

ORIGINAL ARTICLE OPEN ACCESS

Do Snow-Adapted Prey Facilitate Coexistence of the Sierra Nevada Red Fox With Sympatric Carnivores?

Grace Rosburg-Francot¹  | Catherine B. Quinn^{1,2}  | Stevi Lee Vanderzwan¹  | Tal Caspi¹  | Pete Figura³ | Benjamin N. Sacks^{1,4} 

¹Mammalian Ecology and Conservation Unit of the Veterinary Genetics Laboratory, School of Veterinary Medicine, University of California, Davis, Davis, California, USA | ²National Genomics Center for Wildlife and Fish Conservation, USDA Forest Service, Rocky Mountain Research Station, Missoula, Montana, USA | ³California Department of Fish and Wildlife, West Sacramento, California, USA | ⁴Department of Population Health and Reproduction, School of Veterinary Medicine, University of California, Davis, Davis, California, USA

Correspondence: Benjamin N. Sacks (bnsacks@ucdavis.edu)

Received: 21 February 2025 | **Revised:** 8 July 2025 | **Accepted:** 11 August 2025

Handling Editor: Carla Martins Lopes

Funding: Funding for the metabarcoding project was provided from the Christine Stevens Wildlife Award to B.N. Sacks and supplemented through funds of the Mammalian Ecology and Conservation Unit of the Veterinary Genetics Laboratory at the University of California, Davis. Collection of samples from the field and DNA analyses to identify species were supported by the U.S. Fish and Wildlife Service (F16AC0054), U.S. Department of Agriculture (USDA) Forest Service Pacific Southwest Region (12-CS-11052007-021, 16-CS-11051600-023), USDA Forest Service Intermountain Region (11-CS-11041702-046, 16-CS-f 1041730-034), U.S. National Park Service Yosemite National Park (CA-CESU Agreement P15AC01816) and California Department of Fish and Wildlife (P1080019).

Keywords: climate change | competition | coyote | diet | metabarcoding | Sierra Nevada red fox

ABSTRACT

Specialist species in alpine ecosystems may be increasingly threatened by climate-driven habitat loss and encroachment by generalist competitors. Ecological theory predicts that niche differentiation through dietary specialisation can facilitate coexistence with generalist competitors. We quantified dietary overlap between a high-elevation specialist, the Sierra Nevada red fox (SNRF; *Vulpes vulpes necator*) and a widespread generalist, the coyote (*Canis latrans*), as well as other sympatric carnivores. We were especially interested in dietary items that were themselves specialised to alpine habitats, as we expected them to be most critical to SNRF. To characterise diet, we used DNA metabarcoding for vertebrate and plant-based food items of 789 carnivore scats collected from the sites of two SNRF populations (Lassen, Sierra Nevada). As expected for potential competitors, SNRFs exhibited substantial dietary overlap with coyotes overall. Dietary niche overlap was lower between SNRF and both bobcats (*Lynx rufus*) and martens (*Martes caurina*). Compared to coyotes, however, SNRF more frequently consumed snow-adapted prey, including white-tailed jackrabbits (*Lepus townsendii*) and American pika (*Ochotona princeps*) (SIMPER $p \leq 0.005$), especially during periods of deep snow. Whitebark pine (*Pinus albicaulis*; presumably seeds) also appeared more regularly in SNRF winter diets compared to coyotes. These findings support the hypothesis that co-adapted subalpine prey facilitate coexistence between specialist and generalist carnivores by increasing the competitive advantage of specialists under snowier conditions. This environment-mediated shift in competitive dynamics implies that the fates of locally adapted predator and prey may be tightly linked, an important consideration for conservation planning in alpine ecosystems.

This is an open access article under the terms of the [Creative Commons Attribution-NonCommercial-NoDerivs](https://creativecommons.org/licenses/by-nc-nd/4.0/) License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.

© 2025 The Author(s). *Molecular Ecology* published by John Wiley & Sons Ltd. This article has been contributed to by U.S. Government employees and their work is in the public domain in the USA.

1 | Introduction

Climate change has precipitated range expansions of many generalist species (Davey et al. 2012; Rödder et al. 2021). Such expanding generalist species can threaten more range- or habitat-restricted specialist species via competition (Urban et al. 2012). In contrast to temperate zones, where species composition is often more dynamic due to weaker interspecific interactions, extreme climate zones tend to support more specialist species, for which fates may be tightly linked, especially among predators and their prey (Elton and Nicholson 1942; Gilman et al. 2010). In high-elevation zones, specialist communities may be especially at risk, as they face both shrinking bioclimatic envelopes and increased competition from generalist species expanding upslope, whose competitive dominance is favoured by warming temperatures (Rödder et al. 2021). Coexistence of specialist predators with generalist competitors in such communities may depend on the presence of prey also adapted to high elevations that are comparably less vulnerable to generalist predators. In turn, persistence of such specialised prey depends on climatic conditions that favour their adaptive defences against generalist predators. Understanding such linked relationships is essential to conserving increasingly threatened montane specialists.

Some prey species in high-elevation montane ecosystems have evolved low foot loading to facilitate locomotion on snow to evade predators. Reduced snow depth and seasonal duration can therefore render such snow-adapted prey species increasingly vulnerable to generalist predators, threatening their persistence (Niittynen et al. 2018; Peers et al. 2020). Loss of keystone prey (i.e., those with a disproportionate effect on the carnivore community), in turn, could disrupt long-term niche relationships among carnivores, threatening the persistence of more ecologically specialised mesocarnivores. Current projections indicate that snowpack levels in the Sierra Nevada and Cascade mountain ranges of the Pacific Crest in the western United States could decline to less than half of their historical (mid-20th century) averages by the late twenty-first century due to climate change (Knowles et al. 2006; Shulgina et al. 2023). The continued decline of snowpack in high-elevation ranges has likely already contributed to the losses of species specifically adapted to high-elevation habitats and threatens remaining high-elevation specialists with extirpation (Parmesan 2006; Urban 2018).

In the Cascade and Sierra Nevada mountain ranges (i.e., Pacific Crest) of the southwestern USA, multiple snow-specialised species are at risk or in decline, including the endangered Sierra Nevada red fox (SNRF; *Vulpes vulpes necator*), one of three recognised subspecies of an evolutionarily distinct lineage of high-elevation red foxes, and several of its potential prey and vegetative foods, such as white-tailed jackrabbit (*Lepus townsendii*), snowshoe hare (*Lepus americanus*), American pika (*Ochotona princeps*) and whitebark pine (*Pinus albicaulis*).

Both of the two extant California SNRF populations, one in the Sierra Nevada range and the other one near Lassen Peak, have been listed as Threatened under the California Endangered Species Act (CESA) since 1980. In addition, the Sierra Nevada distinct population segment (DPS) of the SNRF was listed in 2021 as Endangered under the U.S. Endangered Species Act due to the existence of too few populations, low abundance and low

genetic diversity (U.S. Fish and Wildlife 2021; Quinn et al. 2019). The Southern Cascades DPS, which includes the Lassen population, also was petitioned for federal listing in February of 2024 (Center for Biological Diversity 2024).

The SNRF occurs sympatrically with multiple other mesocarnivores that likely compete for common resources to varying degrees, including coyotes (*Canis latrans*), bobcats (*Lynx rufus*) and Pacific martens (*Martes caurina*). According to niche theory, when multiple species share similar resource utilisation, the potential for competition increases (MacArthur and Levins 1967; Masters and Maher 2022). Long-term coexistence of species with similar niches, therefore, suggests the possibility that past competition has resulted in sufficient partitioning of fundamental niches (Levin and Paine 1974; Levins 1979). To mitigate interspecific competition, sympatric predators can employ various strategies to reduce niche overlap across spatial, temporal and trophic dimensions (Chase and Leibold 2003; Ferreiro-Arias et al. 2021). Among these strategies facilitating coexistence, differentiation in diet (i.e., reducing niche overlap) may play a crucial role for mesocarnivores (Palomares and Caro 1999; Neale and Sacks 2001a, 2001b; Smith et al. 2018).

Within the SNRF's carnivore guild, the coyote, a larger-bodied canid, is expected to have the most similar ecological niche and, therefore, to present the greatest competitive challenge to SNRFs (Palomares and Caro 1999; Azevedo et al. 2006; Webster et al. 2021). Throughout much of lowland North America, red foxes and coyotes occur in sympatry, with the larger coyote often profoundly affecting red fox abundance in space and time (Levi and Wilmers 2012; Newsome and Ripple 2015). The dynamic coexistence of low-elevation red foxes with coyotes is likely facilitated by the sheer size, habitat variability and continuity of the red fox range. However, these qualities do not apply to high-elevation red fox populations, which exist in isolated sky islands that effectively trap them in place, while coyotes are continuously sourced from lower elevations. Isolation is especially pronounced in red fox populations along the Pacific Crest (Quinn et al. 2022, 2024). Therefore, long-term coexistence of SNRF with coyotes must be achieved through some other mechanism, such as niche partitioning.

Indeed, although red foxes in general are known for their broad ecological niche (Voigt 1987), montane red foxes appear to have several unique adaptations to high-elevation montane ecosystems, some of which may facilitate persistence in the face of competition from coyotes (Buskirk and Zielinski 2003; Van Etten et al. 2007; Perrine et al. 2010). Sierra Nevada red foxes breed later in the year than red foxes at lower elevations adjacent to these mountain ranges. This trait is thought to be important in synchronising parturition with green-up and avoidance of the seasonal period of coldest temperatures and greatest food scarcity (Sierra Nevada Red Fox Conservation Advisory Team [SCAT] 2022). Relative to lowland red foxes at lower elevations, additional specialisations include smaller toe pads more densely covered in fur, resulting in a larger overall surface area, lower body mass and a dense fur coat (Grinnell et al. 1937; Fuhrmann 1998; Perrine et al. 2010; SCAT 2022). The heavily furred toe pads and low body mass of montane red foxes reduce their foot loading on snow, similarly to adaptations of the Canada lynx (*Lynx canadensis*), which aid in the capture

of similarly snow-adapted leporids, such as the snowshoe hare (Fuhrmann 1998, 2002; Perrine 2005). Foot loading in montane red foxes is similar to that of the snowshoe hare and Canada lynx (Fuhrmann 1998; Peers et al. 2020), and is up to 9× lower than foot loading of the coyote, likely shifting the competitive balance in favour of SNRF over coyote in deeper snow conditions (Murray and Boutin 1991).

