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Stable Isotope and Ancient DNA Analysis of Dog Remains From Cathlapotle (45CL1), a Contact-Era Site on the Lower Columbia River

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Stable isotope and ancient DNA analysis of dog remains from Cathlapotle
(45CL1), a contact-era site on the Lower Columbia River.

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Abstract

This study reports ancient DNA (aDNA) and stable isotope analyses of eight dog skeletal elements from the Cathlapotle site on the Lower Columbia River of the western United States. The aDNA analysis confirmed the elements as dogs (*Canis lupus familiaris*). Two haplotypes were found, both of which group within dog Clade A, and have patchy distributions to the north in British Columbia and as far south as Teotihuacan (Mexico). The isotopic analysis showed that the dogs' dietary protein was derived almost exclusively from marine sources. Lower Columbia River ethnohistoric accounts and Cathlapotle zooarchaeological records indicate that while marine fish were dietary keystones, the local diet was more diverse, and included terrestrial organisms and freshwater fishes. This apparent discrepancy raises the possibility the dogs were selectively fed. Thus their diet may not be a close proxy for human diet in this context.

Keywords: Northwest Coast, dogs, aDNA analysis, isotopes, diet.

1. Introduction

There is a long standing debate among archaeologists in Cascadia, a portion of western North America encompassing the Northwest Coast, Intermontane Plateau and portions of the Subarctic (Figure 1), over whether the region's ancient economies were focal, even specialized, structured around a few keystone resources, especially salmon (*Oncorhynchus* spp.) and other marine resources, or whether they were built on multiple keystone resources. This debate emphasizes salmon fishing, which through intensification of harvesting and storage, is often viewed as the prime mover of social and cultural evolution in Cascadia (e.g. Matson and Coupland 1995, Hayden 1997). Others hold that while salmon was a keystone economic resource, a diverse subsistence base was fundamental to supporting the area's large populations and complex social arrangements (e.g. Ames and Maschner 1999). **FIGURE 1 ABOUT HERE.**

This debate is difficult to resolve in part because the available evidence can be contradictory. On one hand, there is growing evidence for a diversified resource base. The salmon focus – what Monks (1987) called “salmonopia”- masks the importance of other marine fish such as herring (McKechnie et al. 2014) and cod (e.g. papers in Moss and Cannon 2011). There was significant modification of intertidal zones to manage and increase mollusk production, at least in some areas (e. g. Groesbeck et al. 2014) as well as management of terrestrial habitats for plant production (e.g. Deur and Turner 2005, Turner 2014). Some scholars (e.g. Campbell and Butler 2010) propose such management extended to animals and fish. While recent studies of zooarchaeological

records suggest that in a few places economies did focus narrowly on salmon (Coupland et al. 2010), more often they did not (Butler and Campbell 2004). Butler and Campbell reviewed zooarchaeological records in two study regions in southern Cascadia, one coastal and one in the interior, and found no evidence of intensification of salmon in either area during the last 7000 years, the period for which they had adequate samples. They measured intensification in two related ways: as specialization (narrowing diet breadth) and resource depression (declining prey abundance) (Butler and Campbell 2004: 337). While salmon were everywhere important, Butler and Campbell found evidence for diverse resource bases reflecting local conditions. On the other hand, stable isotopic evidence seems to point to a narrow, marine diet. Schwarcz et al. (2014), reviewing earlier published (Chisholm et al. 1983) and unpublished isotopic analyses of human remains from the British Columbia coast and interior, conclude protein sources were exclusively marine in origin and were primarily carnivorous fish and sea mammals. They see salmon as the principle resource. The virtual absence of terrestrial resources in their evidence leads them to postulate a dietary taboo on the coast against eating terrestrial animals, despite their exploitation for hides, bone etc. There is no ethnographic support for such a taboo. Human remains from the interior (a 100 km or more away from the coast) show a more mixed diet.

We present the results of stable isotopic and aDNA analyses of dog remains from Cathlapotle (45CL1), a site located on the Lower Columbia River (LCR), in southwestern Washington State USA (Figure 1) dating between AD 1450 and ca. 1830 (Ames and Sobel 2009). Archaeological and ethnohistoric data (reviewed below)

suggest a diverse diet with aquatic (marine, riverine, wet land) and terrestrial components. We compare the isotopic results to those of human and dog isotopic studies elsewhere on the Northwest Coast. aDNA analysis confirms the osteological identification of the canid elements used in the isotopic study as dog. We also compare the aDNA results with canids from Cascadia and North America. The study was originally undertaken to test a hypothesis that the salmon focus developed post-contact as a result of depopulation (Campbell 1991, Butler 2000) using stable isotopes. That test was not successful, for reasons discussed below, but it structured how samples were selected and that information is retained here.

The use of dogs as proxies for human diets is a well-established methodology (e.g. Cannon et al. 1999, Tankersley and Koster 2009, Guiry 2012, 2013), although dog and human isotope values sometimes differ (Eriksson and Zagorska 2000, Bocherens et al. 2014, Tsutaya et al. 2014). When human remains are scarce or where cultural sensitivities regarding destructive testing of human remains exist, as they do in the LCR, dog remains provide a potentially useful way to track human diets. No stable isotope or aDNA analyses of human remains from the LCR or adjacent regions have been accomplished to date nor are there human remains from Cathlapotle available for such analysis were it possible. Our work focused on canid remains from well-defined, pre-and post-contact depositional contexts. An underlying assumption of this and other similar studies is that dogs essentially followed an encounter dietary strategy, consuming the same animal products and in roughly the same proportions as did the

humans with which they are associated (Guiry 2012, 2013) either directly or by eating human feces (caecotrophy) (Tankersley and Koster 2010; Guiry 2012, Mack in press).

One of us (RLL) identified the remains as dog using morphology, which was verified using aDNA analysis carried out by Simon Fraser University Ancient DNA Laboratory. We wished to exclude the possibility that other canids such as *C. latrans* or *C. lupus lupus* were present in the sample. In addition, the DNA produced new haplotype information for dogs in the LCR, which have not been explored to date.

FIGURE 2 ABOUT HERE

2. Background

Cathlapotle is located in the Wapato Valley (aka Portland Basin) portion of the Lower Columbia River (LCR), the lowermost 320 km of the Columbia as it flows into the Pacific Ocean (Figure 2). It is 11 km downriver from Vancouver, WA and Portland, OR. The Wapato Valley encompasses the river's relatively broad floodplain and immediate surrounding uplands between the Sandy and Cowlitz Rivers and contains the metropolitan areas of Portland, OR and Vancouver, WA. The Columbia River was one of the world's foremost rivers for Pacific salmon prior to extensive dam construction after World War II. Salmon are anadromous, hatching in freshwater, moving downstream to the ocean as immature fish, reaching maturity in the Pacific, and then returning to their natal stream to spawn and die, sometimes in prodigious numbers. LCR human populations were comparatively large and dense prior to contact and even after

several major epidemics (Boyd 1999, 2013). Contact began c. 1774, with the first documented exploratory voyages along the coast (Hajda 1984; Gibson 1992; Ames and Maschner 1999). On-going contact began in 1792 with the European exploration of the Columbia River's mouth (Vancouver 1926), and the start of the maritime fur trade on the river (Gibson 1992; Igler 2004).

Cathlapotle was one of the largest and foremost Native American villages in the Wapato Valley (Figure 2). It was noted in multiple historic accounts (Sobel 2004). Lewis and Clark visited the village in 1806 (Moulton 1990). It is clear from these accounts that its people and chief(s) had ongoing and complex relationships with fur traders and other Europeans in the region. Located near the confluence of the Columbia River with four tributaries of differing sizes, its immediate hinterland included a diverse, fine grained patchwork of floodplain wetland, meadow and woodland habitats as well as closely adjacent upland plateaus.

