**Supplemental Materials and Methods**

*Dung Preparation and Identification*

Sixteen bison dung samples from Promontory Cave 1 and one from Promontory Cave 2 were identified based on size and morphology (Kropf et al. 2007; Mead and Agenbroad 1989; Mead et al. 1986). Six modern bison dung samples were collected from Antelope Island State Park and freeze-dried prior to processing. All dung samples were ground into a fine powder with a mortar and pestle prior to isotopic analysis.

*Hair Preparation*

Thirty-three bison hairs from Promontory Cave 1 and two from Franktown Cave were examined using a light microscope to confirm that they were bison (Metcalfe 2018). Most were coarse guard/overhairs or tail/limb-type hairs, but a few underhairs were also sampled (Table 1). Eight samples of modern bison hair from Antelope Island (southeast side of Great Salt Lake) were collected with the permission of Steven Bates (Wildlife Biologist, Antelope Island State Park). All hairs were sonicated with 2:1 chloroform:methanol until the solution remained clear, then air-dried for a minimum of 48 hours prior to analysis. All whole-hair samples had carbon contents of 40 - 46 %, nitrogen contents of 13 - 16 ‰, and atomic C/N ratios of 3.4 - 3.7 (Table 1). There are no widely-accepted ranges for these quality-control indicators particular to bison hair, but these values are well-constrained and within the ranges reported for human hair (O'Connell and Hedges 1999a, 1999b) and bone collagen (Ambrose 1990; van Klinken 1999). Recently, Szpak and Valenzuela (2020) inferred that camelid hairs with C/N ratios between 3.0 to 4.0 had reasonably well-preserved carbon and nitrogen isotope compositions.

Eleven coarse Promontory bison hairs were selected for serial sampling. These hairs ranged in length from about 7 - 27 cm. Each single hair was cut into segments approximately 1 - 3 cm long, depending on the length required to reach a target weight of 0.3 – 0.4 mg. Each segment was analyzed individually, resulting in a record of changes over time during the growth of the hair. A total of 88 serially-sampled segments were analyzed (Supplemental Table 1). All of these hairs had %C, %N, and C/N ratios within the range stated above, except for a few terminal (tip) sections that had somewhat higher C/N ratios (3.9 – 4.1). Importantly, these samples also had lower sample weights than the other hairs in the series. Because of these low weights (≤0.3 mg), their nitrogen peaks were low and their %N and C/N ratios were therefore not as accurately measured. On the other hand, the carbon peaks for these samples remained large enough for accurate analysis, and their δ13C values were consistent with expectations based on the preceding values in the serially-sampled sequence. These samples have therefore been retained for consideration.

*Hide Preparation*

Tiny (~ 5 mg) hide/leather samples (n=19 from Promontory Cave 1, n=6 from Franktown Cave) were sonicated with 2:1 chloroform:methanol until the solution remained clear. After isotopic analysis, some of these samples had higher C/N ratios than would be expected for skin, which is primarily collagen. These and subsequent samples were then submitted to an acid-base-acid treatment (0.5 M HCl and 0.1 M NaOH) adapted from standard pretreatments for radiocarbon dating (Brock et al. 2010). All hide specimens were examined with a microscope to ensure they were free of macroscopic contamination and visible hair inclusions prior to analysis. The clean hide samples had carbon contents of 24 - 44%, nitrogen contents of 8 - 16%, and C/N ratios of 3.2 - 3.5 (Table 1). To our knowledge there are no accepted quality-control values for animal skin/hide, but the values measured here are similar to those of well-preserved bone collagen. Given that skin protein, like bone protein, is primarily Type I collagen, this suggests good sample preservation.

*Bone Collagen Preparation*

Small chunks of dense cortical bone were cut from 12 Promontory Cave 1 and 2 specimens in locations that would minimize damage. Collagen was extracted from bones using a modified Longin (1971) method. Small chunks of cortical bone (n=12) were sonicated with 2:1 chloroform:methanol until the solution remained clear and air dried. Samples were then demineralized in 0.5 M HCl, rinsed to neutrality, treated with 0.1 M NaOH until the solution remained clear, rinsed to neutrality, and gelatinized at 75 ºC in pH 3 HCl. Collagen carbon contents were 38 - 43%, nitrogen contents were 14 - 15%, and atomic C/N ratios were 3.2 - 3.4, indicating excellent preservation and no obvious contamination (Ambrose 1990; van Klinken 1999).

