Isotopes in teeth and a cryptic population of coastal freshwater seals

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Impact statement:

Seals residing in Iliamna Lake, Alaska, are the only known freshwater obligate population of the eastern Pacific harbor seal.

Abstract:

Human activities threaten the biodiversity of aquatic mammals across the globe. Conservation of these species hinges on the ability to delineate movements and foraging behaviors of animals, but gaining such insights is hampered by difficulties in tracing individuals over their lives.

We show how isotope ratios in teeth (^{87}Sr/^{86}Sr, ^{13}C/^{12}C, and ^{18}O/^{16}O) reveal lifelong movement and resource use patterns of a unique freshwater population of a wide-ranging pinniped species (the harbor seal, *Phoca vitulina*) that resides in Iliamna Lake, Alaska, U.S.A. This population's potentially unique migratory behavior and use of different trophic resources is unknown even in the face of proposed large-scale industrial development and a rapidly changing climate. Using isotopes in teeth, we show that seals are born in the lake, remain lifelong residents, and rely principally on resources produced from within the lake even during periods when seasonally abundant and nutrient dense anadromous fish are available (i.e., sockeye salmon *Oncorhynchus nerka*). We present the Iliamna Lake harbor seals as a unique case study into how serial isotope records in teeth, particularly ^{87}Sr/^{86}Sr ratios, reveal the cryptic ecology of an aquatic mammal population residing in an intact ecosystem. The insights we gain are relevant to conservation problems of threatened aquatic mammals across the globe, including those in some of the world's most altered coastal ecosystems, but also provide critical baseline information for the Kvichak River system facing an uncertain future due to a rapidly changing environment.

Introduction:

Coastal marine, estuarine, and freshwater mammals are threatened globally (Pompa et al. 2011). In particular, species and populations that migrate between estuarine and freshwater habitats, or that are freshwater-dependent, include some of the most endangered mammals worldwide (He et al. 2018). At least one extinction has occurred during the last decade (the Yangtze River dolphin or baiji, *Lipotes vexillifer*) (Turvey et al. 2007; Veron et al. 2008). Major threats to their persistence are directly due to human activity (Veron et al. 2008; He et al. 2018). Small and large hydropower dams are proliferating across the globe driving habitat fragmentation and changes to natural flow regimes with little consideration of consequences for aquatic mobile species, particularly mammals (Sabo et al. 2017; Couto & Olden 2018). Riverine and estuarine ecosystems also act as sinks for pollution generated by industrial and agricultural activities of densely populated regions posing significant risks to top predators that bioaccumulate toxins, such as cetaceans (Veron et al. 2008; Sinha & Kannan 2014). Incidental catches of endangered riverine and estuarine mammals during fisheries continue to threaten populations in some of the most intensely impacted rivers and estuaries of the world, e.g. the Gulf of California, Ganges (Sinha & Kannan 2014), Mekong (Smith & Jefferson 2002), Yangtze (Turvey et al. 2007), Amazon, and Orinoco (Gomez-Salazar et al. 2012) River basins.

The conservation of highly mobile species, including coastal aquatic mammals, is dependent on knowledge of complex life histories, population connectivity, and foraging behaviors (Runge et al. 2014). Unfortunately, for many species and populations these fundamental features of their biology remain unknown. This is because tracing the movements and foraging of individuals over the course of their lives is rarely possible, especially for rare and elusive species. Insights are often limited to studies based on visual observations or tagging efforts that yield relatively brief snap shots in time (Gomez-Salazar et al. 2012). Nonetheless, in order to ensure the persistence of such populations in the face of multiple environmental pressures, effective conservation plans must be able to identify

critical habitats and trophic resources, how these vary through time, and how they vary among life stages.

We quantified the lifelong movement and foraging ecology of a unique and small population (~400 individuals) of a wide-ranging pinniped species (the Eastern Pacific harbor seal, *Phoca vitulina richardii*) that resides in Iliamna Lake, Alaska, USA (Figure 1) using serial isotope records in teeth. Like other cryptic aquatic mammals across the globe, the Iliamna harbor seal's migratory and foraging behaviors are largely unknown (Burns et al. 2016). But, unlike most threatened populations, the Iliamna seals live in a relatively undisturbed ecosystem. Therefore, they provide a rare opportunity to gain insights into how populations that are at least in part freshwater-dependent exploit the heterogenous landscapes and trophic resources characteristic of coastal ecosystems, and the extent to which they are connected with proximate marine-dependent populations. It is not known whether these seals migrate between the lake and ocean (an ~80km swim); nor is it known to what extent seals rely on trophic resources produced from within the lake versus the ocean (e.g., seasonally abundant sockeye salmon, *Oncorhynchus nerka*). Thus, this case study is relevant to many of the world's most threatened populations of aquatic mammals, but also provides critical baseline information for the Kvichak River (Figure 1). This watershed is under increasing pressure from proposed large-scale mining activities within its headwaters (EPA 2014) and a rapidly changing climate. Iliamna Lake harbor seals are currently managed as if they are part of the nearby Bristol Bay marine population (Muto et al. 2017). The insights revealed here challenge whether this is an appropriate management strategy.