The site was excavated over several field seasons in the 1990s by Portland State University under Ames' direction (Ames et al., 1999, Sobel, 2004, Smith, 2008). It is large (15,000 m² x 2 m deep), well stratified and contains six large depressions marking the locations of large wooden houses (Ames et al., 1999; 2008; Sobel, 2004; Smith, 2008; Shepard 2014). Chronological control is provided by 52 terrestrial radiocarbon dates (Ames and Sobel 2009) and a large, well stratified assemblage of fur-trade era trade goods. The site was occupied from AD 1450 until ca AD 1830 with very clearly distinguished pre-contact and post-contact era deposits (Ames and Sobel, 2009). The contact era at Cathlapotle begins in 1792 with the first recorded encounter between the

townspeople and Europeans. Euro-american glass and ceramic trade goods (Kaehler, 2002; Cromwell, 2011) appear abruptly about 70 cm below surface. Pre-contact is the period between AD 1450 and 1792 and is stratigraphically below the trade goods. The post contact spans the time between 1792 and ca. 1830 and is represented by the upper 70 cm or so of deposit. Overall, the site produced a rich assemblage of artifacts, including faunal remains analyzed by Butler (2002, Butler and Martin 2013), Lyman (2003), and Frederick (2009).

The Cathlapotle archaeofauna contained an NISP of 41 canids, 3 of which were identified to the level of *Canis lupus familiaris* and 14 as *Canis l. familiaris cf.* None were identified as either *C. lupus* or *C. latrans*. The sample used here was chosen from those 17 elements. Eight of the 17 were post-contact in age, nine pre-contact in age. The elements used were selected as much as possible from sheet midden deposits (Ames et al. 1999) (Figure 3), which are well stratified with abrupt boundaries. They were also selected to avoid sampling the same individual. These strictures produced the sample of eight that we analyzed.

FIGURE 3 ABOUT HERE

The dog elements were not directly dated; we rely instead on stratigraphic position, associated artifacts and associated terrestrial radiocarbon dates. The elements were not radiometrically dated for two reasons: radiocarbon dates from ca. AD 1650 – 1800 can produce high probability calibrations which may be as much as 100 or more years off the sample's true age (Ames and Sobel 2009); and, assuming the dogs' diet was wholly or in part marine in origin, a marine reservoir correction would be necessary

for any calibration. Presently, there is no marine reservoir correction for the waters off the mouth of the Columbia, nor a freshwater correction for the river itself. Consequently any radiocarbon dates on the dog elements would have been difficult to interpret.

3. LCR and Cathlapotle subsistence, dogs and what they may have eaten.

As described in the documentary record, Lower Columbia River subsistence was, in Gahr's apropos phrasing (Gahr 2013, 64), a mix of resource diversity and focal resources. She describes the focal resources as keystone resources, which she identifies as cervids (deer [*Odocoileus* sp.] and wapiti, a.k.a. elk [*Cervus elaphus*]); berries, camas (*Camassia quamash*); wapato (*Sagittaria latifolia*) – these being exploited for their roots - and Western Red Cedar (*Thuja plicata*), used for industrial purposes. Her review did not include fish. In a parallel study using ethnohistoric accounts of fishing, Butler and Martin (2013) found four anadromous fish taxa to have been important. These taxa are: Salmonidae, most importantly the five species of Pacific salmon (*Oncorhynchus* sp.), especially Chinook salmon (*O. tshawytscha*); eulachon (*Thaleichthys pacificus*), a smelt; lamprey (*Entosphenus* sp.), especially Pacific lamprey (*E. tridentate*); and sturgeon (*Acipenser* sp.). Their review of fish archaeofaunas from the LCR (Butler and Martin 2013) indicates that salmon, eulachon and sturgeon are widely abundant, but that some freshwater fish, including minnows (Cyprinidae), suckers (Catostomidae), and Three spine stickleback (*Gasterosteus aculeatus*) are also archaeologically abundant although virtually absent from

documentary accounts. Butler and Martin (2013, 94 - 95) note that stickleback are a traditional human and dog food in southwest Alaska. They also observe that fish bones far outnumber mammal and bird bones in LCR archaeofaunas, pointing to the importance of fish. This is certainly the case at Cathlapotle where fish bones are vastly more common than mammal or avian bones, when NISP is standardized by sampled deposit volume. The diversity in fish resources was compounded by local variability in fish availability along the river resulting from a number of factors, making it difficult to assign keystone status to any particular fish. Thus, the fifth keystone in LCR subsistence is perhaps just fish.

The Cathlapotle mammalian archaeofauna (Lyman 2003), while diverse (Figure 4) (Supplemental Materials Table 1) as measured by taxonomic richness (Lyman, 2003), is dominated by cervid elements. Over 80% of the 6922 Number of Identified Specimens (NISP) are deer and wapiti. The avian archaeofauna, while also diverse, is dominated by water birds (Figure 4): ducks (*Anas* sp.) primarily, but some geese and swans as well (Frederick, 2009). It is worth noting here that birds, as with the freshwater fish discussed below, are not mentioned in the documentary record.

FIGURE 4 ABOUT HERE

The fish assemblage from Cathlapotle represents a diverse fauna (Figure 4), including salmon and trout, sturgeon, and resident fresh-water fish, including the minnows and suckers which were abundant in the wetland backwaters of the Wapato Valley. Remains from the small-bodied eulachon and stickleback were prominent in fine mesh samples. Pacific lamprey has not been identified in archaeological contexts, likely

due to its cartilaginous skeleton (Butler and Martin, 2013). Potential sources of marine protein other than fish are rare in the Cathlapotle archaeofauna. Small numbers of harbor seal (*Phoca vitulina*, NISP = 65, <1%) specimens are present in the mammalian archaeofauna (Lyman et al. 2002). There are no birds that are exclusively marine. Taken as a whole, the archaeofauna represents animals that inhabit marine systems at least during part of their life cycle, fully freshwater habitats, and terrestrial settings.

All of this suggests dogs had a wide array of food from a range of habitats available to them. However, there is little to no information in the documentary record on what they ate or were fed. Fur traders and other early European and American travelers, such as the Lewis and Clark expedition, commonly ate dogs but say nothing in their journals about what the dogs were fed (Mack i.p.). One factor that may have affected whether dogs were fed salmon (or restricted from eating it) and potentially other anadromous fish is so-called salmon-poisoning disease (SPD).

SPD is an infection by a bacterium - *Neorickettsia helminthoeca* – that as part of its life cycle can infect most of the anadromous fish of economic importance along the Lower Columbia (Headley et al. 2011, Table 1). The infection results from eating raw fish. SPD is known to infect 24 species of fish based either on direct observation in the field or experimental data (the fish are infected in a laboratory) (Headley et al. 2011). Among these are all five species of Pacific salmon, lamprey, sucker, minnows, and Three-spine stickleback. Missing from the list are eulachon and sturgeon. What is not discussed by Headley et al. (2011) is whether these are not susceptible or that no one has investigated them. Salmon become infected in freshwater so presumably all of

these other fish do. SPD is endemic to the Pacific coast from the Sacramento River north, including both in the USA and British Columbia because its life cycle requires a snail found only in this region. In the USA, its range extends inland from the coast to the western slopes of the Cascade mountain range. With the recent exception of southern Brazil, this is the only place in the world where it is known to occur. It was first reported in 1814 in northwest Oregon (Headley et al. 2011). Humans, dogs, foxes and coyotes as well as other mammals and birds are susceptible to it. The symptoms of infection are severe and death takes about 18 days after infection. Dogs sometimes survive. A Chinook scholar (Johnson, pers. comm.) discounts its importance for what local dogs ate, observing that dogs do survive and suspecting it would be impossible to keep them away from salmon scraps including viscera. Also, that choice pieces were reserved for people, and that these included often highly boney and cartilaginous cuts. He notes that dogs were, for cultural reasons, not allowed to eat salmon hearts. He also suggests that over time, local dogs would likely develop a resistance or immunity to the parasite as a consequence of natural selection. It is his view, based on inquiries elsewhere along the coast, that today dogs could not survive in indigenous settings without an immunity. The early reports of SPD are coincident with the arrival from elsewhere of dogs presumably without immunity.