Where they have been studied together, red foxes and coyotes typically exhibit high dietary overlap (Masters and Maher 2022), albeit with coyotes more frequently consuming larger prey, such as mule deer (*Odocoileus hemionus*), and red foxes more frequently consuming smaller prey, such as deer mice (*Peromyscus sonoriensis*) (Major and Sherburne 1987; Peterson et al. 2021; Castañeda et al. 2022; Jensen et al. 2023). Based on a small number of limited diet studies of montane red fox diets, the most common year-round prey appears to be pocket gophers (*Thomomys* spp), with snowshoe hare figuring more prominently in winter diets, along with whitebark pine during mast years (Perrine et al. 2010; Cross and Crabtree 2021; SCAT 2022). We predicted that lagomorphs would play a central role in SNRF diets, particularly in winter and early spring. In addition to being better adapted than coyotes to catch these medium-sized prey on snow, we presumed them to compose an energetically optimal prey in winter and early spring, when male foxes must provision pregnant mates, and when most other medium-sized prey (e.g., ground squirrels) would be hibernating and unavailable. In the Lassen population, snowshoe hares represent the primary lagomorph available in subalpine habitat, whereas in the high-elevation Sierra Nevada, white-tailed jackrabbits make up the more abundant species and are thought to be a historically important winter prey for SNRF (Grinnell et al. 1937). Another high-elevation lagomorph, the American pika, occurs in the high Sierra and Cascade peaks, as do whitebark pine trees, both of which represent potentially important subalpine foods on which we expect the SNRF to specialise (U.S. Fish and Wildlife 2018); whitebark pine nuts could present important sources of calories in winter in the form of seed caches buried the previous fall by sciurids (Cross and Crabtree 2021) or Clark's nutcrackers (*Nucifraga columbiana*; McLaren et al. 2023).

As part of a broader effort to inform recovery plans for the SNRF, our overarching aim in this study was to assess the potential role of subalpine-adapted prey or vegetative food species in sustaining SNRF populations, particularly in the context of presumed competition with other specialised and generalist predators of the subalpine community. Our primary objective was to investigate dietary niche relationships between SNRF and coyotes in the Sierra Nevada site, while using smaller samples of SNRF from the Lassen population and from bobcats and martens from the Sierra site to augment these findings. We tested the following predictions: (1) SNRF consume significant quantities of lagomorphs, (2) dietary niche overlap of SNRF is higher with coyotes than with bobcats or martens, (3) relative to coyotes, SNRF consume more lagomorphs and small rodents, but fewer marmots (*Marmota flaviventris*) and deer, (4) lagomorphs make up a greater proportion of the SNRF winter diet than in other seasons, and (5) SNRF consistently consume significant numbers of lagomorphs, whereas coyote consumption of lagomorphs declines in high-snow years, when their higher foot-loading poses the greatest impediment to capture of lagomorphs. We also

investigated and compared consumption of whitebark pine nuts by carnivores to assess the importance of this alpine-restricted dietary item, especially as a winter food source in SNRF.

2 | Materials and Methods

2.1 | Study Area

The Lassen survey area was in the southernmost portion of the Cascade Range in California at elevations ranging 1722–3187 m. Our sampling area included Lassen Volcanic National Park and Lassen National Forest, including the Caribou Wilderness (Figure 1). The vertebrate community included coyotes, martens, bobcats, snowshoe hares, American pikas, pocket gophers, California ground squirrels (*Otospermophilus beecheyi*), Belding's ground squirrels (*Urocitellus beldingi*), golden-mantled ground squirrels (*Callospermophilus lateralis*), chipmunks (*Tamias* spp.), yellow-bellied marmots, voles (*Arvicolinae* spp), deer mice, mule deer and many species of birds. The most abundant trees included whitebark pine and mountain hemlock (*Tsuga mertensiana*) in subalpine habitats, and lodgepole pine (*Pinus contorta*), Jeffrey pine (*P. jeffreyi*), western white pine (*P. monticola*) and red fir (*Abies magnifica*) in upper montane forests. Annual precipitation averages approximately 105 cm in the Lassen survey area, most of which falls during winter as snow (Parker 1991).

The Sierra Nevada survey area was located along a 50-km length of high-elevation habitat in the central Sierra Nevada of California, with sampling efforts focused on subalpine and alpine regions that were 2570–3500 m above sea level (Figure 1). The survey area was entirely within federal public land and primarily in protected wilderness areas (Quinn et al. 2019). The habitat was similar to that of the Lassen study area, except that it was at higher elevation, on average, and whitebark pine was therefore more widespread relative to lodgepole pine and other pines, such as western white pine and Jeffrey pine. Additionally, the main leporid species at the Sierra Nevada site was the white-tailed jackrabbit, rather than snowshoe hare. Average annual precipitation at Sonora Pass was approximately 130 cm, most of which falls in the winter as snow (Schoenherr 1992).

2.2 | Samples

We used 925 DNA extracts from scats obtained during 2011–2017 for previous studies or monitoring efforts (metadata available in Dryad). These scats were previously species-typed using a portion of the mitochondrial cytochrome *b* gene to identify scats as coming from SNRFs, coyotes, martens and bobcats for which methods were previously detailed (Quinn et al. 2019; SCAT 2022). Briefly, searches were conducted on foot along trails, ridges and at passes. In the Lassen area, scats thought likely to be from SNRF were collected. In the Sierra Nevada, all carnivore scats encountered except those of black bear (*Ursus americanus*), which were readily distinguished from those of other carnivores, were collected. Coordinates of the scat locations were recorded before collecting in a paper bag; a small portion of each scat (~1 cm³) was stored in 8–12 mL of 95%–99% ethanol usually on the same day of collection.

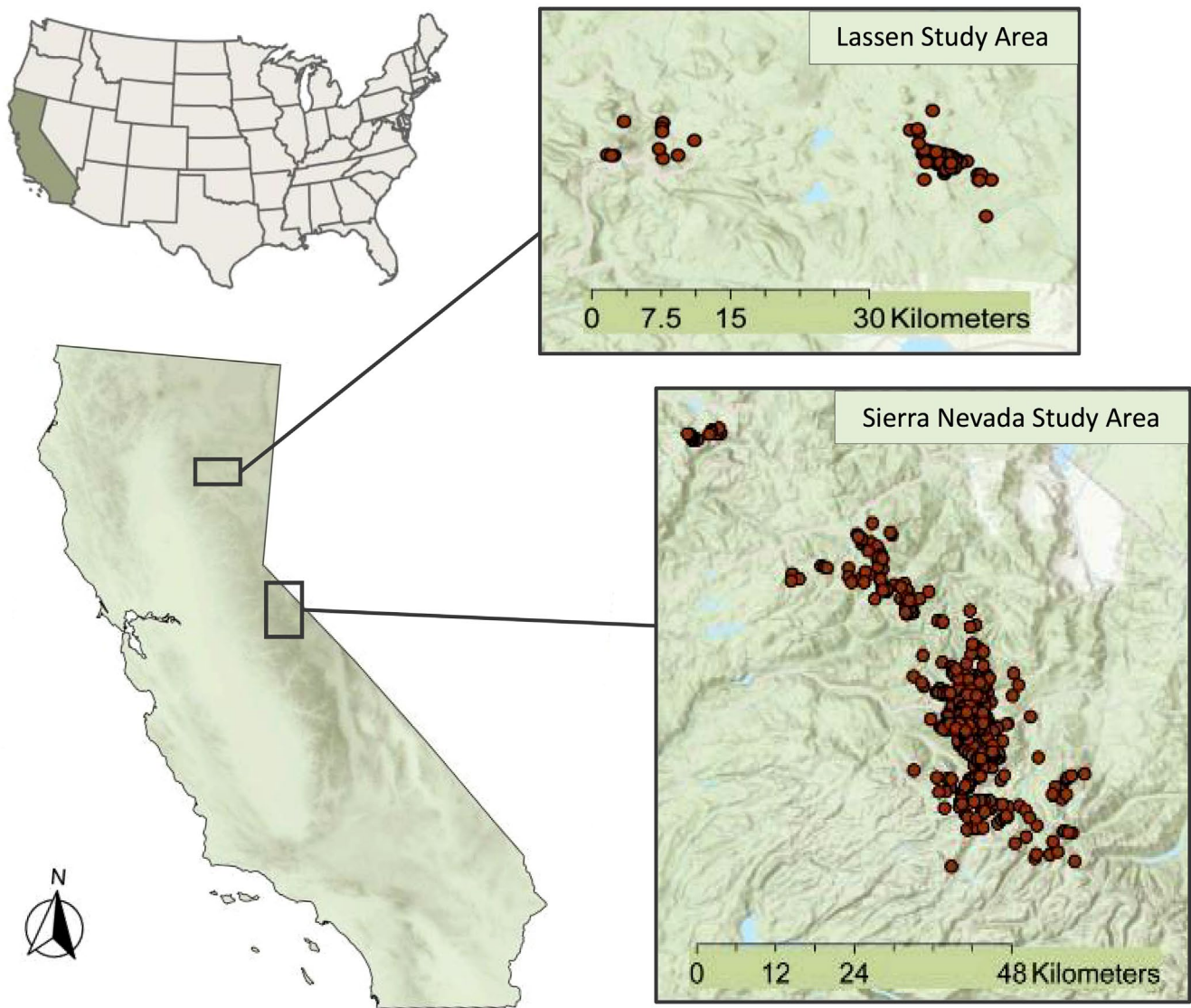


FIGURE 1 | Lassen and Sierra Nevada mesocarnivore diet study areas, along with locations of 925 mesocarnivore scats (brown circles) collected in California, USA. The Lassen area included 133 Sierra Nevada red fox (SNRF) scats collected from 2011 to 2015 in Lassen National Park and Lassen National Forest. The Sierra Nevada area included 317 SNRF, 329 coyote, 88 marten and 58 bobcat scats collected from 2011 to 2017 north of Yosemite National Park in the Stanislaus and Humboldt-Toiyabe National Forests.

Survey effort varied among years, for example, ranging in the Sierra from 31 days to survey 553 km and recovery of 396 scats (including 48 successfully sequenced SNRF samples; 2017) to 118 days to survey 1445 km and recovery of 460 scats (including 89 successfully sequenced SNRF samples; 2013) (Quinn et al. 2019). The DNA was extracted using the QIAamp Stool Kit (Qiagen Inc., Valencia CA) according to manufacturer's instructions except that elution was limited to 50 μ L. Based on 84% of samples in the Sierra site for which we also had associated GPS tracks, collection of a single DNA-verified SNRF scat required on average 1.6 days and 12.9 km of survey effort. The bulk of survey effort involved intensive scat searches conducted in the snow-free periods, when backcountry travel was easiest, which resulted in fewer winter scats compared to the spring, summer and fall seasons. Seasons were defined as winter (Dec–Mar), spring (Apr–Jun), summer (Jul–Aug) and fall (Sep–Nov). Although the age of scats was unknown, we presumed most that were successfully species typed (which

are the only ones used in this study) were deposited recently (e.g., < 1 month). Even if scats are not scattered by wind or other animals, field trials indicate that amplification success of DNA from scats declines steadily with the age of scat, and precipitously when more than a month old (Piggott 2004). Nevertheless, we expect that some portion of the scats was likely deposited in the season prior to that in which they were collected, which would tend to introduce noise into observed seasonal patterns.