Isotope tracers in sequentially growing teeth are a powerful method for unraveling lifelong movement and resource use of elusive animal populations (Kohn & Cerling 2002; Koch 2007; Newsome et al. 2010). Because enamel in mammalian teeth is laid down sequentially during an animal's first years of life (Koch 2007) and dentine grows continuously over the animal's lifetime where layer thickness correlates with body size (Boyd & Roberts 1993; Hoffman et al. 2010), teeth yield lifelong chemical records of an animal's diet and movements. Stable isotope ratios, expressed as $\delta^{18}O$, $\delta^{13}C$, $\delta^{15}N$, and $\delta^{34}S$, are commonly used to study isotopic gradients within and among food webs and habitats of terrestrial, aquatic, and marine ecosystems. Strontium (Sr) isotope ratios ($^{87}Sr/^{86}Sr$) represent a potentially useful tracer to understand movement and resource use patterns of aquatic mammals among freshwater and marine systems. But their use has been limited to a study on the paleoecology of extinct Miocene desmostylians (Clementz et al. 2003) and to a population of endangered beluga whales (Nelson et al. 2018). No study, however, has quantified two critical aspects of Sr biogeochemistry necessary to accurately interpret how $^{87}Sr/^{86}Sr$ ratios inform habitat and resource use of aquatic mammals among freshwater and marine ecosystems: isotopic changes of migratory prey (e.g., diadromous fishes) and concentration dependence. These are particularly important for coastal omnivores and carnivores, such as bears, cetaceans, and pinnipeds known to prey on diadromous fishes during their migrations among freshwater and marine systems. If left unconstrained, they can mislead interpretations of how $^{87}Sr/^{86}Sr$ ratios inform freshwater versus marine habitat and resource use. Nonetheless, much of the world's coastal freshwater habitat is characterized by $^{87}Sr/^{86}Sr$ ratios that differ substantially from their proximate marine ecosystems. The world's oceans are isotopically homogenous ($^{87}Sr/^{86}Sr$=0.70918 ± 0.00006) (Faure &

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Mensing 2005). Due to their tectonic setting along continental margins or ancient shield terranes, many rivers draining regions, such as the Pacific Rim, Amazonian Craton, and the Tibetan Plateau, carve through rocks with $^{87}Sr/^{86}Sr$ ratios that differ widely from the ocean (Palmer & Edmond 1992). They are also home to the world's most threatened mammal populations (Pompa et al. 2011). Given the ubiquity and predictability of such freshwater-marine isotopic gradients across the globe, $^{87}Sr/^{86}Sr$ ratios could be valuable for informing conservation efforts in coastal, estuarine, and riverine ecosystems worldwide. $^{87}Sr/^{86}Sr$ ratios are an established and powerful tool in fish and terrestrial mammal ecology, but their use to study the movement and trophic resource use patterns of coastal mammals is relatively unexplored.

To quantify the lifelong movements and foraging ecology of Iliamna Lake harbor seals, we measured multiple isotope ratios ($^{87}Sr/^{86}Sr$, $^{13}C/^{12}C$, and $^{18}O/^{16}O$) in the enamel and dentine of their canine teeth and characterized the $^{87}Sr/^{86}Sr$ ratios and Sr concentrations [Sr] (mg/kg) of potential trophic resources. Migratory diadromous fishes are a common trophic resource of coastal mammals and when exploited they can motivate movements among these ecosystems. Such fishes are also only ephemerally available, so understanding their relative importance to consumer energy budgets is crucial to understanding the ecosystem processes they support. To this end, we quantified the $^{87}Sr/^{86}Sr$ ratios and [Sr] of resident lake fishes and determined whether the isotopic ratio of the seasonally abundant adult sockeye salmon returning to Iliamna Lake changed over the course of their spawning migration. The physiological transformation that diadromous fish undergo when they migrate between marine and freshwater systems should induce isotopic variation among tissues (e.g., gonads, muscle, and blood) related to tissue turnover rates. Thus, we analyzed Sr concentrations and $^{87}Sr/^{86}Sr$ ratios in eggs, muscle, and blood from female sockeye salmon over the course of a spawning run (SI Appendix, Figure S1, Dataset S1). Constraining time-dependent and tissue-specific $^{87}Sr/^{86}Sr$ changes in sockeye salmon was critical because consumers across the Pacific Rim exhibit selective feeding on anadromous fishes, including among different body parts (Gende et al. 2001; Hauser et al. 2008), and at different times over the course of a spawning run (Schindler et al. 2013; Deacy et al. 2016). Once constrained, the degree to which sockeye salmon isotopically changed over the course of spawning enabled us to test different hypotheses regarding (*i*) whether Iliamna harbor seals migrate to the ocean, and (*ii*) the extent to which they exploit trophic resources produced from within the lake versus the ocean. Furthermore, these time-dependent salmon tissue analyses demonstrate important aspects of the broader applicability of using the $^{87}Sr/^{86}Sr$ isotope system to delineate the migratory and foraging patterns of coastal mammals.