At this point, regarding SPD, we are left with several alternatives: LCR dogs were immune; they were not immune, the immunity was partial; or fish were differentially infected. Since SPD occurs only on the Pacific Northwest, it is safe to assume it was not introduced along with other biological exotics during the fur trade. If the dogs were

immune, the issue is moot. If they were not immune or partially immune, people either tolerated a high death rate among dogs or managed their diets, keeping the dogs away from infested fish. The fish may also have carried different disease loads. Of the fish at issue here, salmon, lamprey and one species of minnow are demonstrated to carry it in the wild; all the rest were infected experimentally. Perhaps they were not as subject to infection in the wild. Regrettably, we do not have the epidemiological evidence with which to evaluate these alternatives. Native testimony is clear their dogs are and were immune.

Mack (i.p.) reviews the documentary evidence on the role of dogs in the LCR and concludes they served as village watch dogs, companions, hunting partners (e.g. Rollins 1935) and as “village sanitation specialists” – cleaning up human feces (caecotrophy). As Guiry (2012) observes, while humans do not do this, dogs do, and the effect of that difference on the use of dogs as a human dietary proxy is not well understood. Following his advice, we can simply note this. The dogs probably also ate butchery scraps, offal and bones. To the north, in the Salish Sea region of southern British Columbia and northwestern Washington, two breeds of dogs existed, the regular village dog and so-called wool dogs. “Village dogs” (Boyko and Boyko 2014, 186) are the common dogs found world -wide that are “relatively free breeding and often times partially free ranging” and scavenge at least some of their food. The dogs in this study are presumably village dogs. Wool dogs were Spitz-like dogs whose fur was used in weaving blankets (Schulting 1994, Crockford 1997, Crockford and Pye 1997, Solazzo et al. 2011). Schulting (1994) suggests there is a faint possibility these dogs were kept on

the LCR; Mack (i.p.) sees no evidence for wool dogs in the LCR. The breeds have been distinguished osteologically in the Salish Sea region (Crockford 1997). Schulting attempted to identify its hairs in a blanket using isotopic analysis. He argued that hairs present in the blanket with a strong marine signature were those of the wool dog, since the hairs from other potential sources, such as coyotes, would have a stronger terrestrial signal, as the dogs would have been fed or scavenged salmon.

4. Expectations

The available documentary and zooarchaeological evidence strongly indicates that the Chinookan peoples of the Lower Columbia River had a diverse diet harvested from a range of sources. Consequently their bone isotope signatures would not be as strongly marine as those reported for other Northwest Coast peoples (e.g. Schwarcz et al, 2014). Of course, stable isotopic data for humans are not available in the LCR nor are they likely to be in the foreseeable future. Assuming, as do most stable isotopic studies of dog diets, that the dogs were village dogs, and at least partially free-ranging and otherwise eating – either being fed or scavenging (Schulting 1994) – what the people ate, we would expect a more generalist signature for the dogs as well.

If salmon harvesting was intensified post-contact, as Butler (2000) hypothesized, our original empirical expectation was that this subsistence shift would be visible in the dogs' isotopic signatures by a rise in trophic level (as measured by $\delta^{15}\text{N}$) as their diets became less diverse and trophically higher ranked as a consequence of consuming

more salmon, a piscivore, and by an increase in the maritime contribution (indicated by relatively enriched $\delta^{13}\text{C}$) as the amount of dietary salmon increased.

Materials and Methods.

4.1 aDNA analysis

The eight canid samples from Cathlapotle identified as dog based on morphology were submitted to the dedicated Ancient DNA Laboratory at Simon Fraser University (SFU) (Table 1), which follows strict contamination control protocols, such as: separation of the pre-PCR (Polymerase Chain Reaction) and post-PCR work spaces; use of ancient DNA dedicated equipment; analysis of both positive and negative controls alongside the ancient DNA samples; and multiple extractions from the same sample in order to replicate the original results.

Samples were chemically decontaminated through submersion in a 6% sodium hypochlorite solution, followed by submersion in 1N HCl and 1N NaOH, then rinsed in double distilled water. The bone samples were then UV irradiated for 60 min, crushed into power and extracted following a silica-spin column method (Yang et al., 1998), modified following Speller et al. 2014.

Two pairs of published PCR primers were used to target two overlapping fragments of *Canis* mtDNA control region to produce a 345bp fragment: MitH52-DL: TCGAGGCATGGTGATTAAGCC /MitL63: ACCCCTACATTCATATATTGAATCACC) (Ishiguro et al., 2000), and DL-4 GCATATCACTTAGTCCAATAAGGG / DL-Hcanx

CCTGAGGTAAGAACCAGATGCC) (Leonard et al., 2002). PCR amplifications were performed on an Eppendorf™ Mastercycler Personal Thermocycler using a 30 µL reaction volume containing 1.5X Applied Biosystems™ Buffer, 0.3 µM of each primer, 2 mM MgCl₂, 0.2 mM dNTP, 1.0 mg/mL BSA, 3.0 µL DNA sample and 2.5 U AmpliTaq Gold LD (Applied Biosystems). Successfully amplified products were sequenced using forward and reverse primers at the Macrogen Ltd. facility in Seoul, Korea. DG2 and DG4 underwent repeat extractions, and all samples were amplified at least twice to ensure the results were replicable.

The ancient sequences were visually edited using ChromasPro software (www.technelysium.com.au), and sequences were truncated to 301bp once primer sequences were removed. Final sequences spanned region 15483-15783 of the *C. familiaris* reference mtDNA genome NC002008 (Kim et al. 1998). The obtained sequences were compared to Genbank sequences through the BLAST application to determine their closest match, and to ensure they did not match any other unexpected species or sequences. Haplotypes were assigned following Pereira et al. (2004). Multiple alignments of the ancient sequences and published canid reference sequences were conducted using ClustalW (Thompson et al. 1994), through BioEdit (Hall 1999). Phylogenetic comparisons with a subset of modern dog (Savolainen et al 2002) haplotypes from Genbank was conducted through Mega3 software (Kumar et al. 2004), using neighbor-joining through the Kimura 2-parameter. Additional comparisons of the ancient haplotypes from Cathalpotle with other ancient haplotypes recovered from pre-contact American archaeological sites (Leonard et al. 2002; Barta 2006; Brown et al.

2013; Thalmann et al. 2013) were conducted though Network 4.6.1.2 (Bandelt et al. 1999).

TABLE 1 ABOUT HERE

4.2 Isotopic analysis

Five of the original eight Cathlapotle canid specimens (Table 2) were submitted to Beta Analytic for isotopic analysis; three specimens were consumed in the aDNA analysis. These three specimen were not replaced with additional ones because that would have significantly depleted the remaining nine *Canis familiaris* elements. Three of the surviving five elements were from post-contact deposits and two were from pre-contact contexts. In addition, to provide background isotopic values for possible prey items of dogs and for carnivores and omnivores operating in the local environment, 12 archaeological remains from common animal taxa recorded at Cathlapotle were analyzed: salmon, sturgeon, wapiti (elk), mallard, black bear (*Ursus americanus*), and raccoon (*Procyon lotor*). Black bear and raccoon were selected as dietary proxies for dogs potentially provisioning themselves in local habitats. Raccoons have a highly varied diet ranging from insects through amphibians to small birds (Maser et al. 1984). Black bears are more omnivorous (Maser et al. 1984). Both would have been encountered on the local floodplain. Neither is a perfect proxy but they are what was available. There are no other canids, i.e. coyotes or wolves, identified in the zooarchaeological assemblage to use for this purpose. After the Beta results were returned, an additional 20 fish elements were submitted to the Isotope Laboratory at the

Department of Human Evolution, Max Planck Institute for Evolutionary Anthropology in Leipzig, Germany (Table 2) to expand the sample of fish and fish habitats. In addition to salmon and sturgeon elements, these elements represented northern pikeminnow (*Ptychocheilus oregonensis*), large scale sucker (*Catostomus macrocheilus*), and eulachon, prominent in the fish fauna at Cathlapotle.