2.3 | Metabarcoding Procedures

Preparation of the sequencing libraries entailed two PCR steps for each DNA extract: (1) amplification of vertebrate and plant markers (amplicon PCR) and (2) annealing of Illumina-compatible adapter sequences and unique 8-bp (forward) and 6-bp (reverse) indexes (index PCR). For the amplicon PCR, we

used a multiplex reaction to simultaneously amplify prey species and plant-based food items with both vertebrate and plant markers (Table S1). For the vertebrate marker, we amplified a 101-bp region of the mitochondrial 12S gene, using a modification (Caspi et al. 2025) of previously published primers (Riaz et al. 2011). Specifically, we added degenerate bases to the first two 5' bases of the forward primer and to the last 3' base of the reverse primer to broaden the taxonomic range of the primer set. We deemed these modifications necessary based on comparisons of the published primers to sequences accessioned in NCBI, which indicated a few consistent mismatched bases. Most importantly, the 3' base of the reverse Riaz et al. (2011) primer mismatched that of all species examined within the family, Geomyidae, which included pocket gophers, an abundant prey item in the montane ecosystems. We also included custom oligonucleotide blockers specific to red foxes, coyotes, martens and bobcats (Table S1) to help reduce predator detections and increase prey item detections (Deagle et al. 2009; Shehzad et al. 2012). We designed these blockers similarly to those of a previous carnivore study that used the 12S locus with a 3' 3-carbon spacer (C3 CPG; Vestheim and Jarman 2008; Shehzad et al. 2012). For the plant marker, we amplified a variable-length (< 100 bp) region of the P6 loop of the trnL intron of the chloroplast DNA, specifically using trnL_g and trnL_h (Taberlet et al. 2007). To both primer sets, we added sequence overhangs to the 5' ends to allow a second round of amplification to attach Illumina adapter and index sequences.

For the amplicon PCRs each 9- μ L reaction included 2- μ L DNA sample, 1.7- μ L of RNase-free water, 5.5- μ L of 2 $\times$ Multiplex Mastermix (Qiagen, Valencia, CA, USA), 1.1- μ L of 10 $\times$ Qiagen Q-solution, 0.8- μ M predator blocker, 0.1- μ M trnL-g and 0.1- μ M trnL-h, 0.08- μ M P5-12SV5 and 0.16- μ M P7-12SV5. Samples were then run on the following thermal profile: Denatured at 95°C for 15 min; followed by 40 cycles of 30 s at 94°C and 90 s at 55°C, then 15°C indefinitely.

We cleaned amplicon PCR products using an exonuclease and shrimp alkaline phosphatase (SAP) solution (Exo-SAP) (Olsen et al. 1991). To clean with Exo-SAP solution, 3- μ L of amplicon product from each sample was added to the following mixture: 0.075- μ L of 20- μ M exonuclease I, 2.775- μ L of TE buffer (10 mM Tris-HCl + 1 mM EDTA), and 0.15- μ L of 1 U/ μ L SAP (New England Biolabs, Ipswich, MA, USA). Samples were then run on the following thermal profile: 37°C for 30 min followed by 80°C for 15 min.

For the index PCR, each 25.5- μ L reaction included 12.5- μ L of 2 $\times$ NEBNext Ultra II Q5 Master Mix (New England Biolabs), 1- μ M indexed P5 primer, 5- μ L of RNase-free water, 2.5- μ L of 10- μ M indexed P7 primer and 3- μ L of cleaned PCR product from the first reaction. Samples were then run on the following thermal profile: 98°C for 30 s, 8 cycles of 98°C for 10 s, 8 cycles of 65°C for 75 s, 65°C for 5 min and 4°C indefinitely. We pooled 2- μ L from each sample of index PCR product and purified it using a QiaQuick column (Qiagen) following the manufacturer's protocol and stored the final product in a 1.5-ml Lo-Bind tube at -20°C until all plates could be pooled. Pooled libraries were sent to Novogene Corporation Inc. (Sacramento, CA, USA) or Admera Biopharma (South Plainfield, NJ, USA) for paired-end 150-bp sequencing on an Illumina NovaSeq S4 or NovaSeq X Plus, respectively.

We processed 95 arbitrarily selected samples twice as independent replicates to assess the repeatability of the metabarcoding results (Ficetola et al. 2015). As a measure of background contamination during the metabarcoding process and to identify artefacts for removal, such as reagent contaminants (e.g., human, potato starch), we also included 104 controls, specifically 8 controls in each of 13 96-well plates: two positive controls composed of separate mixtures of equimolar vertebrate or plant DNA from multiple species foreign to the Cascade and Sierra Nevada ranges, a combination of these two mixtures, and 5 no-template controls (NTCs). The vertebrate mixture was composed of 0.2 ng/ μ L DNA from Laysan albatross (*Phoebastria immutabilis*), elephant seal (*Mirounga angustirostris*), blunt-nosed leopard lizard (*Gambelia sila*), wakasagi smelt (*Hypomesus nipponensis*) and western yellow bat (*Lasiurus xanthinus*). The plant mixture was composed of Joshua tree (*Yucca brevifolia*), desert mariposa (*Calochortus kennedyi*), pickleweed (*Salicornia pacificus*), Thurber's sandpaper plant (*Petalonyx thurberi*) and Bigelow's linanthus (*Linanthus bigelovii*).

2.4 | Data Analysis

We used Cutadapt to remove priming and adapter sequences from the sequencing reads (Martin 2011). We then used the DADA2 R package to reduce sequencing errors and filter the sequencing reads by expected insert length (Callahan et al. 2016). We ran DADA2 separately for 12SV5 and trnL primers, using a minimum filtering length of 80 bp and a maximum of 110 bp for 12SV5 reads and a minimum length of 10 bp and a maximum of 79 bp for trnL reads.

For each marker, we sorted amplicon sequence variants (ASVs) from most frequent to least frequent in the data set and discarded all ASVs with frequencies > 99th percentile. Next, we assigned each of the remaining ASVs to a taxon using the Nucleotide database in GenBank with the Basic Local Alignment Search Tool (BLAST) algorithm (Altschul et al. 1990) or, when no acceptable match was found (e.g., snowshoe hare, *Lepus americanus*; California ground squirrel, *Otospermophilus beecheyi*), from our custom sequence library.

To assign vertebrate ASVs (i.e., 12SV5) to the species level, we required that ASVs matched the corresponding sequence at $\geq 99\%$ of bases (percent identity), overlapped at least 98% of the length of the amplicon (query coverage), occurred in the study area, and were the only species meeting those criteria. Alternatively, when the top hits corresponded to species not present in the study area (e.g., *Peromyscus maniculatus bairdii*), we inferred the most closely related taxon that occurred in the study area, usually for which no sequence was available in GenBank or in our collection (e.g., *Peromyscus sonoriensis*). When multiple species occurring in the study area had equally high identity matches (e.g., several *Tamias* spp), we assigned taxa to the genus level. More generally, when multiple taxa at any level were present in the study area with similarly high identity matches (e.g., several birds in the order Passeriformes), we assigned taxa to the lowest-level taxon possible. In a few cases, when no 12SV5 sequence was available on GenBank or in our collection, the next-highest taxonomic level was assigned. Multiple ASVs that were identified as the same taxon

were pooled into a single operational taxonomic unit (OTU), that is, diet item, by summing the sequence reads across all such ASVs. For data analysis, we used only prey reads, that is, excluding carnivore reads, reagent contaminants identified from controls and other artefacts (e.g., nuclear-embedded mitochondrial DNA sequences [numts]).

To investigate the potential role of whitebark pine nut caches in the diets of SNRF, we used the trnL marker, which resulted in a 45bp sequence for *Pinus* spp., and differentiated lodgepole pine by one substitution from whitebark and other high-elevation pines, including western white pine. Two other montane pines, Jeffery pine and sugar pine (*P. lambertiana*), shared the sequence with whitebark pine, but typically occurred below the elevations of the Sierra Nevada site. We also noted occurrence of fruits, such as manzanita (*Arctostaphylos* spp.) that foxes and coyotes were known to regularly consume (Neale and Sacks 2001a, 2001b; B. N. Sacks, unpublished data), but assumed that most diet items from plants with non-fleshy fruits, such as buckwheats (*Eriogonum* spp.), could have derived secondarily from the ingesta of vertebrate prey.

We removed from each sample diet items composing <1% of the total number of sequencing reads for a given marker (12SV5, trnL). We expressed remaining reads in terms of frequency of occurrence (FOO) and relative read abundance (RRA), where FOO represents the percent of samples containing a given diet item and the RRA is calculated as the average across samples of the proportional composition of reads of a given diet item in each sample. We calculated RRA separately for vertebrate and plant reads. Although RRA is more susceptible than FOO to bias by factors such as uneven PCR amplification of different prey items (Lamb et al. 2019), such biases were expected to apply equally to comparison groups; therefore, they did not invalidate qualitative comparisons among mesocarnivore diets (White et al. 2023; Caspi et al. 2025). Moreover, RRA is less susceptible than FOO to over-representation of frequent but low read numbers due, for example, to background contamination. Consequently, the two measures provide complementary ways of characterising diet.

Although we used these direct measures of diet-item consumption (FOO, RRA) in all statistical analyses unless specifically indicated otherwise, it was also important for the interpretation of the importance of various diet items to assess their contributions to the diet in terms of the proportion of biomass consumed. We used the observation that larger prey contribute proportionally more biomass per occurrence than do smaller prey (Neale and Sacks 2001a, 2001b). We employed correction factors (scalar multipliers of FOO) derived from a previous study of coyote and bobcat diets in terms of both frequency of occurrence and an independent measure of biomass consumption (table 2 in Neale and Sacks 2001b) to grossly estimate the proportion of biomass consumed by each predator. We calculated correction factors for each prey item as the ratio of estimated biomass consumption from table 2 in Neale and Sacks (2001b) divided by FOO for each of the same prey items in the same table, most of which had conspecific or closely related counterparts with similar body sizes in our study (Table S2). Because Neale and Sacks provided biomass and FOO estimates separately for coyotes and bobcats (but which were similar), we averaged the bobcat and coyote

biomass/FOO ratios for each prey for application to all carnivores in this study.