Methods:

We measured $^{87}Sr/^{86}Sr$, $^{18}O/^{16}O$, and $^{13}C/^{12}C$ ratios in the dentine and enamel of canine teeth of four Iliamna harbor seals, and three marine harbor seals from proximate populations within Bristol Bay (SI Appendix, Dataset S2). Iliamna Lake seal samples came from subsistence harvests and were collected in collaboration with the Alaska Native Harbor Seal Commission and the Bristol Bay Native Association and analyzed under Marine Mammal Protection Act Permit #15510 issued to JMB. Iliamna seal ages were determined in the cementum of teeth (Burns et al. 2016). Teeth from marine seals came from University of Alaska's Museum of the North. Dentine was collected from the

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outside edge of each tooth towards the pulp cavity via ~100μm-wide drill paths drilled parallel to visible growth layers using a microdrill (~30 samples/seal) (SI Appendix, Figure S2). Using a Dremel, we milled enamel from three locations: the enamel-dentine junction near base, the middle, and apex of each tooth. All $^{87}Sr/^{86}Sr$ ratios, including salmon tissues, were measured via multi-collector inductively coupled plasma mass spectrometry (MC-ICPMS); $^{18}O/^{16}O$, and $^{13}C/^{12}C$ were measured via isotope ratio mass spectrometry (IRMS) using standard methods (SI Appendix). [Sr] of fish tissues were analyzed via single collector ICPMS. We digested all tissues prior to [Sr] and $^{87}Sr/^{86}Sr$ analyses (SI Appendix).

Because of the large differences in [Sr] between the marine and freshwater lake system, we used concentration-dependent mass-balance equations (Phillips & Koch 2002) to evaluate the degree to which anadromous sockeye salmon contribute to the assimilated biomass of Iliamna harbor seals (SI Appendix). To account for the potentially high spatial and temporal uncertainty in $^{87}Sr/^{86}Sr$ ratios and [Sr] of spawning sockeye salmon in the lake, we evaluated four mass-balance scenarios differing in how the adult salmon endmember was characterized geochemically (Table S1, SI Text). The marine endmember within these scenarios ranged from fully marine to near-senescent salmon (SI Appendix). In all scenarios, the lake endmember was based on measurements of resident fishes within lake (SI Appendix).

Results:

Sockeye salmon and resident fish tissues:

The $^{87}Sr/^{86}Sr$ composition and [Sr] of sockeye salmon tissues changed significantly over the course of the spawning run and the rate and magnitude of the change was tissue dependent (Figure 2, S3-S4, and SI text).

Isotopes in teeth:

Early in the lives of all seals analyzed, $^{87}Sr/^{86}Sr$ ratios in dentine and $\delta^{18}O$ values in enamel reflected values consistent with Iliamna Lake (0.7049-0.7059 and 16.2-17‰ [VSMOW], respectively) (Figure 3, Dataset S2). Later in life, however, $^{87}Sr/^{86}Sr$ ratios in tooth dentine of all four individuals increased to ratios intermediate (0.7065-0.7085) of the global marine value and Iliamna Lake (Figure 3a). Enamel $\delta^{13}C$ values of all four seals ranged from -17.9‰ to -13.2‰ (VPDB), consistent with other freshwater aquatic mammals (Figure 3b). The 16-year-old seal had more enriched enamel $\delta^{13}C$ values than the other seals (5-6 years old). $^{87}Sr/^{86}Sr$ ratios in dentine and enamel of Bristol Bay seals ranged between 0.70911-0.70926 reflecting the global marine value (0.70918±0.00006) (Figure 3a, Dataset S2). $\delta^{18}O$ values in enamel of marine harbor seals from Bristol Bay ranged from 22.2-24.9‰ (VSMOW); $\delta^{13}C$ values ranged from -10.9 to -8.4‰ (VPDB) (Figure 3b).

Mass-balance of resource use:

Fractional contributions of salmon to seal diets varied depending on whether the salmon assimilated was: (*i*) fully marine, (*ii*) had just entered freshwater, (*iii*) was just prior to spawning, or (*iv*) spawned out (Figures 4 and S3, SI Text, and Table S1). These differences were due to rapid

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changes in the ^{87}Sr/^{86}Sr ratios and [Sr] of spawning adult sockeye salmon after entering freshwater (Figure 2a, Tables S1 and S3). From the perspective of assimilated biomass (Figure 4a-e), scenarios where seals ate either fully marine salmon or salmon just entering freshwater (*i* and *ii*) estimated fractional contributions of salmon to be <23%, including the periods later in life. Contributions estimated later in life by scenarios *iii* (salmon just entering freshwater) and *iv* (spawned out salmon) ranged from 10-100%. Early in seal's lives, however, fractional contributions estimated via all scenarios from eating salmon were estimated as <10% assimilated biomass (Figures 4 and S3). Due to relatively high [Sr] in adult salmon tissues, fractional contributions to biomass were always lower than those estimated for the elemental mass of Sr (Figure S4, SI Appendix).

Discussion:

Iliamna seals are born in lake and remain lifelong residents:

Isotopes in teeth indicate that adult seals were born in Iliamna Lake and remained lifelong residents. ^{87}Sr/^{86}Sr ratios in tooth dentine, and δ^{18}O and δ^{13}C values in enamel, of four Iliamna seals revealed general coherence among the early life history patterns of individuals (Figures 3 and 4). During this period, δ^{18}O and δ^{13}C values in enamel fell within ranges consistent with the enamel of freshwater aquatic mammals (Figure 3b) (Clementz & Koch 2001). Similarly, ^{87}Sr/^{86}Sr ratios recorded in the earliest dentine layers also reflected freshwater residence (Figure 3a).

