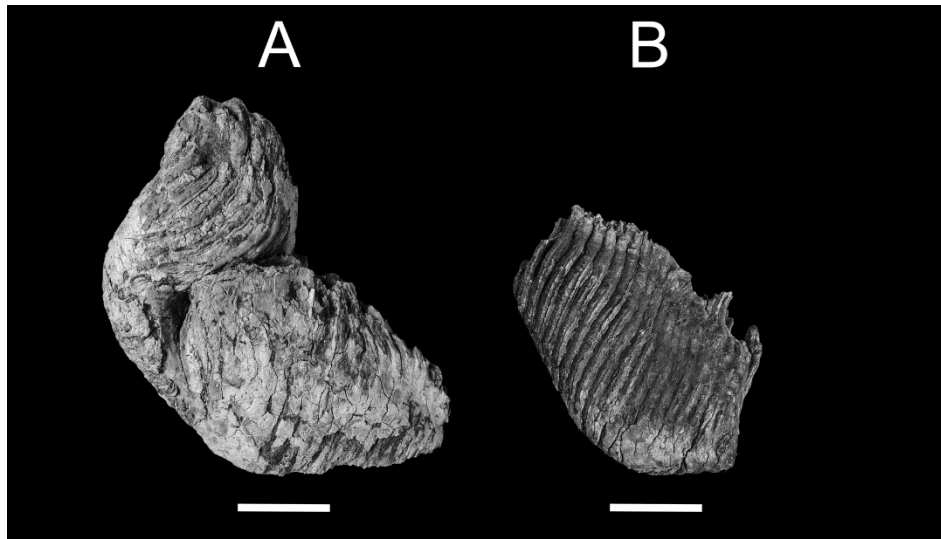
Tissue: D = dentin, B = bone, C = cementum

\* Laboratory in which collagen extraction was performed: LSIS (University of Western Ontario)

1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43

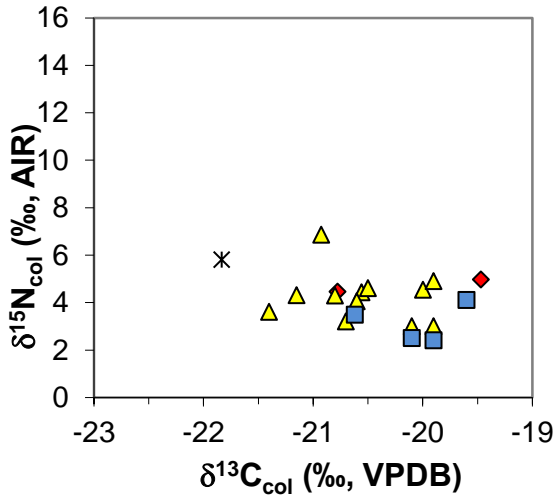


1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43

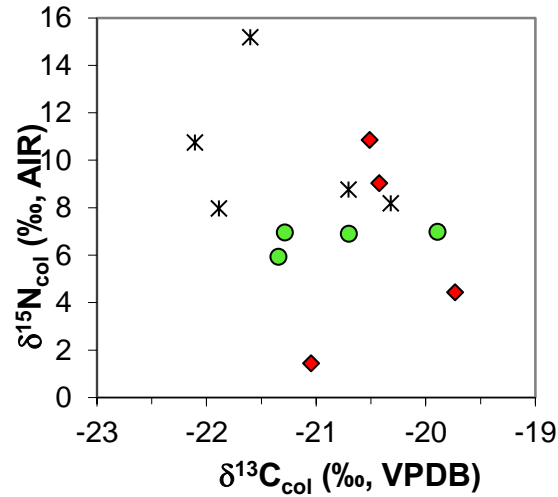


1  
2  
3  
4  
5  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43

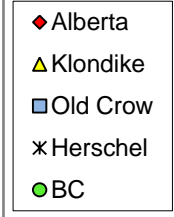
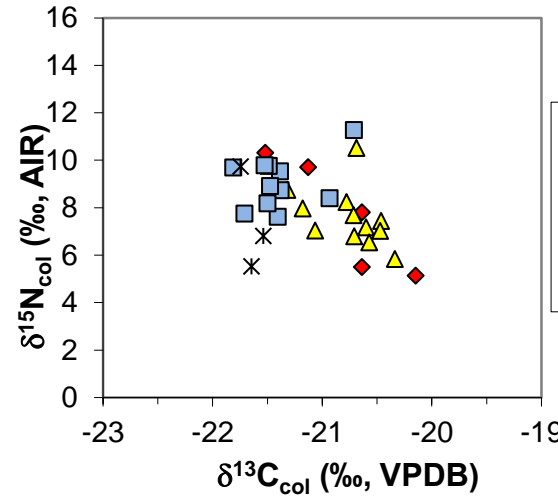
a. Mastodons