*Carbon Isotope Analysis*

Carbon isotope compositions (*δ*13C) and carbon and nitrogen contents (%C, %N) were measured using an Elementar VarioMicro Cube elemental analyzer coupled with an Isoprime stable isotope ratio mass spectrometer in continuous-flow mode. *δ*13C values were calibrated to the VPDB scale using USGS-40 and USGS-41 or 41a (accepted values –26.39, +37.63, and +36.55 ‰, respectively). Internal check standards (gelatin, methionine, red lentil, amaranth; accepted values –15.30, –28.60, –26.12, –13.59 ‰, respectively) and sample replicates (minimum 10% of samples in each run) were used to monitor measurement uncertainty. For bone collagen, hair, hide, and dung (respectively), precision (*u(Rw))* wasdetermined to be ±0.17 ‰, ±0.10 ‰, ±0.19 ‰, and ±0.41‰; accuracy (*u(bias)*) was ±0.14 ‰, ±0.15 ‰, ±0.15 ‰, and ±0.12 ‰; and the total analytical uncertainty (*uc*) was estimated to be ±0.22 ‰, ±0.18 ‰, ±0.24 ‰, and ±0.43 ‰ (Supplemental Table 2) (Szpak et al. 2017). Carbon and nitrogen contents (weight %) were calculated based on the amplitude of the major carbon and nitrogen peaks relative to the weight of the sample, calibrated using USGS-40.

*Ancient DNA Analysis*

We analyzed the FS-305 moccasin ankle wrap specimen at the UCSC Paleogenomics ancient DNA laboratory (PGL) to (1) obtain a taxonomic identification, and (2) determine the sex of the animal. Working in the sterile laboratory facilities at the PGL, we washed the sample in ultra-pure water to remove soil from the exterior and then extracted DNA from 0.15g of tissue following the Dabney et al. (2013) tissue extraction protocol. We prepared two shotgun sequencing libraries following the Meyer and Kircher (2010) method. We labelled the libraries using dual indices with truseq sequencing adapters and, after transferring the PCRs to the modern DNA facility, amplified them with Kappa Hifi for 25 cycles. We pooled and sequenced the libraries across several Illumina Miseq 2x75bp runs. We used Seqprep2 and prinseq (v. 0.20.4) to remove adapters and merge reads, and aligned the resulting data to the nucleotide BLAST database using MEGAN (v. 6.18.0) to identify the taxonomic origin of the tissue. In addition and to confirm the species ID, we used BWA (v. 0.7.12-r1039) to align each read to bison (*Bison bison;* NCBI *GCA\_000754665)*, cattle (*Bos taurus;* NCBI Btau\_4.6.1), bighorn sheep (*Ovis canadensis;* NCBI CP011912.1), and two-toed sloth (*Choloepus hoffmanni;* NCBI *KN174222.1*) genomes. The latter was used to assess what proportion of reads aligned spuriously. We used mapDamage (Jónsson et al. 2013) to verify the sample has expected damage patterns of ancient DNA.

*Species Determination from Ancient DNA*

From the two combined shotgun sequencing libraries derived from the FS-305 ankle wrap sample, we generated 12,289,612 paired reads 11,498,964 of which were merged with *seqprep*. Of these, the BLAST analysis showed that most reads were from bacteria, as expected for ancient samples. The majority of reads identified as mammalian through BLAST aligned to bison and cattle, with the next highest proportion aligning to bighorn sheep, which is also a member of the *Bovidae* family. We next aligned non-duplicated reads to bison, cattle, bighorn sheep, and two-toed sloth genomes directly, the latter to estimate the number of reads that aligned spuriously. After duplicate removal, 288,619 reads aligned to bison, 269,601 reads aligned to cattle (a close relative of bison that was not present on the prehistoric landscape), 138,778 reads aligned to bighorn sheep (a relative of cattle and bison), and 5,430 reads aligned to the two-toed sloth. These genetic results are consistent with a species attribution of bison.

*Sex Determination from Ancient DNA*

To identify the biological sex of the FS-305 ankle wrap sample, we compared the number of reads that mapped to assembled cattle X-chromosome (a contiguous bison genome assembly is not available at present) from four comparatively sized autosomal scaffolds (chromosomes 12-15). Because males have only one copy of the X chromosome, our hypothesis was that we would find approximately half the number of reads mapping to the X as to the autosomal scaffold if the specimen was from a male but approximately the same number of reads mapping to the X and the autosome if the sample was from a female.

*Least Cost Path Analysis*

We determined the least cost paths between the Promontory caves and high-C4 locations in northern Arizona and central Colorado (Franktown Cave). The least cost path analysis was conducted using a digital elevation model (DEM) (courtesy USGS) with a 100 m scale. A slope raster was created using ArcGIS Pro’s “Slope” tool. Using Promontory Cave 1 as the origin location, the “Cost Distance” tool was used to create a cost distance surface as well as a backlink raster based on slope. The “Cost Path to Polyline” tool was used to create the least cost paths from Promontory Caves to the other locations. The paths shown in Supplemental Figure 1 demonstrate least cost paths based solely on slope and no other factors.

When unconstrainted, the shortest path to Arizona travelled west around Great Salt Lake. We forced the path to go around the east site of Great Salt Lake, given the Promontory Phase sites in that area and that the east side of Great Salt Lake is primarily salt flats, by clipping the DEM at the northwest corner of Great Salt Lake. This produced the route between Promontory and northern Arizona, shown below.



Supplemental Figure 1. Least-cost paths from the Promontory Caves to locations with abundant C4 grasses in northern Arizona and central Colorado.

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