To compare prey-item richness among groups with different sample sizes, we created species accumulation curves (SAC) using the 'specaccum' function (Ugland et al. 2003) in the R package vegan (Oksanen et al. 2024). The SAC curves were created with the 'random' method choice, which finds the mean SAC and its standard deviation from random permutations of the data (Gotelli and Colwell 2001). To then estimate the expected prey richness (i.e., under infinite sample size) for each mesocarnivore, we calculated Chao richness extrapolation values and standard errors with the vegan subpackage 'specpool' (Chao 1987). Shannon diversity was calculated for each mesocarnivore using the 'specnumber' and 'diversity' functions in vegan.

We computed a pairwise matrix of Jaccard dissimilarity indexes based on FOO values and a pairwise matrix of Bray Curtis dissimilarity indexes based on RRA values among samples using the 'vegdist' function in vegan. We used these dissimilarity matrices to create both FOO-based and RRA-based nMDS plots for the diet of all our sampled mesocarnivores. We then performed a series of PERMANOVA tests using the 'adonis2' function in vegan to assess statistically significant differences in diets among mesocarnivores, where 'difference' in this case was defined as a difference in the centroid (diet composition), the dispersion (diet breadth), or both (Anderson and Walsh 2013). We compared the prey items of the Sierra Nevada SNRF to the prey items of Lassen SNRF, the Sierra Nevada coyote, Sierra Nevada marten and Sierra Nevada bobcat by running separate PERMANOVAs for each pairing.

We additionally calculated Pianka's niche overlap indices (0: no overlap, 1: full overlap; Pianka 1973) using the 'spaa' subpackage in vegan (Zhang and Ma 2014) and FOO values of each prey item. Niche overlap tables were created for rarefied samples to account for different sample sizes among the mesocarnivores. We used a null model analysis to test whether the observed niche overlap values differed from what would be expected by chance if diets were identical, using the 'EcoSimR' package to compare results with 1000 permutations of a null model that retained the dietary niche width of each species while randomising prey item values (Gotelli et al. 2015). We used the 'simper' function in vegan to perform a similarity percentage (SIMPER) analysis (Clarke 1993) between Sierra Nevada SNRF and Sierra Nevada coyotes on our Jaccard dissimilarity matrix data to determine which prey items contributed the most to dietary niche differentiation between these two mesocarnivores.

To further investigate the role of SNRF adaptation to capture of white-tailed jackrabbits in snow, we examined the relationship between annual snowpack and both SNRF and coyote consumption of white-tailed jackrabbits. To test the prediction that the difference in consumption of this prey by the two predators would increase with increasing snowpack (SNRF consumption positively and coyote consumption negatively correlated), we compared the snow water content measured as of April 1st of each year in the Central Sierra Nevada weather stations downloaded from the California Department of Water Resources (<https://cdec.water.ca.gov>) to the frequency of

TABLE 1 | Sample sizes and dietary diversity metrics for 103 Sierra Nevada red fox (SNRF) scats from Lassen from 2011 to 2015 and 296 SNRF scats, 279 coyote scats, 61 marten scats and 50 bobcat scats from the Sierra Nevada from 2011 to 2017.

Predator	No. of scats (n)	No. Vertebrate Prey items	Chao estimated richness	Chao SE	Shannon diversity
Lassen SNRF	103	53	93.74	23.55	3.19
Sierra SNRF	296	50	74.00	15.99	2.54
Sierra coyote	279	55	69.99	9.00	2.76
Sierra marten	61	30	44.88	12.28	3.01
Sierra bobcat	50	36	88.92	40.11	3.12

occurrence of white-tailed jackrabbits in scats of the two predators. Although seasonal patterns of snowpack vary somewhat across years, April 1 represents a relatively consistent measure of peak annual snowpack. As an additional indicator of specialisation versus opportunistic consumption, we compared the multiannual consistency between the carnivores in consumption of both white-tailed jackrabbits and pine. Although we had no data on the abundance of lagomorphs or mast across the years of our study, both are known generally to vary multiannually (e.g., Hodges 1999; Bartel et al. 2008; Crone et al. 2011). Our expectation, therefore, was that a more specialised diet would be reflected in consistent consumption across years (i.e., regardless of fluctuations in availability), whereas opportunistic consumption would be reflected by more variable multiannual consumption, consistent with consumption tracking availability.

3 | Results

3.1 | Vertebrate Diet Items

We attempted to sequence 925 predator scats, along with 95 replicates (approximately 10%), 26 positive controls and 78 negative control samples (i.e., including 65 NTCs and 13 positive controls for each marker). Positive controls ($n = 26$) contained an average of 39,546 (SD = 34,546) vertebrate sequence reads each, nearly all of which corresponded to the expected positive-control species. Positive controls contained only 5.6 (range: 0–40) contaminant prey reads (0.01%) on average. From negative controls ($n = 78$), we identified 176 contaminant prey reads per sample on average, with 95% of them containing < 300 prey reads. As a minimum threshold for total prey read count for including a sample in analyses, we used double the number of reads corresponding to the 95th percentile for contaminant reads in negative controls (i.e., 300), which translated to excluding scat samples with < 600 prey reads.

We identified 90 vertebrate dietary items composed of 151 ASVs (Tables S3 and S4). Most ASVs were assigned at the species level, although some ASVs were consistent with multiple species with a 100% match (e.g., *Tamias* spp., *Microtus* spp). The resulting dataset contained 866 mesocarnivore samples with > 600 reads, including 789 distinct samples and 77 replicates (Table 1). Based on the 77 pairs of replicate samples, the RRA correlation was nearly perfect (Pearson $r = 0.999$), indicating that the dietary composition of the samples was highly repeatable. The average

number of vertebrate reads in samples was 18,399 (SD = 15,497). The number of vertebrate prey items per sample averaged 2 and ranged from 1 to 6.

3.2 | SNRF Diet in Lassen and Sierra Nevada Populations

Our sample size was higher for the Sierra SNRF than for the Lassen SNRF (Table 1). Nevertheless, the Lassen SNRF population showed higher breadth and variation in scat prey items compared to Sierra Nevada SNRF in terms of SACs, total numbers of prey items recorded (despite smaller sample size and increasing SAC), and Chao richness and Shannon diversity estimators (Figure S1; Table 1). We detected significant differences in prey composition of diets between the Lassen and Sierra Nevada populations for both FOO and RRA values (Table 2; Figure S2).

We observed both commonalities and differences in the prey in the diets of SNRF from the two populations. For example, pocket gophers were the top prey item in both populations in terms of estimated biomass, FOO and RRA (Figure 2; Tables S3 and S4). Lagomorphs were also important in both populations, although the primary lagomorph species in the diets differed as expected given the ranges of these prey species. In terms of biomass consumption, snowshoe hare was the third highest ranked prey item in the Lassen population (after pocket gophers and golden-mantled ground squirrels), whereas white-tailed jackrabbit and American pika were the second and third highest ranked prey items, respectively, in the Sierra Nevada population (after pocket gophers). Patterns were similar based on RRA, although snowshoe hare was the second highest ranked prey item in Lassen (Table S4). In contrast to the Sierra population, the Lassen population also frequently consumed fish, principally rainbow trout.

3.3 | Dietary Breadth and Overlap of SNRF With Sympatric Mesocarnivores

Compared to SNRF and coyotes in the Sierra Nevada site, our sample sizes were relatively small for bobcats and marten (Table 1). Nevertheless, bobcat diets showed the highest prey richness and breadth based on SACs, Chao estimators and the Shannon diversity index (Figure S1; Table 1). Based on both PERMANOVA and Pianka's niche overlap index, we detected significant differences in diet composition between SNRF and each of the other mesocarnivores, including coyotes, for both

TABLE 2 | Results of permutational multivariate analysis of variance (PERMANOVA) performed on frequency of occurrence (FOO) and relative read abundance (RRA) in Lassen (Las) and Sierra populations of Sierra Nevada red fox (SNRF) and Sierra SNRF with sympatric coyotes, martens and bobcats.^a

Data	Pairing	PERMANOVA			
		No. of scats (n)	R ²	F ₂	p
FOO	Sierra SNRF/ Las SNRF	296/103	0.036	14.67	≤0.001
	Sierra SNRF/ coyote	296/279	0.029	16.83	≤0.001
	Sierra SNRF/ marten	296/61	0.053	19.70	≤0.001
	Sierra SNRF/ bobcat	296/50	0.056	20.29	≤0.001
RRA	Sierra SNRF/ Las SNRF	296/103	0.033	13.30	≤0.001
	Sierra SNRF/ coyote	296/279	0.014	8.30	≤0.001
	Sierra SNRF/ marten	296/61	0.067	24.88	≤0.001
	Sierra SNRF/ bobcat	296/50	0.056	20.28	≤0.001

^aAnalyses used 103 SNRF scats from Lassen from 2011 to 2015 and 296 SNRF scats, 279 coyote scats, 61 marten scats and 50 bobcat scats from the Sierra Nevada from 2011 to 2017.

FOO and RRA (Tables 2 and 3; Figure S3). Niche overlap was highest for SNRF and coyote (0.89). Among all the mesocarnivore pairings, coyote and marten diets overlapped least (0.46).

Certain prey items occurred more frequently in scats of some mesocarnivores than others. Of all the Sierra Nevada mesocarnivores, SNRF had the highest proportion of white-tailed jackrabbit (FOO=0.29) and deer mouse (FOO=0.34), whereas coyotes had the highest frequency of mule deer in scats (FOO=0.15) (Table S3). Converting these numbers to estimates of biomass consumption similarly indicated high use of white-tailed jackrabbit by SNRF and deer by coyote, but indicated that smaller-bodied deer mice represented only an estimated 7% of the biomass consumed by SNRF despite their occurrence in 34% of scats (Figure 2). Pocket gophers occurred less frequently in marten (FOO=0.23) and bobcat scats (FOO=0.28), whereas squirrels (Sciuridae), especially golden-mantled ground squirrels and Douglas squirrels (*Tamiasciurus douglasii*), occurred more frequently in marten

(FOO=0.20, 0.13, respectively) and bobcat scats (FOO=0.34, 0.20) compared to SNRF (FOO=0.04, 0.00) and coyote scats (FOO=0.04, 0.01) (Table S3). In general, sciurids (squirrels) composed 46% and 52% of the estimated biomass consumed by martens and bobcats, respectively, but only 7% of estimated biomass consumed by SNRF; one sciurid, in particular, the yellow-bellied marmot, composed 11% of the estimated biomass consumed by coyote (Figure 2).