The procedures for collagen extraction and isotope measurement at the Beta laboratories are described as follows (Hood, pers com): In the Beta Analytic laboratory, the material was first tested for friability ("softness"). Very soft bone material is an indication of the potential absence of the collagen fraction (basal bone protein acting as a "reinforcing agent" within the crystalline apatite structure). It was then washed in de-ionized water, the surface scraped free of the outer most layers and then gently crushed. Dilute, cold HCl acid was repeatedly applied and replenished until the mineral fraction (bone apatite) was eliminated. The collagen was then dissected and inspected for rootlets. Any rootlets present were also removed when replenishing the acid solutions. This was followed by additional pretreatment with sodium hydroxide (NaOH) to ensure the absence of secondary organic acids. Beta Analytic did not provide collagen criteria indicators such as the C:N ratio, %C, %N, or collagen yields. At the Department of Anthropology, UBC (Vancouver, Canada) and at the Department of Human Evolution in Leipzig, collagen was extracted from approximately 300 mg of whole bone, following the procedures outlined in Richards and Hedges (1999) with the addition of an ultrafiltration step (Brown et al., 1988). Samples were demineralised in 0.5M HCl at 5 °C for 5 days, then rinsed in deionised water, the resultant solid

gelatinised at 70 °C for 24 hours and then the solution filtered through 30 kDa ultrafilters. Carbon and nitrogen isotope values were measured using a Thermo-Finnigan Flash EA coupled to a Delta V isotope ratio mass spectrometer. All results had acceptable C:N ratios (between 2.9 and 3.6, DeNiro 1986) and %C and %N values (Ambrose, 1990) and the errors on the $\delta^{13}\text{C}^0_{/00}$ and $\delta^{15}\text{N}^0_{/00}$ measurements are less than 0.2 per mil. **TABLE 2 ABOUT HERE**

Salmon was the only taxon successfully extracted and measured in both laboratories and the values are quite similar. Our salmon values are also close to those given by Szpak et al. (2009, Table 1) (n=19, mean $\delta^{13}\text{C}^0_{/00} = -15.1 \pm 0.7$, mean $\delta^{15}\text{N}^0_{/00} = 12.2 \pm 0.8$) and Scharcz et al. (2014, Table S3) (n=4, mean $\delta^{13}\text{C}^0_{/00} = -17.8 \pm 1.3$, mean $\delta^{15}\text{N}^0_{/00} = 15.4 \pm 0.5$). Therefore, we believe that the results from both laboratories are comparable and we combine the data from both below. The absence of reported collagen criteria indicators for the specimen analyzed by Beta Analytic that were not duplicated could be seen as problematic, especially for the sturgeon, since efforts to replicate those results at the UBC laboratory had poor C:N. The reported sturgeon values should consequently be viewed with caution. However, the results for the other animals, most importantly the dogs, are consistent with, although not identical to, stable isotope values reported elsewhere (e.g. Cannon et al. 1999, Grier 2006, Corr et al. 2009, Szpak et al. 2009, Schwarzcz et al. 2014) and we are confident of them.

5. Results

5.1 aDNA

Seven of the eight canid samples yielded amplifiable DNA (Table 1), resulting in two mitochondrial haplotypes detected in five and two individuals, respectively. Several criteria point to the authenticity of the sequences and rule out contamination from modern sources: 1) no PCR amplifications were observed in blank extracts and PCR negative controls; 2) the presence of two haplotypes within the samples as a whole, as well as within different extraction batches, minimizes the likelihood that they were from the same contamination source; 3) a subset of the sequences displayed characteristic damage patterns associated with degraded DNA (Brotherton et al. 2007, Gilbert et al. 2007); and 4) repeat extractions and/or repeat amplifications of all successfully amplified samples confirmed haplotype identifications.

According to the SFU lab protocols, species identification is assigned to a sample only if it matches identically or very closely with published reference sequences while being clearly distinctive from the most closely related species and if no other evidence including reproducibility tests or additional sequencing of the same sample indicates the possibility of base pair ambiguities. Both haplotypes matched identically with *Canis lupus familiaris* reference sequences in Genbank (Table 1). Phylogenetic comparison of the Cathlapotle sequences with modern canid reference sequences showed all the aDNA sequences clustered together with those of modern dogs (Figure 5), differing from the closely related coyote (*Canis latrans*). In addition to confirming the identification of the canid specimens as dog, the aDNA analysis helps allay concerns

about samples representing the same dog. The two samples recovered spatially closest together, samples DG3 and DG4, have different haplotypes.

Importantly, the aDNA analysis also provides additional mitochondrial haplotype data concerning ancient domestic dogs in Cascadia. Comparisons with previously published dog haplotypes indicated that ancient Cathlapotle sequences corresponded to Haplotype A3 (15627, 15639^{T/A}) as described by Savolainen et al. 2002, and Haplotype D25 (15621, 15639^{T/A}) as described by Vilà et al. 1997 (Table 1). When phylogenetically compared to other modern dog haplotypes, both haplotypes fall within the most diverse of the dog clades, Clade A (Savolainen et al. 2002), which includes a range of modern breeds from European, African, Indian, and American dogs, and is the most common lineage found in ancient North American dogs studied to date (Leonard et al., 2002; Brown et al. 2013; Barta 2006).

The two ancient haplotypes identified in this study match those recovered from other archaeological dog remains from the Americas (Figure 6). Haplotype A3 was detected in two archaeological sites on the Central Coast of British Columbia (Barta, 2006, 'hapC'): Namu (5 samples), and McNaughton Island (2 samples). A3 has also been detected in a 1,000 year old site in Florida as well as the ca. 8,500BP Koster site, in Illinois (Thalmann et al. 2013). The second haplotype, D25, has also been recovered from one archaeological individual each from Nitlilquotlank on the Central Coast of British Columbia, Dionisio Point on Galiano Island in the Salish Sea, and T'ukw'aa on the west coast of Vancouver Island (Barta, 2006, 'Hapl') (Figure 1), as well as at Teotihuacan, Mexico dating to 1300 BP (Leonard et al., 2002).

FIGURE 5 ABOUT HERE

FIGURE 6 ABOUT HERE

5.2 Isotopic analysis

The small sample size – five dogs – is a limitation, although it is equivalent to samples in other recently published studies (e.g. Byrd et al. 2013, Bocherens et al. 2014). Further, the close grouping of the isotopic values from dogs widely dispersed in time and space indicates the small sample size is not as serious a problem as it might be. The isotopic values of the pre and postcontact samples cluster tightly (Figure 7) while dogs from widely separated excavation units are also either identical or very similar in their values. For example, the two elements with virtually identical isotopic values, samples 4 and 6, were recovered 2 m apart in depth from units 20 m distant and one is pre-contact (6) and the other postcontact (4) in age. The coherence of the results allows confidence in making inferences at the site level.

Isotope ratios for the potential dog dietary items from freshwater and terrestrial habitats are extremely distinct from dietary items with a marine life-phase (Table 2, Figure 7a). Carbon isotopes of the freshwater fish (minnows, suckers), terrestrial mammals, and ducks have much more depleted $\delta^{13}\text{C}_{\text{‰}}$ values (range: $-17.8 \delta^{13}\text{C}_{\text{‰}}$ to $-23.3 \delta^{13}\text{C}_{\text{‰}}$) than do the anadromous sturgeon, salmon and eulachon (range: $-13.4 \delta^{13}\text{C}_{\text{‰}}$ to $-15.9 \delta^{13}\text{C}_{\text{‰}}$). The relatively low $\delta^{13}\text{C}_{\text{‰}}$ values for the five dog specimens (range: $-14.1 \delta^{13}\text{C}_{\text{‰}}$ to $-15.1 \delta^{13}\text{C}_{\text{‰}}$) clearly indicate consumption of marine resources, which were the dominant protein source in their diets (Richards and Hedges 1999).