The lifelong residence of Iliamna harbor seals in the lake is inferred on the basis of evaluating different mass-balance scenarios, which indicated that given high [Sr] of salmon prior to entering freshwater, it was unlikely that seals had a fully marine diet. If seals migrated to the ocean down the Kvichak River they would have been required to of fed entirely on marine resources. Assimilation of even relatively small amounts of such resources would have resulted in tooth ^{87}Sr/^{86}Sr ratios that reflected the marine value (Figure 4e). ^{87}Sr/^{86}Sr ratios in teeth were always <14% fully 'marine' salmon. We assessed whether such a fraction could be explained by short migrations to the ocean by computing the expected ^{87}Sr/^{86}Sr ratios in teeth if seals assimilated a mixture of fully 'marine' salmon, 'just prior to spawning salmon', and resident lake fish during the approximately two-month period corresponding to each 100μm-drillpath making up each tooth analysis (Figure 4e, SI Text). We estimated that the maximum amount of time during this period that a seal (maximum ^{87}Sr/^{86}Sr ratio=0.7085) could have spent eating fully marine fish in the ocean or estuary would have been ~2 weeks (0.14x2months, Figure 4e, SI Text). This also assumes that while the seal was in the lake during the remainder of this period it only ate resident lake fish; if instead it also ate spawning salmon, the length of such a foray to the ocean would have been correspondingly shorter (Figure 4e, SI Text). Although such migrations seem biologically unrealistic due to their short duration and associated energetic cost, they are within the range of movements of marine populations (Lowry et al. 2001).

Foraging ecology of Iliamna seals:

Isotopes in dentine suggest that earlier in life, seals relied principally on lake food resources; later in life, seals shifted to rely more heavily on the seasonally abundant sockeye salmon. All mass-

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balance scenarios indicated that adult sockeye salmon contributed a small proportion of the Sr (Figure S4) and biomass (Figure 4) assimilated by Iliamna seals early in life; δ^{13}C ratios in enamel also indicated a primarily freshwater diet during this period (Figure 3b). All individuals indicated a large shift in ^{87}Sr/^{86}Sr ratios later in life towards the global marine ratio (Figures 3a and 4). This isotopic change likely reflects an ontogenetic shift in diet towards increased consumption of salmon. Similar shifts have been documented in coastal bear populations (Van Daele et al. 2013), and also for estuarine bottlenose dolphins (Rossman et al. 2015a). Iliamna seals are almost always seen at the east end of the lake on a series of islands (Figure 1) (Burns et al. 2016). Sockeye salmon are likely most vulnerable to seal predation once they start actively spawning along the island beaches. Sockeye salmon enter the Kvichak River in July and spawning in the lake occurs at the end of August. Actively spawning salmon have likely spent more time in freshwater than those that are holding. Thus, the salmon most vulnerable to seals would tend to have a geochemical composition reflecting salmon that have been in freshwater for an extended period (e.g., scenarios *iii* and *iv*, respectively) (Table S1). Observations of seal scat and seal-killed salmon on these islands indicate that during the spawning season, sockeye salmon represent a major diet source for seals (Hauser et al. 2008). But, seals often consumed only the belly of female fish (>50% of females), presumably seeking eggs, and typically consumed the whole body of male salmon (>95% of males). Sex ratios of sockeye salmon spawning on these island beaches have been reported as approximately 0.9:1 (female:male) (Stewart et al. 2003), but surveys of seal-killed salmon had sex ratios ranging from approximately 1.2:1 to 5.4:1 across three years (Hauser et al. 2008). Taken together, this suggests that the salmon eaten by seals may be disproportionately pre-spawned actively spawning female salmon (e.g., scenario *iii*). Such selective feeding behavior has been documented for other mobile consumers in salmon ecosystems and is thought to reflect animals seeking to optimize nutrient density and the energetic costs of metabolism (Gende et al. 2001). Given that ^{87}Sr/^{86}Sr ratios early in the life of seals show little influence from marine sources, it is unlikely that young seals consume large amounts of salmon prior to spawning, as well as those near-senescence (Figure 4).

Even late in life, sockeye salmon contribute 10% (minimum of scenario *iii*) to 100% (maximum of both scenarios *iii* and *iv*) to the assimilated biomass of seals. This large range later in life is in part due to differences among individual seals (Figure 4), suggesting some seals rely more heavily on sockeye salmon (seal03 and seal07), while others do to a lesser extent (seal02 and seal04). Individual specialization of foraging habits has been documented for other generalist consumers, such as coastal cetaceans (Rossman et al. 2015b), sea otters (Estes et al. 2003), and bears (Deacy et al. 2016). The food resources produced from within the lake available to seals include abundant juvenile sockeye salmon and a diverse array of resident fishes, all of which would isotopically reflect lake water (Figures 2 and 3). Furthermore, gut and scat contents of seals contained resident fishes even during periods when spawning sockeye salmon were available (Hauser et al. 2008; Burns et al. 2016). The coherent ontogenetic switch to relying more heavily on sockeye salmon later in life may reflect that young seals are unable to effectively hunt adult salmon prior to spawning when fish are still spry.