3.4 | Diet Dissimilarity Between SNRF and Coyotes in the Sierra Nevada

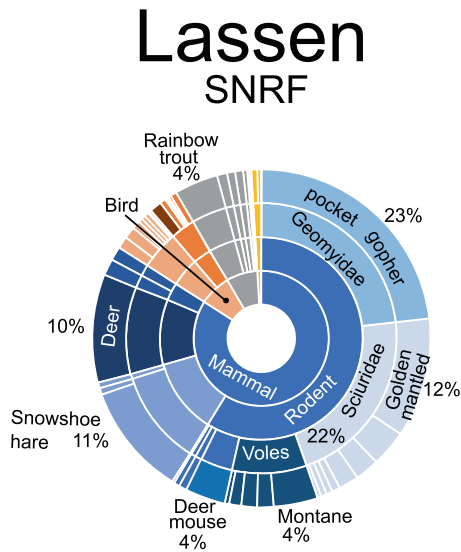
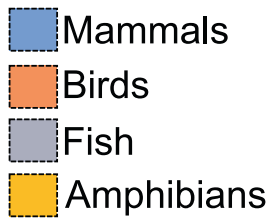
Between the two canids, diets differed in terms of a small number of prey species, consistent with niche differentiation due both to body size and adaptive specialisations of both prey and predators. Specifically, SIMPER analysis showed that white-tailed jackrabbit ($p=0.005$), deer mouse ($p=0.001$) and American pika ($p=0.004$) were consumed significantly more frequently by SNRF than by coyotes (Table S5). In contrast, coyotes more frequently consumed mule deer ($p=0.001$), montane vole ($p=0.004$), yellow-bellied marmot ($p=0.0001$) and dark-eyed junco ($p=0.005$). Most prey species contributed similarly to the biomass consumed by these canids, with the exception of the smallest and largest prey (deer mice, deer and marmots) and lagomorphs, white-tailed jackrabbit and pika, that were specialised to the subalpine environment (Figure 3).

Seasonal differences provided additional insights into niche partitioning between SNRF and coyotes (Table S6; Figure S4). Pianka's overlap values were lowest (0.69) during winter when food resources were likely to be most limiting, and higher during spring (0.91), summer (0.89) and fall (0.86). All comparisons differed significantly from 1.0 ($p<0.001$).

Seasonal dietary breadth also varied for Sierra Nevada SNRF and coyotes. The SACs and Chao species richness estimates taken together indicated a higher species richness for summer and fall seasons than winter and spring for both species (Figure S5; Table S7). Occurrence of prey items in SNRF scats was more even in winter than in other seasons, resulting in similar Shannon diversity estimates of the winter diet to other seasons (Table S7). Hibernating mammals, such as yellow-bellied marmots and other ground squirrels, were absent in all SNRF and coyote winter scats, contributing to the lower number of species consumed (Table S6). Conversely, white-tailed jackrabbits were consumed more frequently in winter and spring than summer or fall, with the difference most pronounced in coyotes (Figure 4A). This pattern was consistent with the absence in fall and summer of two prey (marmots, deer) that composed a large portion of biomass consumed by coyotes (but not SNRF) overall, suggesting coyotes preyed secondarily on white-tailed jackrabbits.

3.5 | Whitebark Pine and Other Vegetation

For plant-based diet items, positive controls ($n=26$) contained an average of 3386 (SD=10,208) positive control sequence reads each, along with 35 (range: 0–385) contaminant reads (1%) on average. From negative controls ($n=78$), however, we identified



Sierra

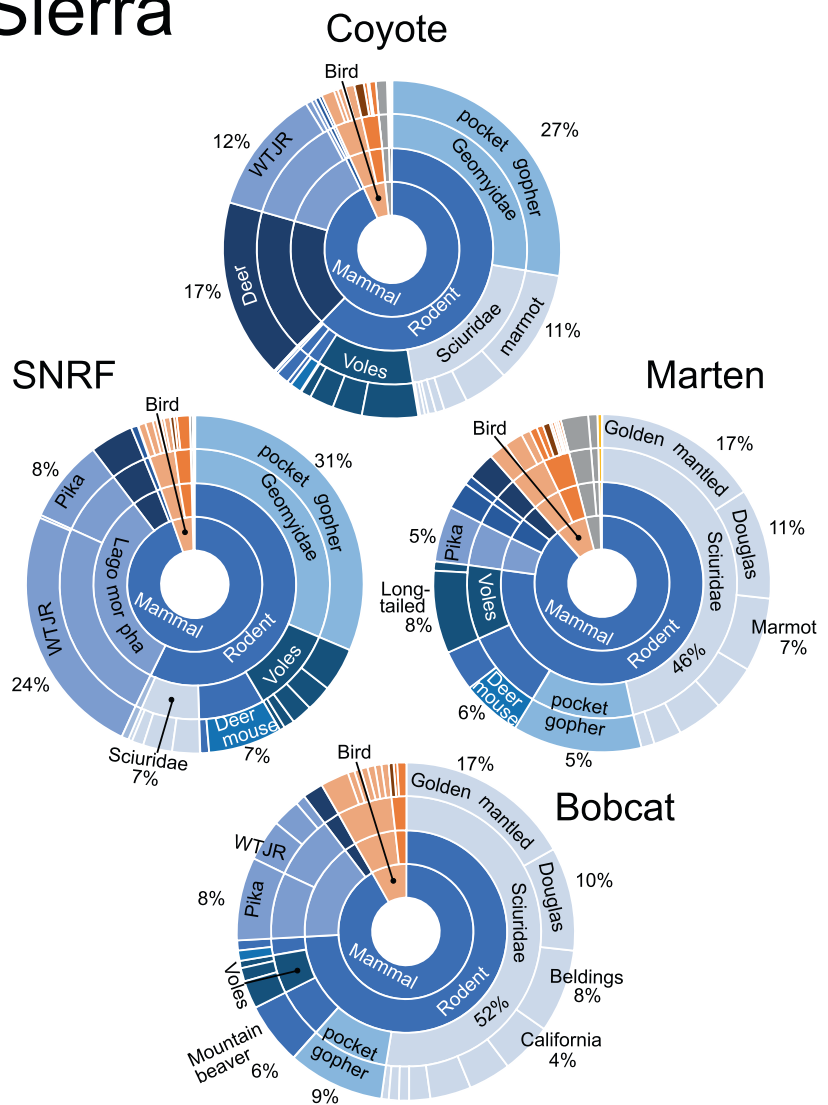


FIGURE 2 | Dietary composition in terms of estimated proportional biomass contributions of prey calculated as frequency of occurrence multiplied by a correction factor (Table S2) for mesopredators including 103 Sierra Nevada red fox (SNRF) scats from Lassen from 2011 to 2015 and 296 SNRF, 279 coyote, 61 marten and 50 bobcat scats from the Sierra Nevada from 2011–2017. Rings from inner to outer represents taxonomic class, order, family (or subfamily) and species (or lowest known operational taxonomic unit). For convenience, common names and abbreviations were used at the species level and for some higher-order taxa, for example, to indicate Arvicolinae (voles), Mammalia (mammal), Aves (bird), Rodentia (rodent) or white-tailed jackrabbits (WTJR). Complete taxonomic information for prey items is provided in Table S2.

4996 contaminant plant reads per sample on average, with a median of 1155 contaminant reads. The ASV sources of contamination showed no consistent pattern, indicating that when samples contained no template plant DNA (i.e., to dominate annealing of primers) they became highly sensitive to background contamination. Therefore, we focused specifically on pines, which occurred in low frequency in controls ($n = 5$ of 104 controls) and were of greatest interest, but also noted the occurrence of manzanita, which did not show up in any controls.

We documented a significant occurrence of pine in the diets of both SNRF populations, as well as by coyotes and especially martens (Tables S8 and S9). Specifically, the ASV associated with whitebark and other pines (but not lodgepole) was found in

scats of the Lassen SNRF population (FOO = 0.26, RRA = 0.11), and in the Sierra Nevada in SNRFs (FOO = 0.13, RRA = 0.04), coyotes (FOO = 0.11, RRA = 0.07) and martens (FOO = 0.43, RRA = 0.34), but as expected given its hyper-carnivorous status, rarely in bobcats (FOO = 0.02, RRA = 0.03). The Lassen SNRF scats also frequently contained lodgepole pine (FOO = 0.20, RRA = 0.03), in contrast to all carnivores in the Sierra Nevada, including SNRF (FOO = 0.01, RRA = 0.01). This difference was consistent with the higher elevation of the Sierra Nevada than Lassen study areas (and populations).

Another difference likely reflecting this elevational difference was in the consumption of manzanita (*Arctostaphylos* spp.), which was found in 44% of SNRF scats (RRA = 0.19) from Lassen

TABLE 3 | Pianka's (1973) niche overlap index (below diagonal) and *p*-values (above diagonal) estimated for each pair of mesocarnivores from frequency of occurrence values using rarefaction analysis.^a

	Sierra SNRF	Sierra coyote	Sierra bobcat	Sierra Marten
Sierra SNRF	—	0.001	0.002	0.004
Sierra Coyote	0.89	—	0.006	0.013
Sierra Bobcat	0.59	0.56	—	0.001
Sierra Marten	0.56	0.46	0.74	—

^aAnalyses used 296 Sierra Nevada red fox (SNRF) scats, 279 coyote scats, 61 marten scats and 50 bobcat scats from the Sierra Nevada from 2011 to 2017. The *p*-values refer to the null hypothesis that niche overlap was 100%.

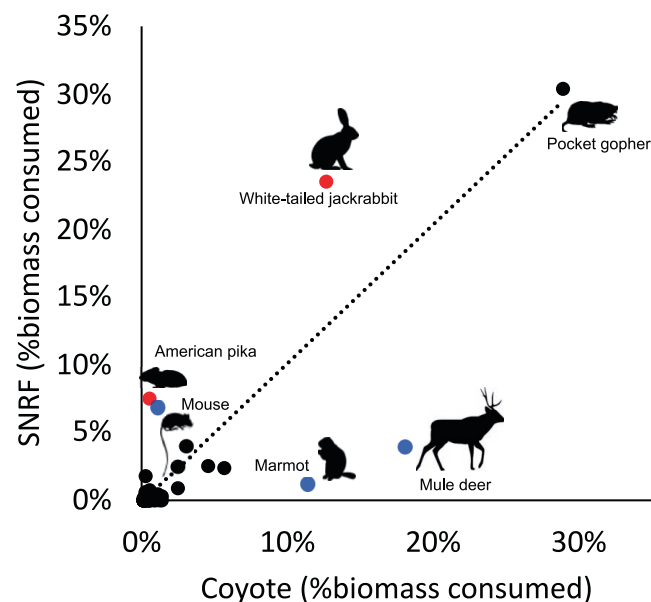


FIGURE 3 | Estimated proportional biomass contributions of prey species to the diets of Sierra Nevada red fox (SNRF) and coyote prey based on frequency of occurrence in scats multiplied by a prey body-size correction factor (see Table S2), illustrating that most prey species constitute similar proportions of the diets of both canids, with exceptions noted in blue and red symbols. Red symbols indicate prey species with specialised adaptations to the subalpine zone of the Sierra Nevada on which SNRF may be especially well-adapted to capture; blue symbols represent prey species likely to differ between carnivore diets due to body sizes. Pocket gophers contributed the greatest biomass to the diets of both carnivores. Data were based on 296 SNRF and 279 coyote scats from the Sierra Nevada from 2011 to 2017.

and <1% of SNRF scats (RRA = 0.001) in the Sierra Nevada. The only plant-based items with high frequency in bobcat scats were buckwheat (*Eriogonum* spp.) (FOO = 0.18, RAA = 0.24) and wild or bitter cherry (*Prunus* spp.) (FOO = 0.18, RRA = 0.17), which were also frequently found in the scats of the other carnivores, suggesting they likely sourced indirectly from the ingesta of herbivorous prey.