Nitrogen isotopes also indicate the dogs mainly consumed creatures that spent part of their life cycle at sea. The $\delta^{15}\text{N}/_{00}$ values for freshwater fish, ducks, and mammals are consistently much lower (range: $3.8 \delta^{15}\text{N}/_{00}$ to $9.6 \delta^{15}\text{N}/_{00}$) than those for the anadromous fish (range: $13.0 \delta^{15}\text{N}/_{00}$ to $16.4 \delta^{15}\text{N}/_{00}$); which show close similarity to the dog $\delta^{15}\text{N}/_{00}$ values (range: $15.2 \delta^{15}\text{N}/_{00}$ to $17.2 \delta^{15}\text{N}/_{00}$). Since the dogs' carbon and nitrogen isotope values are close to those for salmon, eulachon and sturgeon (Figure 7b), it is clear they derived little protein from freshwater wetland or upland habitats – i.e. they were not eating minnows, suckers or cervids to any significant degree.

FIGURE 7 ABOUT HERE.

Two fish that could potentially have been important dietary items for which we do not have isotopic values are the sticklebacks, which are well documented zooarchaeologically at the site, and lamprey, which are documented in the ethnohistoric record (Miller 2012, Butler and Martin 2013). Sticklebacks are freshwater fish that would have been taken in backwater habitats so it is probable their isotopic signature would be similar to the minnows and suckers we tested. It therefore seems unlikely then that the dogs in our sample were eating them in any numbers. Lamprey, however, are anadromous so their isotopic signature would presumably be similar to the anadromous fish in our sample. Their skeleton lacks calcified structures and their absence from LCR archaeofaunas is likely because of preservation biases (Butler and Martin 2013).

Which marine-anadromous taxa the dogs were eating is slightly less clear. Predators are trophically a step above their prey and will be enriched in nitrogen relative

to their prey. Predator – prey relationships are established using a stepwise offset between isotopic values. A standard predator-prey nitrogen offset is 3.4 ‰ (e.g. Post, 2002; Minigawa and Wada, 1984). Carbon is more stable across trophic levels with very small and variable offsets (Schoeninger and DeNiro, 1984; Fischer et al., 2007). We use ranges of 0 – 2 ‰ for carbon and 3 – 5 ‰ for nitrogen, following Bocherens and Drucker (2003). Our data suggest the Cathlapotle dogs were primarily eating eulachon of the anadromous fish that we tested. Their mean carbon values differ (1.12 ‰) more than they do for salmon and sturgeon. The mean carbon values for the dogs, salmon and sturgeon are very close (-14.5 ‰, -14.1 ‰ and -13.8 ‰ respectively). The differences (dog: salmon -0.4 ‰ and dog: sturgeon -0.7 ‰) are within the range of potential measurement error and too small to be useful. The nitrogen data show an offset of 2.5 ‰ between dogs and eulachon suggesting the dogs were eating eulachon, perhaps some salmon but little sturgeon. It is also quite plausible they were eating lamprey for which we do not have values. The mean nitrogen values for the dogs is 16.4 ‰, 16.4 ‰ for the sturgeon, and 14.5 ‰ for the salmon with differences of dog:salmon of 1.86 ‰ and dog:sturgeon of 0.0 ‰, again well within the range of measurement error. Zooarchaeological data adds support to the inference that the dogs were primarily eating eulachon. Eulachon elements constitute 56% of the fish NISP in bulk samples collected at Cathlapotle (Table S3, Butler 2002). The village was close to the Lewis River, which hosted a large eulachon run historically. Ethnohistoric accounts (e.g. Boyd and Hajda 1987, Sobel 2004, Butler and Martin 2013) indicate eulachon were taken and stored in prodigious amounts so the dried fish would have been

available over much of the year. The issue of which fish the dogs were eating aside, it is clear is that the dogs' isotopic values have a very strong marine signal and they were not eating terrestrial or freshwater organisms. This is consistent with other stable isotope analyses of Northwest Coast dogs (Cannon et al. 1999, Barta 2006, Grier 2006).

To put Cathlapotle in a larger context, we compared our isotope data to the published records for dogs and human remains from other Cascadia archaeological sites. Figure 7a compares the $\delta^{13}\text{C}$ $^0_{\text{‰}}$ values for the Cathlapotle dogs to those for dogs from the Namu (Barta, 2006; Cannon et al., 1999) and Keatley Creek (Barta, 2006) archaeological sites; and to a sample of 38 human remains from Namu (Chisholm et al., 1983, Schwarzc 2014, Table S1) and 16 human remains from 8 localities in interior British Columbia (Schwarcz et al. 2014 Table S3). Namu is located on the central British Columbia Coast and Keatley Creek is in the south-central interior plateau of British Columbia (Figure 1) on the Thompson River, which supports a major salmon run. The other 8 Interior localities are in the same general interior region and have salmon runs. The closeness of the $\delta^{13}\text{C}$ $^0_{\text{‰}}$ values for the Namu human and dog samples indicates the dogs are good proxies for human diet, at least at Namu (Cannon et al. 1999). The Namu zooarchaeological record also shows an overwhelming emphasis on marine resources (Cannon, 1991). Barta (2006) suggests the relatively high $\delta^{13}\text{C}$ $^0_{\text{‰}}$ values of the Keatley Creek dogs reflects a mixed diet of salmon and terrestrial resources, given Keatley Creek's interior location. We wonder whether the pattern may also reflect salmon becoming more carbon depleted given that they stop eating as they ascend

rivers in their runs. The Interior human remains (which do not include remains from Keatley Creek) display an even stronger mixed pattern, probably reflecting the diverse sources of the remains. The Cathlapotle dog's values are intermediate between Namu and Keatley Creek, suggesting they had more terrestrial foods than the Namu dogs but less than the Keatley dogs. However, the Cathlapotle dogs' isotopic values align very closely with the Cathlapotle salmon pointing to the possibility that their position on the graph *vis a vis* Namu may reflect some variation in isotopic values among North Pacific marine resources either through time (Misarti 2007) or among different species of salmon (Romanuk and Levings 2005) as much as it does variation in the proportion of marine\terrestrial resources in their diet. **FIGURE 7 ABOUT HERE** Whatever the Cathlapotle dogs were eating, they were all eating much the same thing, given how tightly their $\delta^{13}\text{C}_{\text{‰}}$ values cluster. The spread of the Cathlapotle $\delta^{13}\text{C}_{\text{‰}}$ values is 1.4 ‰. Keatley Creek's, with a comparable sample size, is 2.4 ‰, and with a much larger sample, Namu's spread is almost identical to Keatley Creek's at 2.3 ‰.

The $\delta^{15}\text{N}_{\text{‰}}$ values of the Namu, Keatley Creek and Cathlapotle dogs and the Namu and interior humans (Schwarz 2014, Tables S1 and S3) are compared in Figure 7b, again including the Cathlapotle salmon. The mean $\delta^{15}\text{N}_{\text{‰}}$ value for the Namu humans is 19.3 ‰, which is higher than the Namu dog samples by more than 2 ‰. Similar offsets between human and dog values have been observed elsewhere (e.g. Tankersley and Koster 2009, Rick et al. 2011). Nitrogen values for the Namu and Cathlapotle dogs are very close and both are higher than the Cathlapotle salmon values, indicating both sets of dogs were eating salmon to some degree. The nitrogen

values of the Keatley Creek dogs fall much closer to the terrestrial prey and thus again, suggest a greater proportion of terrestrial food sources were consumed at this interior location than at Cathlapotle. Nitrogen values for Interior humans cover an extreme range from essentially terrestrial herbivore to top ranked carnivore. At minimum, as with the Interior human $\delta^{13}\text{C}_{\text{‰}}$ values, these point to a varied interior diet.

6. Discussion

6.1 aDNA

The primary goal of the ancient DNA analysis was to confirm the osteological identification of the skeletal elements as domestic dog. It did so. DNA species identification of archaeological remains is no longer new; this study reinforces the usefulness of DNA species identification of faunal remains for subsequent isotopic analyses (e.g. Byrd et al. 2013).