The estimates of dietary contributions reported here may also be affected by Iliamna seals drinking lake water, which would constitute a source of freshwater Sr in addition to fishes produced

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from within the lake that contributed to the total assimilated mass by Iliamna seals. No study has constrained the degree to which drinking water influences the Sr recorded in teeth of aquatic mammals. Freshwater drinking is thought to be limited in pinnipeds because they achieve water balance via metabolic and dietary sources of water (Ortiz 2001). Studies of terrestrial mammals that do regularly drink freshwater indicate that drinking water contributes <10% of the assimilated biomass (Lewis et al. 2017). Because Iliamna Lake water is isotopically very similar to its resident fish (Figure 2 and 3), then a proportion of the assimilated mass of seals from lake resources could be from drinking water. However, this proportion is probably negligible given the low [Sr] in Iliamna Lake.

Using $^{87}Sr/^{86}Sr$ to quantify marine versus freshwater resource use of consumers:

Although $^{87}Sr/^{86}Sr$ ratios have been used extensively to study the ecology of migratory fishes and terrestrial mammals, their use to study how coastal mammals differentially use marine and terrestrial ecosystems is relatively unexplored, especially compared to the lighter stable isotope systems ($^{15}N/^{14}N$, $^{13}C/^{12}C$, and $^{34}S/^{32}S$). This study demonstrates the importance of two critical aspects of using $^{87}Sr/^{86}Sr$ ratios to quantify migration and trophic resource use of coastal mammals among ecosystems: concentration dependence and tissue turnover of migratory prey. In coastal areas situated along continental margins (e.g., Pacific Rim) or shield terranes (e.g. Laurentian Plateau and Amazonian Craton), marine environments have much higher [Sr] compared to proximate freshwater and terrestrial ecosystems (Palmer & Edmond 1992). This contrast in [Sr] results in high sensitivity of $^{87}Sr/^{86}Sr$ to discern marine resource and habitat use of principally terrestrial or freshwater coastal consumers (e.g., bears). However, this [Sr] gradient yields less sensitivity to discern freshwater resource and habitat use of principally marine consumers. The mass-balance scenarios here illustrate that small fractions of the assimilated biomass from marine resources with high [Sr] (<5%) will correspond to large $^{87}Sr/^{86}Sr$ isotopic changes recorded in teeth (Figure 4e). Thus, even ephemeral periods of marine resource use by freshwater or terrestrial consumers can be detected. However, $^{87}Sr/^{86}Sr$ will be less sensitive for quantifying freshwater habitat and resource use of marine consumers because they would have to assimilate large amounts of freshwater resources in order to detect isotopic changes in their teeth. Furthermore, when coastal mammals exploit populations of diadromous fishes during their migrations between ocean and freshwater, this food resource will undergo rapid isotopic change. Therefore, $^{87}Sr/^{86}Sr$ ratios in teeth of principally marine consumers known to target anadromous fishes during spawning migrations (e.g., beluga whales (Nelson et al. 2018)) should be interpreted with caution, as departures from the global marine value could be due to feeding on a marine resource that partially reflects the freshwater isotopic endmember (e.g., 'bright' salmon, Table S1). It is imperative to constrain such processes (i.e., concentration-dependence and isotopic changes of migratory prey) with respect to $^{87}Sr/^{86}Sr$ ratios.

Implications for conservation:

Using serial isotope records within teeth, we quantified two fundamental biological dimensions of an elusive freshwater-dependent pinniped population. These insights have important implications for their conservation. The patterns we revealed for four seals are suggestive for the

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population overall, but are still subject to the issues of a small sample size. Although small, it corresponds to 1% of the population (~400), which is a relatively large sampling density in ecological studies, including of Alaska's harbor seal genetic population structure (~0.3%) (Muto et al. 2017). Nonetheless, Iliamna seals clearly exhibit distinct patterns of habitat and resource use compared to proximate and closely related marine harbor seals in Bristol Bay. The absence of migratory connectivity among these populations further emphasizes the need to protect Iliamna seals as an ecologically unique population, as endemic populations are more vulnerable to environmental change (Ceballos & Ehrlich 2002; Pompa et al. 2011). Furthermore, because Iliamna seals are principally dependent on lake resources, especially early in life, responses of this population to environmental change will most likely differ from that of Bristol Bay marine populations. Rapidly warming temperatures and increasing pressure of proposed mining activities in the region (EPA 2014) further emphasize the need to establish baseline ecological information and an effective conservation plan for this population.

A recent expert review of the conservation status of Iliamna harbor seals found that this population was genetically distinct, but due to a paucity of quantitative genetic, morphometric, and ecological data it was ambiguous about whether this population was 'significant' to the broader *P. v. richardii* taxon (Boveng et al. 2016). This review identified the need for data that is able to elucidate whether Iliamna seals have adapted to their unique environment as a measure of its genetic and ecological significance to the broader taxon. Adaptation of unique foraging behaviors in unusual ecological settings can constitute such a measure of significance, as in the case of the island-associated Hawaiian false killer whales (Oleson et al. 2010). The unique foraging ecology of Iliamna seals, as shown here, supports the conclusion that this population has adapted to their unusual ecological setting. This includes their reliance on lake resources and consistent ontogenetic shift from a diet composed principally of lake resources to one that exploits seasonally abundant salmon. Both imply locally-adapted abilities to exploit a food web unlike that of any other *P. v. richardii* population across the Eastern Pacific, which are significant to the evolutionary potential of the broader taxon.