Pine consumption by SNRF in the Sierra Nevada site varied seasonally, consistent with winter reliance on caches of white-bark pine nuts (Figure 4B). We had too few winter SNRF scats from Lassen to investigate seasonal trends in that population. In the Sierra Nevada site, pine FOO values were highest in SNRF winter scats (winter: 0.38) compared to the other three seasons (spring: 0.11 summer: 0.13, fall: 0.09; $\chi^2 = 15.94$, $df = 3$, $p = 0.001$). Pine FOO values remained relatively even across seasons for coyotes (winter: 0.13, spring: 0.10, summer: 0.11, fall: 0.16; $\chi^2 = 1.12$, $df = 3$, $p = 0.77$). The RRA patterns were similar, with pine in SNRF scats highest in winter (winter: 0.19) compared to the other three seasons (spring: 0.04, summer: 0.03, fall: 0.02; Kruskal–Wallis $\chi^2 = 17.65$, $p = 0.0005$). Pine RRA values remained relatively even across seasons for coyotes (winter: 0.05 spring: 0.04 summer: 0.08 fall: 0.08; Kruskal–Wallis $\chi^2 = 0.93$, $p = 0.82$).

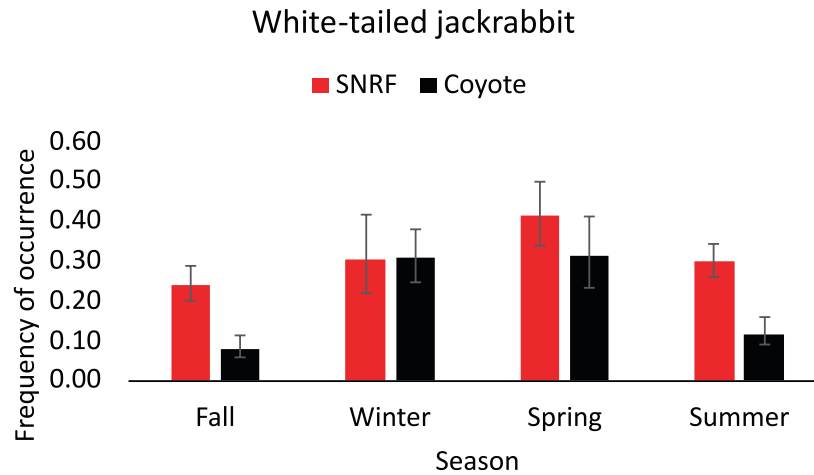
3.6 | Multiannual Patterns

We examined patterns of consumption of white-tailed jackrabbits and pine in the Sierra Nevada site across 6 years, where we had sufficient sample sizes for SNRF and coyotes. The FOO of white-tailed jackrabbit was more stable across years in SNRF than in coyote scats, which exhibited a downward trend from 2012 through 2017 (Figure 5A). In general, the frequency of occurrence of white-tailed jackrabbit in SNRF scats was positively correlated with snow pack, whereas in coyote scats it was negatively correlated with snow pack, resulting in little to no difference in consumption by SNRF and coyotes of white-tailed jackrabbits in low-snow years and large differences in high-snow years (Figure 5A,B). In contrast, pine FOO varied somewhat erratically across years with no clear relationship to snowpack and did so similarly for coyotes and SNRF (Figure 5C), consistent with opportunistic consumption by both carnivores.

4 | Discussion

In this study, we sought to understand which prey were most crucial to the persistence of Sierra Nevada red foxes in their subalpine environments. Although we did not and could not test for competition per se, our inferences about key prey were guided by the presumption that prey were frequently limiting in this environment, particularly during winters and springs (e.g., Schoenherr 1992), and therefore that niche partitioning was an important determinant of coexistence within the mesocarnivore community, especially for SNRF relative to coyotes (e.g., Newsome and Ripple 2015). Our sampling of sympatric mesocarnivores in the Sierra site, therefore, provided greater insights regarding the importance of prey to the Sierra SNRF population, whereas comparisons between diets of the two SNRF populations provided additional evidence that lagomorphs were likely to be broadly important to SNRF across a range of elevational zones. Based on previous observations reviewed in the Introduction, we predicted that SNRF were specialised to capture lagomorphs, particularly white-tailed jackrabbits and snowshoe hares, in snowy environments and that this adaptation facilitated coexistence with coyotes, which were less able to efficiently utilise these hypothetically key prey resources,

A.



B.

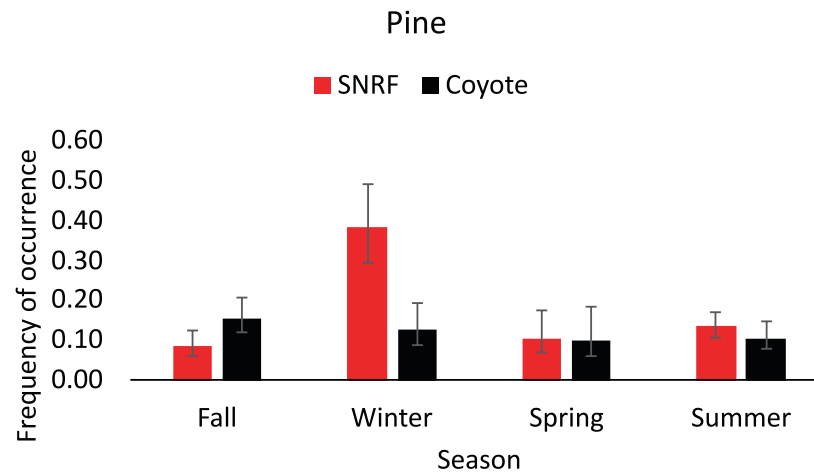


FIGURE 4 | Seasonal frequency of occurrence of (A) white-tailed jackrabbit and (B) pine for Sierra Nevada red fox (SNRF) and coyote scats in the Sierra Nevada from 2011 to 2017. Numbers of scats analysed for vertebrates were 23 and 52 winter, 41 and 29 spring, 130 and 94 summer and 100 and 101 fall SNRF and coyote scats, respectively. Numbers of scats analysed successfully for plant dietary items were 26 and 47 winter, 38 and 30 spring, 119 and 85 summer and 94 and 77 fall SNRF and coyote scats, respectively. Bars in both panels denote standard errors.

particularly in winters of high-snow depth. Below, we evaluate these hypotheses in light of our findings, first those of the two SNRF populations and then those of the Sierra SNRF relative to sympatric carnivores.

4.1 | Lassen and Sierra Nevada SNRF Diets

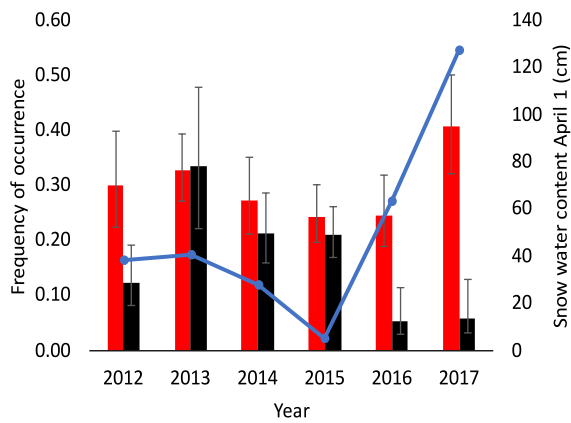
The frequent consumption of snow-adapted lagomorph species by both populations of SNRF was consistent with our prediction that SNRF would specialise on high-elevation lagomorph species with which they historically coexisted and therefore potentially co-evolved. Snow-adapted lagomorphs were within the top 3 diet items for both Lassen and Sierra Nevada SNRF populations. Snowshoe hares were the primary lagomorph consumed at Lassen, while white-tailed jackrabbits were the primary lagomorph consumed at the Sierra Nevada site. Snowshoe hares are rare or absent from the Sierra site, and white-tailed jackrabbits are rare or absent from the Lassen site. Although both study areas contained American pika, they only composed a significant portion of the diet in

the Sierra Nevada site. Pika tend to be more abundant in high-elevation areas (Yandow et al. 2015), which might explain this difference; elevation averaged approximately 1000 m higher in the Sierra (3105 m, range = 2570–3500 m) than in the Lassen site 2100 m (range = 1900–2700 m).

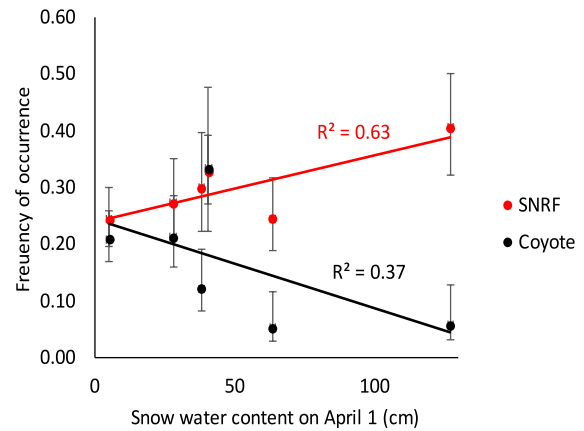
Both Lassen and Sierra Nevada SNRF had high proportions of pine in their scats. The near absence of pine in bobcat scats, a hyper-carnivore that consumed considerably more sciurids than did SNRF (52% vs. 7% biomass consumption, respectively), suggests that most pine occurrences in scats of other mesocarnivores resulted from direct consumption rather than secondary consumption (i.e., consumption of small mammals that consumed pine). Additionally, some prey taxa that are known to consume large amounts of pine nuts, such as ground squirrels and chipmunks, hibernate and were not detected in winter scats, the season when SNRF FOO values for pine were highest.

The Lassen SNRF population exhibited a broader diet than that in the Sierra Nevada population, including frequent consumption of fish, such as trout, chubs and dace. To our knowledge,

A.



B.



C.