The analysis also pinpoints the presence of two ancient North American haplotypes within the LCR. Both haplotypes are relatively well dispersed in North America, and, for haplotype A3, may represent one of the earliest matrilineal types to enter the Americas, as evidenced by its presence at the Koster site in Illinois (Thalmann et al. 2013). The relatively large geographic distribution of both haplotypes points to the challenges in using mtDNA alone to effectively separate regional dog populations since dogs may have been regularly exchanged among human groups. Although, based on the limited ancient mtDNA data that has been generated to date (Figure 6), there

appears to be little overlap between haplotypes identified in pre-contact Arctic sites (Brown et al. 2013) with those on the Northwest Coast and further south into Central and South America (Figure 6). With further data, it will be intriguing to investigate to what extent these phylogeographic patterns reflect human migration and interaction patterns in the New World. The genetic structure of regional dog populations has the potential to yield valuable data on the regional and local structure of human behavior, however, this type of analysis will require systematically collected samples targeting the same genetic locus in order to produce comparable-population level data. The wide geographic and temporal distribution of both Cathlapotle haplotypes also suggests neither is a candidate to represent the wool dog.

6.2 Isotopic analysis

The dog isotope values are an average of all of the protein that was consumed by the dogs while the bones were formed and remodeled over their lifetimes, so provide a general view of the protein sources of diet over many years. As isotope analysis is a general measure of diet, it cannot determine occasional consumption of other foods like seals or salmon, but instead indicates the main dietary protein sources, and in this case the isotope results show the dogs' protein intake was largely from marine sources, conforming closely to other isotopic analyses of archaeological dogs from Cascadia (e.g. Barta, 2006; Grier, 2006; Cannon et al., 1999). At Cathlapotle the fish are most likely eulachon primarily based on the low nitrogen isotope values.

The apparently narrow diet for dogs contrasts sharply with the ethnohistorical and zooarchaeological records, which together strongly indicate a great diversity of animals was part of human subsistence, more like the diet in the British Columbia interior than on the coast. Dogs had available to them a rich and diverse array of resources harvested in terrestrial and freshwater habitats but they were not foraging across the full spectrum of animals being brought into Cathlapotle. Taking three examples, the people at Cathlapotle took wapiti in large numbers. The wapiti at Cathlapotle were much larger than modern wapiti in North America (Lyman 2006). The Roosevelt elk is the largest modern wapiti. Bulls weigh 300-500 kg and cows 260-285 kg. Many of the Cathlapotle wapiti would have been even larger based on skeletal evidence. The animals were hunted at Cathlapotle for their hides, meat and tool bone. The processed hides were certainly traded (Gibson 1992) and virtually all bone tools were made from wapiti metapodials (Fuld 2011). While the meat was eaten by humans, it was not being fed to the dogs to any degree. For example, only a very small number of wapiti bones (100 of an NISP of 3224) (Lyman 2003) show carnivore gnawing. Several freshwater fish taxa were taken, especially minnows and suckers, in large numbers that would have been available to scavenging dogs. Again, the dogs were not eating these fish. Finally, the dogs were not eating the harvested water birds present in the site. We conclude from this that the dogs were not simply eating what they encountered from the human diet (i.e. Rick et al., 2011), nor were they provisioning themselves in the local environment assuming their feeding habits were similar to coyotes', which primarily eat small mammals but take animals as large as deer and as

small as insects as well as seasonal fruit (Maser et al.1984). Presumably coyotes' isotopic values would fall within the terrestrial cluster in Figure 5 (e.g. Warsen et al. 2014). Rather, the dogs appear to have been fed anadromous fish, which we think were mainly eulachon, with some salmon and sturgeon. They also may have been fed lamprey for which we have no isotopic data.

This conclusion raises questions about the management of dogs in the LCR and perhaps elsewhere on the Northwest Coast. Controlling their diet implies the dogs were tied or penned and that there was selectivity in their feeding. Penning or tying would be necessary to keep dogs from scavenging for scraps. Currently the wool dog is the only evidence for dog management on the Northwest Coast. The development of a functionally specific dog breed requires long-term controlled breeding (Schulting 1994). Of course, this is not evidence for controlled feeding, but indicates it is not outside the realm of possibility. We have already been told LCR dogs were not allowed salmon hearts or boney salmon fillets but that the dogs had ready access to other salmon scraps. The early documentary record is mute as to whether dogs were tied or restrained in any way (e.g. Mack i.p.) although Mack quotes one journal passage which implies the presence of dog packs. These apparent contradictions are not resolvable with the data at hand. The questions also include why preferential feeding? PSD is one possibility. However, this potentially is carried by most anadromous and some freshwater fish, many of which the dogs were eating. Additionally, if that was the problem, why not feed the dogs the available cervid meat. They were feeding the dogs eulachon, which may not have carried SPD. If that can be demonstrated to be the case,

SPD could explain the eulachon preference. The occupants of Cathlapotle may not have been alone in feeding their dogs small, abundant marine fish. Grier has argued that the marine isotopic dietary signals from dog bones at the Dionisio Point site in the Salish Sea of southern British Columbia resulted from a diet heavy in herring (*Clupea* sp.), another small fish taken in vast numbers (Grier 2006). Since they do not enter fresh water, herring are at no risk of SPD infection. If the dogs were immune to SPD, the preferential feeding may rather be the result of the same kinds of cultural rules that prohibited dogs' access to salmon fillets and hearts.

The strong discrepancies between the dog isotope values and the zooarchaeological record pose the issue whether the dogs' diet is a close proxy for the diet of Cathlapotle's human occupants who harvested the diverse array of food resources. We suggest the answer is no, it is not, for the reasons developed above. The alternative hypotheses is that the town's occupants did not eat the deer or elk meat they brought in or any of the freshwater fish they harvested in large amounts. We think that unlikely. There are other documented cases where the diets of coresident dogs and humans differed (e.g. Bocherens et al. 2013, Tsutaya et al. 2014). It is for these reasons that we do not see our results as a test of Butler's original hypothesis (Butler 2000).

Our results do not undermine the potential to use dogs as proxies for human diets. Rather our study highlights the importance of using additional data such as documentary and zooarchaeological records, and other lines of evidence, (e.g. Guiry 2012) to evaluate the linkage. It also reinforces that isotopic analyses on a range of

potential prey items and background species in a site is an essential part of establishing the use of dog isotopes as a proxy for human diet. .

7. Conclusions

Our results are consistent with other stable isotope analyses of archaeological dog bones on the Northwest Coast: the dogs were eating a diet dominated by marine fish. However, these results are not consistent with other lines of evidence about human diet at Cathlapotle and in the LCR which suggest a more diverse diet in which marine fish were but one keystone resource, and included numerous terrestrial and freshwater resources. The dogs were not foraging across the human diet nor self-provisioning in the local environment. This points to the strong likelihood the dogs at Cathlapotle were being preferentially fed the marine fish, most likely eulachon (and possibly lamprey) with perhaps the addition of salmon and occasional sturgeon. These results reinforce the need for background isotopic values for a range of animals present in the zooarchaeological collection from a site.

Our study points to the need for better understanding PSD in order to evaluate its effects, if any, on dog diets in the region of North America in which it is endemic. The issue is moot if dogs in the region were immune. If they were not, then it likely shaped what they were fed which has obvious implications for stable isotope studies in the affected region. One particularly pressing issue is determining which fish carried it and which did not.

The aDNA analysis confirmed the osteological identification of the skeletal elements as dog, thus eliminating a potential source of error. We suggest aDNA analysis should also be used to confirm ambiguous identifications increasing the number of specimens available for isotopic analysis. We also believe that in the long run, the population structures of ancient dogs provide a very promising line of evidence about large scale human interactions, and requires a large-scale analysis of pre-contact canid remains to address the issue.

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1087

1088 Table 1 Cathlapotle canid samples, PCR amplification, species identification and

1089 haplotype results.