The endemism and unique foraging ecology of Iliamna Lake harbor seals is important because wide-ranging generalist consumers (bears, pinnipeds, and cetaceans) are predicted to fare better than spatially restricted specialist consumers in the face of environmental change due to their increased ability to integrate across variable habitats and resources (Miller-Rushing et al. 2010). Food resources are often only available for short periods of time and their productivity can exhibit high spatial heterogeneity (Armstrong et al. 2016). Mobility, plus the ability to integrate across resources that exhibit asynchronous and complimentary temporal dynamics, enables generalist consumers to exploit emergent portfolio effects of ecosystems (the dampened variance typical of aggregate scales of complexity due to weak covariance at finer scales). Such effects buffer consumer populations from variability of single prey populations and extend the period of time they are available. Run-timing varies by two weeks among sockeye salmon stocks returning to the major rivers of Bristol Bay, Alaska. These stocks also exhibit ~70% more inter-annual variability on average than the aggregate of all Bristol Bay stocks (Schindler et al. 2010). Coastal marine mammal populations have the ability to integrate across this spatial and temporal variability, which makes the

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resource more reliable (Schindler et al. 2010). However, consumer populations restricted within a single basin – e.g., Iliamna Lake – would not be able to exploit these portfolio effects in the same way. Iliamna sockeye stocks were the most abundant stocks prior to the mid-1990s in Bristol Bay, Alaska, but returns through the mid-2000s were some of the lowest on record (Hilborn et al. 2003). Harbor seals endemic to Iliamna Lake are subject to these large shifts; whereas their closely related marine populations are not. The stability of the population over this period (Boveng et al. 2018) may in part be due to the Iliamna seal's ability to integrate across lake and marine resources.

The insights we gained into the cryptic ecology of Iliamna's seals are relevant to other transient and obligate freshwater coastal mammals across the globe – many of which constitute the world's most threatened mammal species and populations. The Iliamna seals clearly integrate across trophic resources produced from disparate ecosystems. The degree to which one ecosystem versus another supports their energy budget depends on life stage and also the individual. The differential exploitation of resources among life stages reflects the fact that particular prey sources are needed or preferred during different times (Van Daele et al. 2013; Rossman et al. 2015a). Similarly, individual foraging behaviors suggests that individuals adopt various feeding strategies in the face of spatially and temporally dynamic prey sources (Estes et al. 2003; Rossman et al. 2015b; Deacy et al. 2016). In both instances, there is an important interaction between foraging strategy, trophic resources, and the role of heterogeneity. This highlights why conservation of ecosystem heterogeneity and the processes that generate it is essential to the persistence of consumer populations.

In addition to changing overriding climatic conditions, human activities across the globe are degrading (e.g., via pollution), reducing, and simplifying (e.g., fragmentation of habitats via dams) the habitats of aquatic mammals. Conservation in the face of such environmental change depends on identifying critical habitats and trophic resources over the lives of individuals. Many of the most threatened mammal populations, including those within the largest and most highly impacted rivers and estuaries of the world, use coastal marine, estuarine, and riverine habitats and resources that are defined by predictable isotopic gradients. Thus, isotopes in sequentially growing teeth represent a promising way to elucidate fundamental aspects of their ecology. We show here that isotopes in teeth, in particular $^{87}Sr/^{86}Sr$ ratios, are a powerful approach to understanding the movement and foraging ecology of these cryptic aquatic mammals.

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Supplemental Information:

The Supplemental Information associated with this manuscript is available as an online appendix, including details of analytical and modeling methods and full data tables.

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Figures Legend:

Figure1: Iliamna Lake in Southwest Alaska, U.S.A., the Kvichak River basin, and the range of Eastern Pacific harbor seals. The seals in Iliamna Lake are the only known freshwater-associated population of this subspecies of harbor seals.
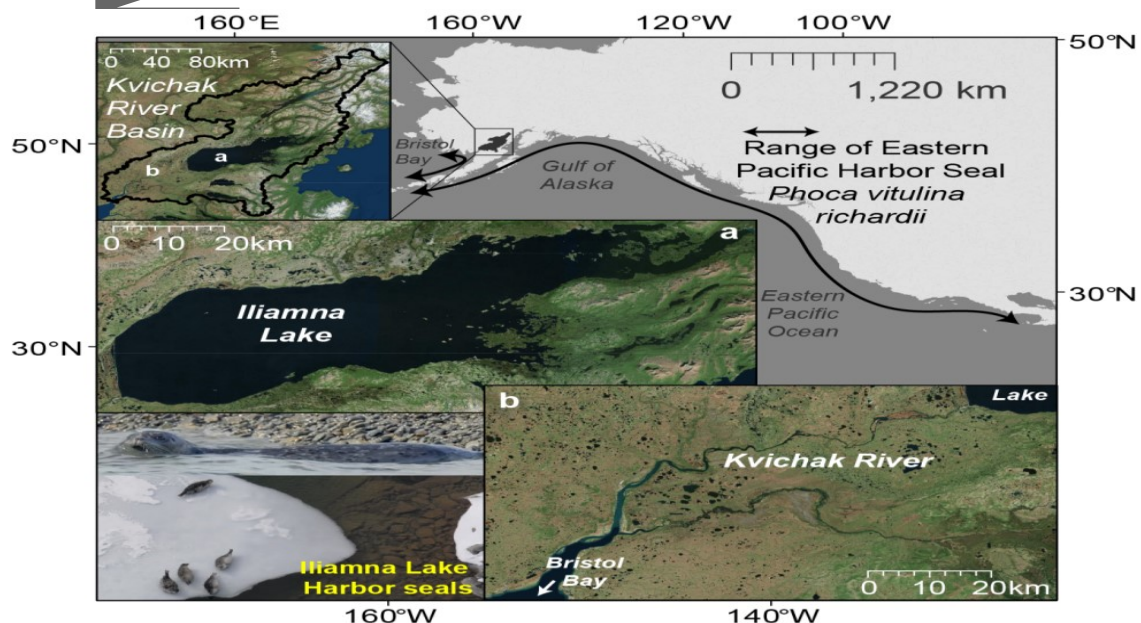