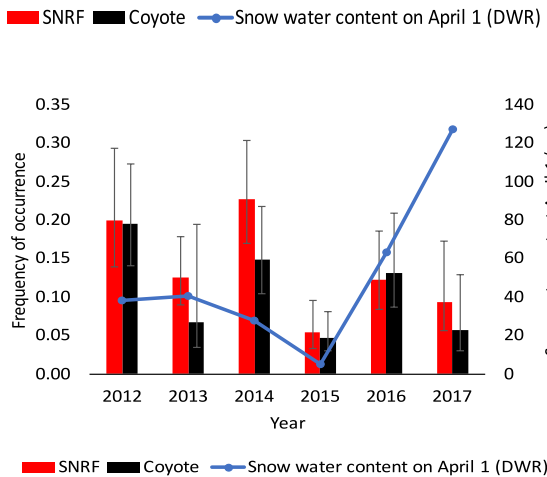


FIGURE 5 | Annual frequency of occurrence of (A, B) whitetail-jackrabbit and (C) pine in relation to annual snowpack for 293 Sierra Nevada red fox (SNRF) and 262 coyote scats in the Sierra Nevada from 2012 to 2017. Bars in all panels denote standard errors. Snowpack was derived from the California Department of Water Resources (<https://cdec.water.ca.gov/reportapp/javareports?name=DLYSWEQ.20180401>).

extensive foraging on non-anadromous freshwater fish has not been previously documented in red foxes and seems generally uncommon in canids. It is possible that ingestion of fish by foxes in our study was derived partly or completely through scavenging. Studies of Great Lakes wolves (*Canis lupus × lycaon*), however, have documented active foraging on freshwater fish under certain conditions, indicating the capacity to do so in canids (Freund et al. 2023). The generally broader diet of Lassen SNRF compared to Sierra SNRF may have reflected the elevational differences between the two populations. A limitation of our study with respect to assessing the importance of lagomorphs to SNRF beyond the Sierra Nevada population was that our Lassen sample included very few winter and spring scats, which is when lagomorphs were predicted to be most significant.

4.2 | Niche Relationships Among Carnivores

Our results revealed multiple prey items potentially important to SNRF populations, only some of which likely facilitated niche partitioning. Frequency of occurrence, estimated % biomass, and RRA data indicated that SNRFs in both the Lassen and the Sierra Nevada sites consumed pocket gophers more than any other prey item; yet pocket gophers were also detected in high

frequencies for coyotes in the Sierra Nevada site, indicating that this prey taxon served as a staple for these canids. Because we lacked specific reference sequences for the two species of pocket gopher present at both sites, we were unable to differentiate them in the diets of the four carnivore species. Thus, we do not know to what extent northern pocket gopher (*T. talpoides*) versus mountain pocket gopher (*T. monticola*) made up the diets of SNRF and how these compositions compared to those of coyotes and the other sympatric carnivores.

As predicted, Sierra Nevada SNRF exhibited the greatest prey overlap with coyote and much lower overlap with bobcat and marten. Compared to SNRFs and coyotes, bobcats and martens more frequently consumed tree and ground squirrels (Sciuridae). Despite high overlap between SNRFs and coyotes, several prey species were identified in our SIMPER analysis as differentiating their diets, including species we predicted based on the body sizes of the two predators and the specialisation of SNRF to the subalpine environment. In terms of body size, coyotes more frequently consumed larger prey items, particularly mule deer and yellow-bellied marmots, whereas SNRF more frequently consumed the smallest prey, principally deer mice. Deer and marmots tend to be unavailable in winter in subalpine habitats and deer mice may be too small to alone support the

energetic demands associated with provisioning pregnant mates or offspring during winter and spring. Indeed, our estimate of the proportion of biomass in the Sierra SNRF diet composed of deer mice was only 7%. In contrast, mid-sized prey, such as lagomorphs, are likely to be especially important during the colder seasons. Despite the relatively large body sizes of white-tailed jackrabbit and American pika, SNRF consumed these lagomorphs more frequently than did coyotes, consistent with specialisation by SNRF on this subalpine-adapted prey.

4.3 | Do Keystone Prey Sustain SNRF Specialists in the Presence of Competition?

The high frequency of white-tailed jackrabbit in SNRF scats in the Sierra across all study years also supported the hypothesis of specialisation by SNRF on this lagomorph and a competitive advantage over coyotes in capturing this prey. Multiannually, SNRF consumed white-tailed jackrabbits at consistently high frequencies, whereas coyotes consumed them almost exclusively in low-snow winters and springs. We found a positive correlation for SNRF and a negative correlation for coyotes between consumption of these lagomorphs and snowpack, consistent with a competitive advantage by SNRF over coyotes for capture of jackrabbits in high-snow conditions. A similar pattern has been described for boreal predators, whereby predation on snowshoe hare by a specialist predator, the Canada lynx, was relatively constant regardless of snow depth, but that by coyotes inversely corresponded to snow depth (Peers et al. 2020).

Although we had no data on annual white-tailed jackrabbit abundance in our study area, it is possible based on other studies of hares in snowy environments that their abundance varied across years (e.g., Hodges 1999; Bartel et al. 2008; Crone et al. 2011). If so, the observed patterns of jackrabbit consumption by SNRF and coyotes also could reflect opportunistic predation by coyotes (i.e., in proportion to abundance, i.e., type I functional response) but selective predation by SNRF on hares (type II functional response) during a declining trend (Holling 1965; Todd and Keith 1983; Spencer et al. 2017). A similar example would be of Canada lynx (specialist) and coyote (generalist) predation on snowshoe hares in boreal Canada, whereby lynx exhibited a type II functional response, consistent with specialisation, but coyotes exhibited a functional response that was equally well characterised by a linear (type I) relationship to snowshoe hare abundance, consistent with a more opportunistic relationship (O'Donoghue et al. 1998). Regardless of the relative roles of snowpack and hare abundance in affecting coyote predation on hares in our study, our finding that most consumption of white-tailed jackrabbit by coyotes occurred during the winters and springs of years when snowpack was light (Figures 4A and 5A) suggests that they represented a secondary prey to coyotes. Altogether, these observations suggest that SNRF are especially reliant on white-tailed jackrabbit prey, presumably more so in the presence of coyote competitors.

Occurrence of pine in scats was significantly higher in winter than in other seasons for SNRF but not for coyotes, also potentially reducing niche overlap between the two canids during winter. We observed variable frequency of occurrence of pine in scats of both carnivores across years, possibly reflecting

annual mast. Although our use of the *trnL* marker was unable to differentiate whitebark pine from several other pine species, western white pine and Jeffrey pine were the only other pines that occurred within the Sierra Nevada site and were generally lower in elevation and less abundant. Thus, it is likely that whitebark pine, in particular, composed most of the pine consumption we documented in the Sierra Nevada site and possibly the Lassen site as well, although Jeffrey pines also were abundant in the Lassen site. The frequencies of pine occurrence in scats across seasons and carnivore species were most consistent with primary consumption, suggesting whitebark pines provide direct nutritional benefit to SNRF, as was indicated for Rocky Mountain red foxes through physical dissection of scat components (Cross and Crabtree 2021).

For competition to occur between species sharing similar diets, prey must be limiting (Chesson 2000; Begon et al. 2003). Although we had no data on prey abundance and the specific energetic needs of the carnivores, our presumption that prey were generally limiting in the Sierra Nevada was supported by a variety of indirect lines of evidence. First and most generally, the subalpine habitats of the Pacific Crest and other mountains are low in productivity and support low diversity of vertebrate and plant species; winters are particularly harsh in such regions (Rahbek 1995; Asner et al. 2014; Schoenherr 1992; Lee et al. 2021). Second, the considerably larger size of SNRF home ranges than coyote home ranges in our Sierra study area indicates that the available resource density is effectively lower for red foxes than for coyotes. The SNRF home range averages 66 km² (range: 22–126 km²; SCAT 2022), many times larger than that of the coyote, the larger-bodied carnivore (approximately 10 km²; B. N. Sacks and C. B. Quinn, unpublished data), and nearly an order of magnitude greater than that of lower-elevation red fox populations (Ables 1969; Storm et al. 1976; Voigt 1987; Lewis et al. 1993). Because home range size is broadly a function of prey availability in carnivores (Ward et al. 2018), it is difficult to explain these observations without invoking competitive exclusion of SNRF by coyotes. In situations where prey items are limiting, niche partitioning is key to co-existence between two similar predators (Chesson 2000; Steinmetz et al. 2021). Thus, despite our lack of direct evidence of competition, which is notoriously difficult to demonstrate in nature, the totality of our findings in this study strongly implicate subalpine lagomorphs as playing a keystone role in allowing SNRF to persist in sympatry with coyotes in their historical range.

4.4 | Conservation Implications

Our study highlighted several species in the diet of SNRF that could be critical to the recovery of endangered SNRF populations in California. In particular, white-tailed jackrabbits, snowshoe hares, American pika and whitebark pine, which are adapted to snow and other harsh winter conditions of the subalpine zone, likely play a key role in sustaining SNRF through winters. We currently have almost no data on population status or trends in abundance and distribution of the lagomorphs, though all are likely facing severe threats from climate change. For example, among better studied populations, both snowshoe hare and white-tailed jackrabbit are exhibiting more severe camouflage mismatch, where their fur colour is no longer synchronised

with snow presence, increasing their vulnerability to predation (Zimova et al. 2020; Ferreira et al. 2023). Such decreases in temperature and snow also could threaten white-tailed jackrabbits both directly and through competition with black-tailed jackrabbits (*Lepus californicus*) as the latter expand their ranges upward in elevation in response to warming temperatures (Brown et al. 2020; Schlater et al. 2021). Climate change and snowpack reduction also have been implicated as causes of northward shifts of the southern range limits of snowshoe hares in Wisconsin (Saultaire et al. 2016). Although effects of climate change on the American pika are less clear (Stewart et al. 2015, 2017; Johnston et al. 2019; Smith 2020), their occurrence in warming high-elevation ecosystems warrants concern and close monitoring.

Additionally, many high-elevation pines are facing challenges driven by climate change (Matías and Jump 2014; Shirk et al. 2018). The whitebark pine has been declining throughout its range due to mountain pine beetle (*Dendroctonus ponderosae*) infestations and white pine blister rust (*Cronartium ribicola*) and it is listed as Threatened pursuant to the Endangered Species Act (U.S. Fish and Wildlife Service 2022). Currently, these threats are most pronounced in the Rocky Mountains, although elevated mountain pine beetle-caused mortality has occurred in northeastern California's Warner Mountains (Meyer et al. 2023). Although pine-beetle infestations are still rare among high-elevation pines in the southern Sierra Nevada (Logan et al. 2010; Nesmith et al. 2019), this situation could change with increasing temperatures, which are required for explosive population growth of pine beetle populations (Millar et al. 2012; Macfarlane et al. 2023). The reduction of prey species and whitebark pine that facilitate niche partitioning between SNRF and coyotes could reduce their ability to co-exist in sympatry, thereby threatening the persistence of SNRF. Monitoring efforts associated with SNRF conservation up to now have emphasised the distribution and abundance of the species itself. While monitoring of SNRF remains a priority, so too should monitoring the distribution, abundance and population trends of the climate-sensitive subalpine specialist species on which it depends for nutritional sustenance and coexistence with an otherwise dominant competitor.