1090 Table 2 Stable isotope values for archaeological remains of dog and common fauna

1091 from Cathlapotle

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Figure 1 Map of Western North America, showing sites mentioned in the text and the distribution of Cathlapotle Haplotypes as well as other dog haplotypes identified by Barta (2006).

Figure 2. Location of Cathlapotle (45CL1) and the Wapato Valley on the Lower Columbia River in western North America.

Figure 3. Map of Cathlapotle showing positions of the houses, locations of excavation units and units from which the canid elements used here were selected. Map by Emily Shepard.

Figure 4. Pie charts showing percent NISP of different taxa among a) mammals, b) birds and c) and d) fish recovered at Cathlapotle. Mammalian NISP is total NISP recovered in ¼" screens from all 50 excavation units (240 m³) (Lyman 2003). Taxa represented here have NISP >100. The "Other" category includes all taxa with NISP <100. The avian fauna is the analyzed sample recovered in ¼" screens from 21 excavation units (114 m³) (Frederick 2009). The fish fauna is the analyzed sample from 12 10 ltr bulk samples from 12 excavation units (Butler 2002): c) is 4 mm mesh, d) is 2 mm mesh. The bulk samples were water screened in the field and sorted in the lab. The pie charts are based on data presented in Tables S1-S3, Supplemental Materials.

1122
 1123 Figure 5 Phylogenetic tree displaying the relationships between obtained haplotypes
 1124 and some modern *Canis* haplotypes (Savolainen et al. 2002), with coyote (*Canis*
 1125 *latrans*) as the outgroup. Accession numbers and haplotypes are listed for Genbank
 1126 samples. The numbers at the nodes indicate those bootstrap values above 50% after
 1127 2000 replications. Squares represent Cathlapotle Haplotype A3 and triangles Haplotype
 1128 D25.

1129
 1130 Figure 6 Median-joining network comparing Cathlapotle dogs to other ancient domestic
 1131 dog haplotypes discovered in the Americas. Each node depicts a separate D-loop
 1132 haplotype, and node sizes are proportional to haplotype frequencies in the data set, and
 1133 node distances are proportional to the number of mutations between them. Numbers
 1134 beside the nodes indicate haplotypes identified on the Northwest Coast (black) and LCR
 1135 (white).

1136
 1137 Figure 7 Plots of the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values of the Cathlapotle dogs and other taxa
 1138 tested. a) displays the plot of all tested specimen. b) displays the dogs, and taxa with
 1139 marine signatures: salmon, sturgeon and eulachon.

1140
 1141 Figure 8. Box and whisker graphs comparing the isotopic results for the Cathlapotle
 1142 dogs and salmon with Northwest Coast humans (panel a) (see text) and Namu and
 1143 Keatley Creek dogs. a) presents $\delta^{13}\text{C}$ values, b) $\delta^{15}\text{N}$ values. The open circles are the

1144 data points. The midlines in the diamonds mark the medians, the small internal boxes

1145 the means. The lower hinge is at 25%, the upper hinge at 75%.

1146

1147 Ames et al. List of Supplemental Materials

1148 Table S1. NISP of mammalian taxa represented at Cathlapotle (Lyman 2002). The

1149 “Other” category in Figure 4 includes the taxa with NISP less than 100. The total NISP

1150 for “Other” is 379.

1151

1152 Table S2. NISP of avian taxa represented at Cathlapotle (Frederick 2009) used in

1153 Figure 4.

1154

1155 Table S3 NISP of Cathlapotle fish taxa recovered in 4 and 2 mm mesh from 10 l bulk

1156 samples (Butler 2002) used in Figure 4.

1157

1158

Table 1 Cathlapotle canid samples, PCR amplification, species identification, haplotype and stable isotope results

Sample	aDNA Code	Element	Provenience	Map Code ^a	Level	Elevation m ASL	Component	
1	DG1	Astragalus	N106-107/W77-81	4	4	6.35	Post contact	
2	DG2	Femur shaft	N161-163/W104-106	1	4	5.6 - 5.45	Post contact	
3	DG3	Metacarpal	N75-77/W76-78	5	4	6.75	Post contact	
4	DG4	Mandible & teeth	N75-77/W76-78	5	2	6.95	Post contact	
5	DG5	Radius & ulna	N107-109/W98-100	3	8	5.45	Post contact	
6	DG6	Humerus	N107-109/W98-100	3	11	4.9	Pre-contact	
7	DG7	Mandible & maxilla	N107-109/W98-100	3	18	3.1	Pre-contact	
8	DG8	Premolar (P4)	N159-160/W103-107	2	7	5.25	Post contact	

Sample	PCR Sequence ^b	Weight (g)	Species	D-loop Haplotype	Genbank Accession Match	13C/14C	15N/14N
1	0	0.49	<i>Canis lupus familiaris</i>	No amplification	-		
2	6	0.71	<i>Canis lupus familiaris</i>	A3	AF531656	-14	15.2
3	7	0.54	<i>Canis lupus familiaris</i>	A3	AF531656		
4	6	0.41	<i>Canis lupus familiaris</i>	D25	AF005295	-15.1	16
5	4	1.2	<i>Canis lupus familiaris</i>	A3	AF531656	-13.7	17
6	4	0.63	<i>Canis lupus familiaris</i>	A3	AF531656	-15.1	16.5
7	7	0.73	<i>Canis lupus familiaris</i>	A3	AF531656	-14.4	17.2
8	4	0.41	<i>Canis lupus familiaris</i>	D25	AF005295		

^a. See Figure 3.

^b. Indicating how many DNA sequences were obtained from two overlapping DNA fragments.

Table 2 Stable isotope values for archaeological remains of dog and common fauna from Cathlapotle.

Lab number									
(PSU sample number	Species	Element	Component	collagen %	13C/ 14C	15N/ 14N	%C	%N	C:N
Beta 262006									
(PSU 2)	<i>Canis lupus familiaris</i> (Dog)	femur shaft	Postcontact	ND ^a	-14	15.2	ND	ND	ND
Beta 262007		mandible with 2							
(PSU 4)	<i>Canis lupus familiaris</i> (Dog)	teeth	Postcontact	ND ^a	-15.1	16	ND	ND	ND
Beta 262008									
(PSU 5)	<i>Canis lupus familiaris</i> (Dog)	radius and ulna	Postcontact	ND ^a	-13.7	17	ND	ND	ND
Beta 262009									
(PSU 7)	<i>Canis lupus familiaris</i> (Dog)	mandible	Pre-contact	ND ^a	-14.4	17.2	ND	ND	ND
Beta 262010									
(PSU 6)	<i>Canis lupus familiaris</i> (Dog)	humerus	Pre-contact	ND ^a	-15.1	16.5	ND	ND	ND
Anadromous									
Beta 262019	<i>Acipenser</i> (Sturgeon)	1 subopercle		ND ^a	-14.2	16.4	ND	ND	ND
Beta 262020	<i>Acipenser</i> (Sturgeon)	1 sop		ND ^a	-13.4	16.4	ND	ND	ND
S-EVA-	<i>Acipenser</i> (Sturgeon)	scute		0					

16217								
S-UBC 1606	<i>Acipenser</i> (Sturgeon)	scute	0.31	-16.6	14.9	16.6	4.4	4.4 ^b
S-UBC 1607	<i>Acipenser</i> (Sturgeon)	scute	0.75	-17.2	15.5	35.5	9.8	4.2 ^b
S-UBC 1608	<i>Acipenser</i> (Sturgeon)	scute	0.85	-21.3	14.1	11.8	1.8	7.7 ^b
		5 vertebral						
Beta 262017	<i>Oncorhynchus sp.</i> (Salmon)	pieces	ND ^a	-13.8	14.7	ND	ND	ND
		11 vertebral						
Beta 262018	<i>Oncorhynchus sp.</i> (Salmon)	pieces	ND ^a	-14	14.9	ND	ND	ND
S-EVA-								
16210	<i>Oncorhynchus sp.</i> (Salmon)	Vertebrae 2	2.5	-12.4	15.4	35.8	13.1	3.2
S-EVA-								
16211	<i>Oncorhynchus sp.</i> (Salmon)	Vertebrae 2	1.5	-15.9	13.0	15.1	4.9	3.6
S-EVA-								
16212	<i>Oncorhynchus sp.</i> (Salmon)	Vertebrae 2	1.7	-13.7	14.7	30.6	11.0	3.2
S-EVA-								
16213	<i>Oncorhynchus sp.</i> (Salmon)	Vertebrae 3	0.5	-14.5	14.4	17.7	6.1	3.4
S-EVA-	<i>Thaleichthys pacificus</i>							
16218	(Eulachon)	Vertebrae	0					
S-EVA-	<i>Thaleichthys pacificus</i>							
16219	(Eulachon)	Dentary	0					