b. Unknown proboscideans



c. Mammoths





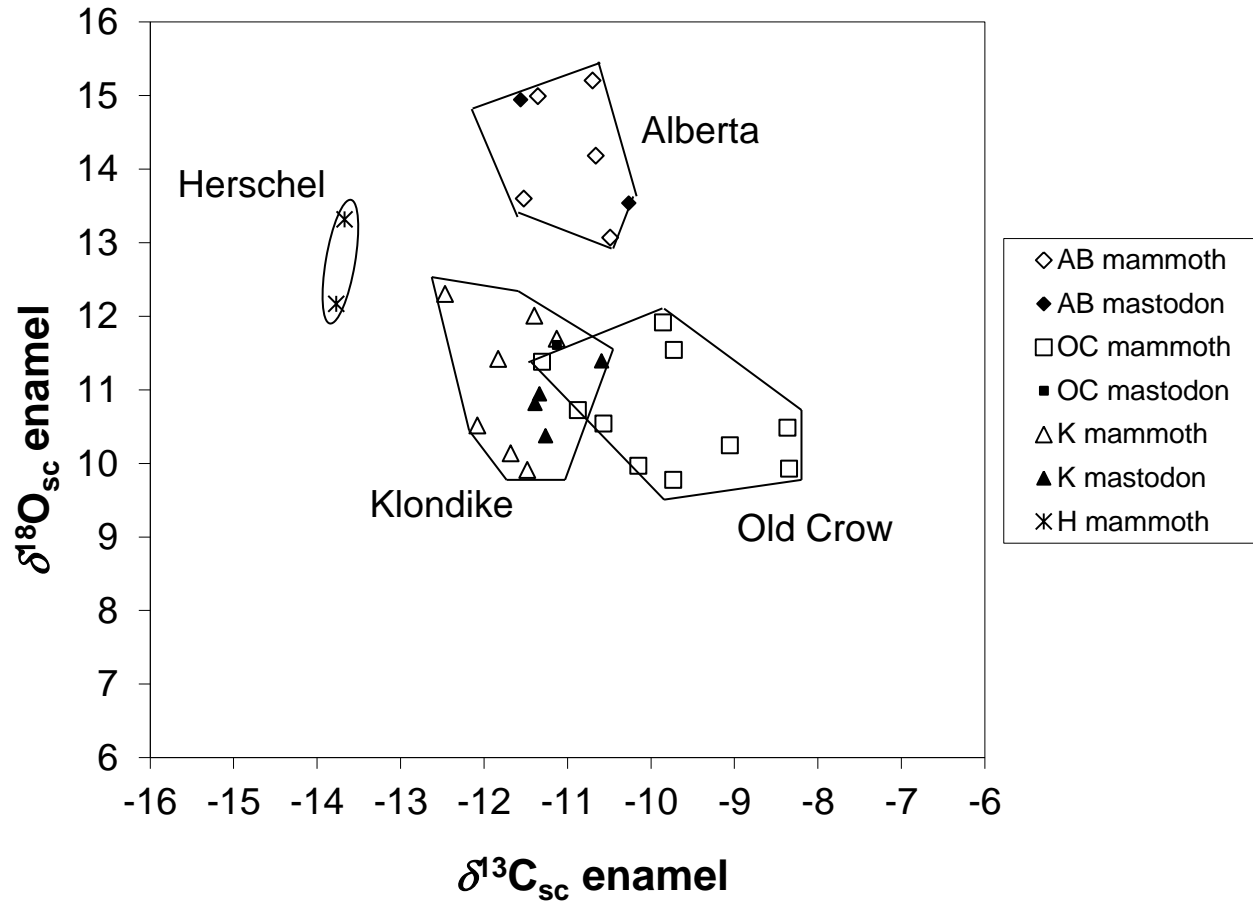


Table S1. Isotopic results for collagen extracted from different tissues within the same individuals (excluding AB5).