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Figure2: [Sr] and ^{87}Sr/^{86}Sr ratios in tissues of adult sockeye salmon and resident lake fish over the course of the spawning run (A). Resident fish (black-filled circles) are not scaled by [Sr], as these analyses includes [Sr] in muscle and vertebrae of char, and whole stickleback. [Sr] (B) and ^{87}Sr/^{86}Sr (C) ratios in different tissues adult sockeye salmon and lake water; these distributions represent samples collected across the duration of the spawning migration.
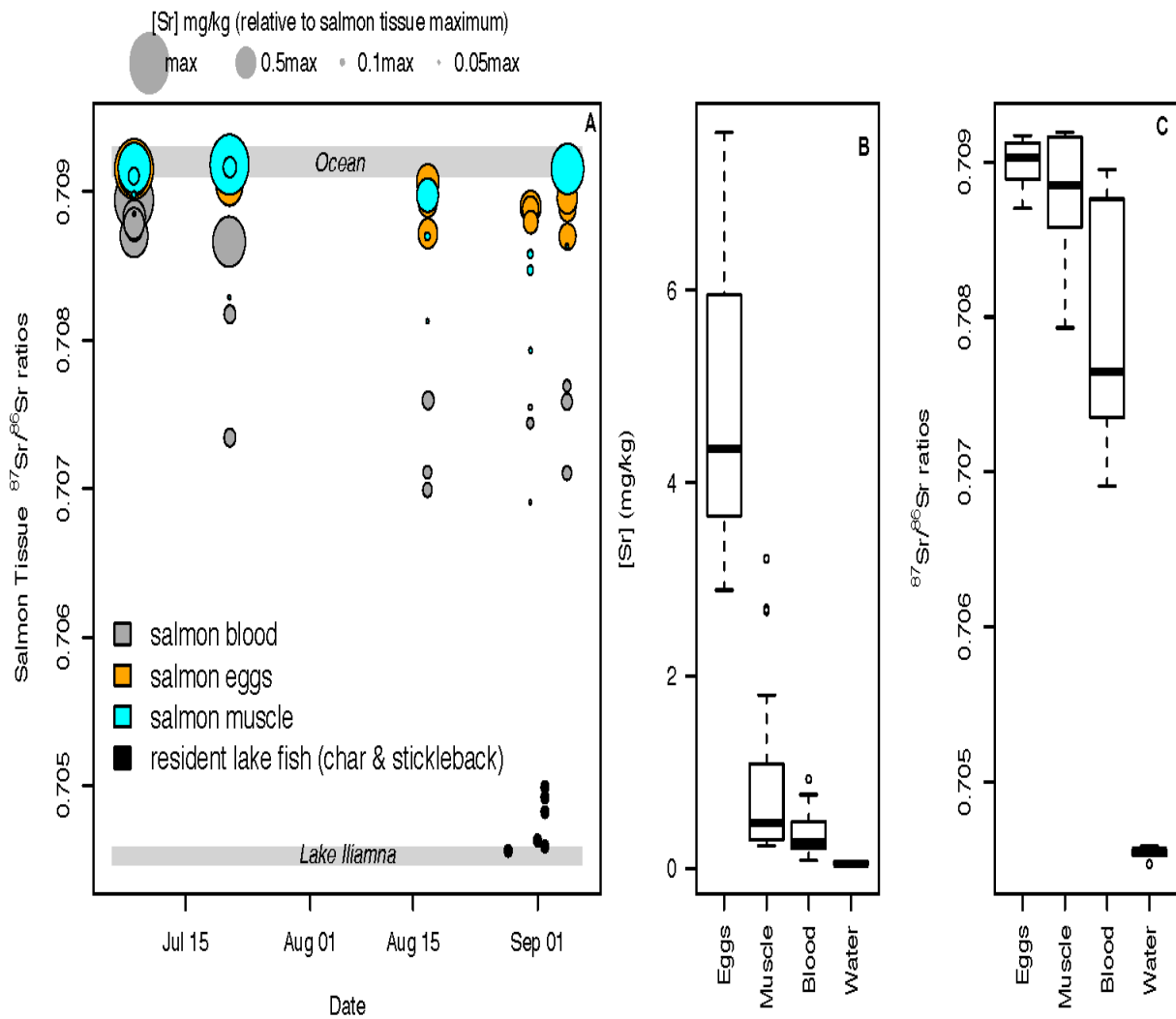
Figure3: [87]Sr/[86]Sr profiles (A) in dentine of Iliamna and marine harbor seals. The range of marine harbor seal (n=3 seals) enamel, dentine, and ocean water (Dataset S2), and that of Iliamna Lake fish and water is indicated by light and dark gray polygons, respectively. δ^{13}C and δ^{18}O (B) of seal teeth in enamel. Black-lettered distributions are from Clementz