S-EVA-	<i>Thaleichthys pacificus</i>								
16220	(Eulachon)	Abdom.Vert.		0					
	<i>Thaleichthys pacificus</i>								
S-UBC 1609	(Eulachon)	Vertebrae		1.94	-13.2	13.8	40.4	14.2	3.3
S-UBC	<i>Thaleichthys pacificus</i>								
16010	(Eulachon)	Vertebrae		1.75	-14.2	13.7	40.8	13.9	3.4
	<i>Thaleichthys pacificus</i>								
S-UBC 1611	(Eulachon)	Vertebrae		2.91	-12.6	14.1	43.1	16.0	3.1
Freshwater									
Beta 262021	<i>Anas platyrhynchos</i> (Mallard)	R.d. humerus	Postcontact	ND ^a	-17.8	4.8	ND	ND	ND
Beta 262022	<i>Anas platyrhynchos</i> (Mallard)	I.p. humerus	Pre-contact	ND ^a	-23.3	6.7	ND	ND	ND
S-EVA-	<i>Catostomus macrocheilus</i>								
16214	(Large Scale Sucker)	Maxilla		0.3	-22.9	6.9	38.0	13.4	3.3
S-EVA-	<i>Catostomus macrocheilus</i>								
16215	(Large Scale Sucker)	Dentary		1.6	-23.2	5.2	9.6	3.2	3.5
S-EVA-	<i>Catostomus macrocheilus</i>								
16216	(Large Scale Sucker)	Dentary		0.4	-19.9	6.0	31.3	10.9	3.3
S-EVA-	<i>Ptychocheilus oregonensis</i>								
16207	(Northern Pikeminnow)	Hyomandibula		0.5	-20.3	9.6	32.1	11.1	3.4
S-EVA-	<i>Ptychocheilus oregonensis</i>	Dentary		4.2	-21.4	9.6	38.3	13.6	3.3

16208	(Northern Pikeminnow)								
S-EVA-	<i>Ptychocheilus oregonensis</i>								
16209	(Northern Pikeminnow)	Quadrate		0.3	-19.5	11.1	18.4	6.0	3.6
Terrestrial									
Beta 262015	<i>Cervus elaphus</i> (Wapiti)	Calcaneum	Postcontact	ND	-21.8	3.8	ND	ND	ND
Beta 262016	<i>Cervus elaphus</i> (Wapiti)	d. Tibia. Shaft	Pre-contact	ND	-23.2	4.2	ND	ND	ND
Beta 262011	<i>Procyon lotor</i> (Raccoon)	Humerus	Postcontact	ND	-22	10.2	ND	ND	ND
Beta 262012	<i>Procyon lotor</i> (Raccoon)	Mandible	Pre-contact	ND	-21.8	8	ND	ND	ND
Beta 262013	<i>Ursus americanus</i> (Black Bear)	Humerus	Postcontact	ND	-20.1	5.8	ND	ND	ND
Beta 262014	<i>Ursus americanus</i> (Black Bear)	Ischium	Pre-contact	ND	-21.9	4.5	ND	ND	ND

^a ND Data not reported by laboratory

^b Poor C:N

Ames et al. Stable isotope and aDNA analysis of dog remains from Cathlapotle

(45CL1), a contact-era site on the Lower Columbia River.

Highlights:

- aDNA and $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values from dogs from a fur-trade era site on the Lower Columbia River were analyzed.
- Two haplotypes present among the dogs were patchily present to the north in British Columbia and at Teotihuacan in Mexico.
- Isotopic values indicated a completely marine diet despite ethnohistoric and zooarchaeological data indicating a mixed marine and terrestrial diet.
- The dogs were probably being fed eulachon, a smelt.

Table S1. NISP of mammalian taxa represented at Cathlapotle (Lyman 2002). The “Other” category in Figure 4 includes the taxa with NISP less than 100. The total NISP for “Other” is 379.

Taxon	Common Name	NISP
Cervus	Wapiti	3224
Odocoileus	Deer	2376
Castor	Beaver	392
Procyon	Raccoon	207
Aplodontia	Mtn. Beaver	136
Ondatra	Procupine	106
Ursus	Bear	102
Microtus	Mouse	68
Phoca	Seal	65
Lutra	River Otter	64
Lepus	Hare	50
Canis	Dogs	39
Mustela	Weasel	29
Lynx	Lynx	26
Felis	Mtn. Lion	12
Vulpes	Fox	8
Peromyscus	Deer mouse	5
Sorex	Shrew	4
Scapanus	Mole	3
Mephitis	Skunk	3
Martes	Martin	2
Ovis	Sheep	1
Total		6922

Table S2. NISP of avian taxa represented at Cathlapotle (Frederick 2009) used in Figure 4.

Taxon	Common Name	Cathlapotle NISP
Swans and Geese		
Cygrinae	Swans	32
Anserinae	Geese	52
Ducks		
Anatinae	Dabbling Duck	120
Aythya	Diving Duck	26
Merginae	Mergansers	7
Duck, Undistinguished	Duck	117
Other Water Birds		
Podicipedidae	Grebes	1
Phalacrocoracidae	Cormorants	4
Ardeidae	Hérons	13
Gruidae	Cranes	3
Laridae	Gulls	2
Rallidae	Rails, Gallinules and Coots	1
Raptors		
Accipitridae	Hawks and Eagles	32
Strigidae	Owls	8
Grouse		
Phasianidae	Grouse	7
Perching Birds		
Corvidae	Jays, Magpies, Crows	31
Picidae	Woodpeckers	6
Columbidae	Pigeons and Doves	4
Muscicapidae	Thrushes, Bluebirds etc	2
Emberizidae	Towhees, Sparrows etc	1
Icteridae	Blackbirds, Orioles etc	1
Total Identified Birds		470

Table S3 NISP of Cathlapotle fish taxa recovered in 4 and 2 mm mesh from 10 bulk samples (Butler 2002) used in Figure 4.

Taxon	Common Name	>4 mm	%	> 2 mm	%
Salmonidae	Salmon, Trout, Whitefish	112	32.5	490	19.8
<i>Acipenser</i> sp.	Sturgeon	67	19.4	158	6.4
<i>Thaleichthys pacificus</i>	Eulachon	1	0.3	1374	55.5
Cyprinidae	Minnow	56	16.2	83	3.4
<i>Ptychocheilus oregonensis</i>	Northern pikeminnow	6	1.7	9	0.4
<i>Mylocheilus caurinus</i>	Peamouth	5	1.4	10	0.4
<i>Acrocheilus alutaceus</i>	Chiselmouth			2	0.1
<i>Gila bicolor</i>	Tui chub			1	0.0
<i>Rhinichthys osculus</i>	Speckled dace			1	0.0
Catostomidae	Sucker	27	7.8	52	2.1
<i>Catostomus macrocheilus</i>	Large scale sucker	1	0.3	2	0.1
Cyprinidae/Catostomidae	Minnow/Sucker	51	14.8	236	9.5
<i>Cottus</i> sp.	Sculpin			1	0.0
<i>Gasterosteus aculeatus</i>	Three-spine stickleback	19	5.5	56	2.3
Total		345	100	2475	100