LSIS #	Tissue	Collagen					Yield (%)	Lab*
		C (%)	$\delta^{13}\text{C}$ (‰)	N (%)	$\delta^{15}\text{N}$ (‰)	C/N (Atomic)		
AB1	B	44.4	-20.8	16.7	+7.3	3.1	16.0	LSIS
AB1	C	39.7	-20.6	14.9	+8.0	3.1	>11.0	LSIS
AB1	D	38.6	-20.5	15.1	+8.2	3.0	>5.9	LSIS
AB1 mean	B,C,D	40.9	-20.6	15.6	+7.8	3.1	>5.9 to 16.0	LSIS
AB2	C	42.1	-21.1	15.6	+9.9	3.1	14.4	AMS
AB2	D	30.1	-21.1	10.9	+9.5	3.2	>15.5	LSIS
AB2 mean	C, D	36.1	-21.1	13.3	+9.7	3.2	14.4 to >15.5	AMS&LSIS
AB4	B	43.5	-20.9	16.3	+4.1	3.1	13.6	LSIS
AB4	D	37.1	-20.7	13.8	+4.8	3.1	>11.9	LSIS
AB4 mean	B, D	40.3	-20.8	15.0	+4.5	3.1	>11.9 to 13.6	LSIS
AB11	C	43.8	-20.3	16.3	+5.9	3.1	16.1	AMS
AB11	D	25.9	-21.0	9.7	+5.1	3.1	>4.3	LSIS
AB11 mean	C, D	34.9	-20.6	13.0	+5.5	3.1	>4.3 to 16.1	AMS & LSIS
YT 11	C	44.1	-21.5	15.9	+8.3	3.2	18.2	LSIS
YT 11	RD	47.2	-21.5	17.6	+9.5	3.1	14.3	LSIS
YT 11 mean	C, RD	45.6	-21.5	16.7	+8.9	3.2	16.3	LSIS

## Appendix S1. Developmental timing of the AB5 (P90.7.1) mammoth M6/M7

Interpretation of the isotopic results for the AB5 (P90.7.1) mammoth depends in part on knowledge of mammoth tooth development, which is not yet complete. Although growth rates of proboscidean molar crowns and the timing of eruption and wear have been estimated (Dirks et al., 2012; Haynes, 1991; Laws, 1966; Maschenko, 2002; Metcalfe and Longstaffe, 2012; Metcalfe et al., 2010; Uno et al., 2013), we lack complete knowledge of (1) the time required for a full M6 or M7 to develop, (2) the relative timing (i.e., degree of overlap) of the development of these teeth, and (3) the amount of time that elapsed between the development of the M6/M7 and the AB5 (P90.7.1) mammoth's death. These issues are explained more fully below, and inferences about the developmental timing of the AB5 (P90.7.1) specimen are made based on our best estimates for the timing of mammoth tooth development and the age-at-death of the AB5 (P90.7.1) mammoth.

In proboscidean teeth, lamellae (plates) are formed in sequence from anterior to posterior. Anterior crown dentin forms first, followed by anterior enamel and cementum (Hillson, 2005; Maschenko, 2002). Root formation begins after the lamellae of the crown are present, but before the cementum is completely formed (Maschenko, 2002). In general, as one tooth is in wear another is developing posterior to it, but there can be overlap in the development time of successive teeth (Laws, 1966). Growth of mammoth and modern elephant teeth crowns in the occlusal-basal direction occurs at a rate of 13 to 22 mm/a (Dirks et al., 2012; Metcalfe and Longstaffe, 2012; Uno et al., 2013). Dirks et al. (2012) estimated that in *Mammuthus columbi* it took about 10.6 years for an enamel plate of 180.9 mm height to develop, but this does not necessarily include the full mineralization time, and is an estimate for only one lamella. The full

1  
2  
3 development time for a single tooth also depends on the number of lamellae present, the height  
4  
5 of each lamella, and the developmental timing of the crown dentin, roots, and cementum, which  
6  
7 remains unknown. Since the anterior of an M6 is in wear while the posterior is still developing, it  
8  
9 is not possible to physically measure the “full” tooth at any given time. The rate of development  
10  
11 of the M7, which may differ from that of the M6, also remains unknown. Based on the general  
12  
13 development of proboscidean teeth, we can infer the following relative timing of development  
14  
15 for our subsamples: (1) anterior M6 dentin, (2) anterior M6 cementum, (3) M6 root dentin, (4)  
16  
17 M7 crown cementum, (5) cementum between the M6 and M7, (6) M7 root dentin. The potential  
18  
19 for overlap in the developmental timing of the M6 and M7 is discussed below.  
20  
21  
22  
23