and Koch (2001) of enamel from offshore (OS), nearshore (NS), estuarine (E), and freshwater (FW) aquatic mammals from California and Oregon, USA.
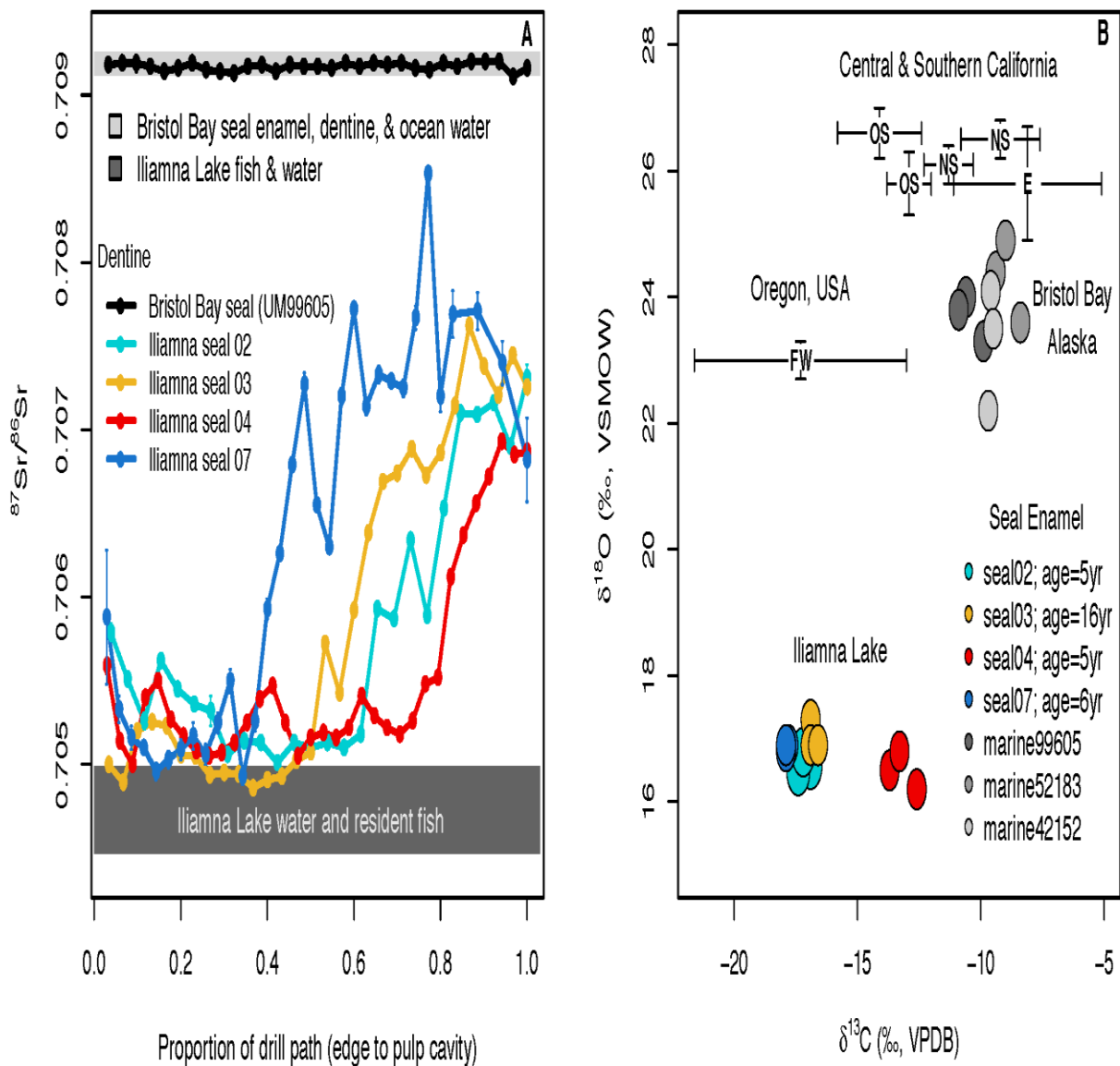
Figure4: Different scenarios of the proportion of adult sockeye salmon biomass assimilated by Iliamna seals if seals fed solely on 'marine', 'bright', 'just prior to spawning', or 'zombie' salmon versus lake resources (A-D). Endmembers for each mass balance scenario are described in SI Appendix (SI Text, Table S1). Expected $^{87}Sr/^{86}Sr$ ratios in teeth (E) based on concention-wieghted mass balance if seals ate a mixture of fully marine salmon (fm), spawning salmon (fs), and resident lake fish (fl). This illustrates the overwhelming influence marine resources can have on $^{87}Sr/^{86}Sr$ ratios recorded in seal teeth due to concentration differences among ocean and lake even in scenarios where seals eat relatively small amounts of these resources.