24 We lack any general knowledge of the developmental timing of M7 relative to that of  
25  
26 M6, since the former is not normally present in elephant dentition. Based on the development  
27  
28 and wear of elephant and mammoth teeth (Haynes, 1991; Laws, 1966), full crown formation of a  
29  
30 *Mammuthus* M6 likely occurred occur over a period of about 17 years, between ages 26 and 43  
31  
32 (Metcalf et al., 2010). Burns et al. (2003) estimated the AB5 mammoth to be about 47-48 years  
33  
34 old at the time of death. If these inferences are correct, then only about 4-5 years elapsed  
35  
36 between complete formation of the M6 crown and the animal’s death. Burns et al. (2003:82)  
37  
38 infer that the M7 “was undoubtedly deformed from a very early stage of its development because  
39  
40 it is so thoroughly distorted... virtually all lamellae were disengaged from their neighbours and  
41  
42 later cemented at odd angles.” They further argue that “the M6 was fully erupted when the M7  
43  
44 was forced into place, but that the M6 functioned for some years before the M7 came to rest”  
45  
46 (Burns et al., 2003:82). Thus, the morphological evidence indicates that deformation of the M7  
47  
48 occurred after formation of the M6 was complete. However, the M7 lamellae may have begun to  
49  
50 form either before or after the posterior/basal portions of the M6 were still developing. As  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

1  
2  
3 described in the main text, the isotopic compositions of the teeth can provide additional insight  
4  
5 into this problem. Similar isotopic compositions for the two teeth would support the idea that  
6  
7 there was significant overlap in their development times, whereas different isotopic compositions  
8  
9 would suggest that they developed largely at different times. The latter proved to be the case:  
10  
11 different  $\delta^{15}\text{N}_{\text{col}}$  values of the M7 crown cementum and the M6 tissues (including the M6 root,  
12  
13 the last part of the M6 to develop) support the hypothesis that the M6 was fully formed when the  
14  
15 M7 began to develop. Thus, if all of the above inferences are correct, the isotopic data we  
16  
17 obtained for the M7 thus represents some portion of the last 4-5 years of the animal's life,  
18  
19 whereas the M6 developed earlier, well in advance of the animal's death. Unfortunately, the M7  
20  
21 root and the cementum linking the two teeth, which likely represented the time closest to death,  
22  
23 did not contain well-preserved collagen (see main text). Crown cementum develops after crown  
24  
25 enamel and dentin, so the M7 crown cementum isotopic results could still represent a period of  
26  
27 time close to death. It is currently not possible to determine whether development of the M7  
28  
29 crown cementum would overlap the period of impaired chewing and acute nutritional stress  
30  
31 represented by the secondary wear facet on the M7, which apparently developed over a  
32  
33 "relatively short time" (Burns et al., 2003: 83).  
34  
35  
36  
37  
38  
39  
40  
41  
42

## 43 References

- 44  
45 Burns, J.A., G., B.C., Mol, D., 2003. An extraordinary woolly mammoth molar from Alberta,  
46 Canada. *Deinsea*, 9: 77-85.  
47  
48 Dirks, W., Bromage, T.G., Agenbroad, L.D., 2012. The duration and rate of molar plate  
49 formation in *Palaeoloxodon cypriotes* and *Mammuthus columbi* from dental histology.  
50 *Quaternary International*, 255: 79-85.  
51  
52 Haynes, G., 1991. *Mammoths, mastodonts, and elephants: biology, behavior, and the fossil*  
53 *record*. Cambridge University Press, Cambridge New York, 413 pp.  
54  
55 Hillson, S., 2005. *Teeth*, Cambridge manuals in archaeology. Cambridge University Press, New  
56 York.  
57  
58 Laws, R.M., 1966. Age criteria for the African elephant, *Loxodonta a. africana*. *East African*  
59 *Wildlife Journal*, 4: 1-37.  
60

- 1  
2  
3 Maschenko, E., 2002. Individual development, biology and evolution of the woolly mammoth.  
4 Cranium, 19(1): 4-120.  
5 Metcalfe, J.Z., Longstaffe, F.J., 2012. Mammoth tooth enamel growth rates inferred from stable  
6 isotope analysis and histology. Quaternary Research, 77(3): 424-432.  
7 Metcalfe, J.Z., Longstaffe, F.J., Zazula, G.D., 2010. Nursing, weaning, and tooth development in  
8 woolly mammoths from Old Crow, Yukon, Canada: Implications for Pleistocene  
9 extinctions. Palaeogeography Palaeoclimatology Palaeoecology, 298: 257-270.  
10 Uno, K.T. et al., 2013. Bomb-curve radiocarbon measurement of recent biologic tissues and  
11 applications to wildlife forensics and stable isotope (paleo)ecology. Proceedings of the  
12 National Academy of Sciences of the United States of America, 110(29): 11736-11741.  
13  
14  
15  
16  
17  
18  
19  
20  
21  
22  
23  
24  
25  
26  
27  
28  
29  
30  
31  
32  
33  
34  
35  
36  
37  
38  
39  
40  
41  
42  
43  
44  
45  
46  
47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60