5 | Conclusion

Our study highlights the role of environment-dependent competition in facilitating species coexistence in high-elevation ecosystems. Despite strong dietary overlap between SNRF and coyote in general, competition for snow-adapted prey in particular was lessened during the seasonal periods and years when conditions and prey resources as a whole were likely to be most limiting. These findings underscore a potentially critical ecological linkage: the survival and persistence of alpine specialist predators may closely depend on the continued availability of subalpine-adapted prey species, particularly as climate change allows generalist species to invade or become more numerous in higher elevations. Conversely, disruptions to specialised prey populations due to climate-induced habitat changes could precipitate cascading effects on alpine predator species, highlighting yet another vulnerability of high-elevation communities to climate change. More broadly, preserving specialised predator-prey relationships could be increasingly vital for the conservation of alpine biodiversity in the face of climate change.

Author Contributions

B.N.S., C.B.Q. and G.R.-F. conceived the study; C.B.Q. and P.F. designed and executed field sampling; G.R.-F., C.B.Q., S.L.V. and T.C. performed laboratory analyses. G.R.-F. and B.N.S. analysed the data. G.R.-F. wrote the initial draft. All authors contributed to the writing of latter drafts.

Acknowledgements

Funding for the metabarcoding project was provided from the Christine Stevens Wildlife Award to B.N. Sacks and supplemented through funds of the Mammalian Ecology and Conservation Unit of the Veterinary Genetics Laboratory at the University of California, Davis. Collection of samples from the field and DNA analyses to identify species were supported by the U.S. Fish and Wildlife Service (F16AC0054), U.S. Department of Agriculture (USDA) Forest Service Pacific Southwest Region (12-CS-11052007-021, 16-CS-11051600-023), USDA Forest Service Intermountain Region (11-CS-11041702-046, 16-CS-f 1041730-034), U.S. National Park Service Yosemite National Park (CA-CESU Agreement P15AC01816) and California Department of Fish and Wildlife (P1080019). We thank our many collaborators for their assistance in data collection and logistical support: E. Burkett and C. Stermer (California Department of Fish and Wildlife), S. Eyes, S. Stock and R. Mazur (Yosemite National Park), A. Irvin (U.S. Marine Corps), R. Kalinowski and A. Rich (Stanislaus National Forest), M. Easton, S. Lisius, J. Lowden and A. Orlando (Humboldt-Toiyabe National Forest), K. Boatner (USDA Forest Service Intermountain Region), J. Buckley (Central Sierra Environmental Resource Center), J. Power (field volunteer) and M. Statham and Z. Lounsbury (University of California, Davis) for laboratory assistance. We thank the field and laboratory technicians that made this work possible: C. Angulo, N. Barney, N. Broman, E. Burke, W. Deacy, A. Fitzmorris, M. Fought, M. Holz, N. Goddard, T. Kalani, R. Kuffle, A. Lee, K. Miles, J. Owen, J. Pagano, C. Sanchez, L. Sanders, N. Turner, G. Wardlaw and D. Wolfson. Lastly, we thank C. Aylward, C. White, J. A. Smith, A. Schreier and M. Statham for assistance in study design and helpful edits to early stages of the manuscript.

Disclosure

Benefit Sharing Statement: Genetic resources made available to the public (<https://doi.org/10.5061/dryad.15dv41p7c>) comprise a valuable resource for conservation of biodiversity, both in terms questions as addressed in this study, as well as providing data on lower-trophic biodiversity of the subalpine and alpine study areas. All primary collaborators were included as co-authors. The contributions of all individuals to the research were listed in the Acknowledgements.

Research Material Availability: The authors are willing to share the DNA samples, notes and advice upon reasonable request for non-commercial purposes.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

All data have been made available through the Dryad repository under <https://doi.org/10.5061/dryad.15dv41p7c>. Metadata is also accessioned in Dryad, <https://doi.org/10.5061/dryad.15dv41p7c>.

References

- Ables, E. D. 1969. "Home-Range Studies of Red Foxes (*Vulpes vulpes*).
Journal of Mammalogy 50: 108. <https://doi.org/10.2307/1378635>.
- Altschul, S. F., W. Gish, W. Miller, E. W. Myers, and D. J. Lipman. 1990. "Basic Local Alignment Search Tool." *Journal of Molecular Biology* 215: 403–410. [https://doi.org/10.1016/S0022-2836\(05\)80360-2](https://doi.org/10.1016/S0022-2836(05)80360-2).

- Anderson, M. J., and D. C. I. Walsh. 2013. "PERMANOVA, ANOSIM, and the Mantel Test in the Face of Heterogeneous Dispersions: What Null Hypothesis Are You Testing?" *Ecological Monographs* 83: 557–574. <https://doi.org/10.1890/12-2010.1>.
- Asner, G. P., C. B. Anderson, R. E. Martin, et al. 2014. "Landscape-Scale Changes in Forest Structure and Functional Traits Along an Andes-to-Amazon Elevation Gradient." *Biogeosciences* 11: 843–856. <https://doi.org/10.5194/bg-11-843-2014>.
- Azevedo, F. C. C., V. Lester, W. Gorsuch, S. Larivière, A. J. Wirsing, and D. L. Murray. 2006. "Dietary Breadth and Overlap Among Five Sympatric Prairie Carnivores." *Journal of Zoology* 269: 127–135. <https://doi.org/10.1111/j.1469-7998.2006.00075.x>.
- Bartel, R. A., F. F. Knowlton, and L. C. Stoddart. 2008. "Long-Term Patterns in Mammalian Abundance in Northern Portions of the Great Basin." *Journal of Mammalogy* 89: 1170–1183. <https://doi.org/10.1644/07-MAMM-A-378.1>.
- Begon, M., J. L. Harper, and C. R. Townsend. 2003. *Ecology: Individuals, Populations and Communities*. 3rd ed, reprint. Blackwell.
- Brown, D. E., A. T. Smith, J. K. Frey, and B. R. Schweiger. 2020. "A Review of the Ongoing Decline of the White-Tailed Jackrabbit." *Journal of Fish and Wildlife Management* 11: 341–352. <https://doi.org/10.3996/042019-JFWM-026>.
- Buskirk, S. W., and W. J. Zielinski. 2003. "Small and Mid-Sized Carnivores." In *Mammal Community Dynamics: Management and Conservation in the Coniferous Forests of Western North America*, 207–249. Cambridge University Press.
- Callahan, B. J., P. J. McMurdie, M. J. Rosen, A. W. Han, A. J. A. Johnson, and S. P. Holmes. 2016. "DADA2: High-Resolution Sample Inference From Illumina Amplicon Data." *Nature Methods* 13: 581–583. <https://doi.org/10.1038/nmeth.3869>.
- Caspi, T., M. G. Serrano, S. L. Vanderzwan, J. Kessler, C. J. Schell, and B. N. Sacks. 2025. "Impervious Surface Cover and Number of Restaurants Shape Diet Variation in an Urban Carnivore." *Ecosphere* 16: e70152. <https://doi.org/10.1002/ecs2.70152>.
- Castañeda, I., T. S. Doherty, P. A. Fleming, A. M. Stobo-Wilson, J. C. Z. Woinarski, and T. M. Newsome. 2022. "Variation in Red Fox *Vulpes vulpes* Diet in Five Continents." *Mammal Review* 52: 328–342. <https://doi.org/10.1111/mam.12292>.
- Center for Biological Diversity. 2024. https://ecosphere-documents-production-public.s3.amazonaws.com/sams/public_docs/petition/11760.pdf.
- Chao, A. 1987. "Estimating the Population Size for Capture-Recapture Data With Unequal Catchability." *Biometrics* 43: 783–791.
- Chase, J. M., and M. A. Leibold. 2003. *Ecological Niches: Linking Classical and Contemporary Approaches*. University of Chicago Press.
- Chesson, P. 2000. "Mechanisms of Maintenance of Species Diversity." *Annual Review of Ecology and Systematics* 31: 343–366. <https://doi.org/10.1146/annurev.ecolsys.31.1.343>.
- Clarke, K. R. 1993. "Non-Parametric Multivariate Analyses of Changes in Community Structure." *Australian Journal of Ecology* 18: 117–143. <https://doi.org/10.1111/j.1442-9993.1993.tb00438.x>.
- Crone, E. E., E. J. B. McIntire, and J. Brodie. 2011. "What Defines Mast Seeding? Spatio-Temporal Patterns of Cone Production by Whitebark Pine." *Journal of Ecology* 99: 438–444. <https://doi.org/10.1111/j.1365-2745.2010.01790.x>.
- Cross, P. R., and R. L. Crabtree. 2021. "Recent Findings Suggest Adding Red Fox (*Vulpes vulpes*) to Climate-Threatened Whitebark Pine (*Pinus albicaulis*) Trophic System." *Canadian Journal of Zoology* 99: 618–623. <https://doi.org/10.1139/cjz-2020-0198>.
- Davey, C. M., D. E. Chamberlain, S. E. Newson, D. G. Noble, and A. Johnston. 2012. "Rise of the Generalists: Evidence for Climate Driven Homogenization in Avian Communities." *Global Ecology and Biogeography* 21: 568–578.
- Deagle, B. E., R. Kirkwood, and S. N. Jarman. 2009. "Analysis of Australian Fur Seal Diet by Pyrosequencing Prey DNA in Faeces." *Molecular Ecology* 18: 2022–2038. <https://doi.org/10.1111/j.1365-294X.2009.04158.x>.
- Elton, C., and M. Nicholson. 1942. "The Ten-Year Cycle in Numbers of the Lynx in Canada." *Journal of Animal Ecology* 11: 215–244.
- Ferreira, M. S., T. J. Thurman, M. R. Jones, et al. 2023. "The Evolution of White-Tailed Jackrabbit Camouflage in Response to Past and Future Seasonal Climates." *Science* 379: 1238–1242. <https://doi.org/10.1126/science.ade3984>.
- Ferreiro-Arias, I., J. Isla, P. Jordano, and A. Benítez-López. 2021. "Fine-Scale Coexistence Between Mediterranean Mesocarnivores Is Mediated by Spatial, Temporal, and Trophic Resource Partitioning." *Ecology and Evolution* 11: 15520–15533. <https://doi.org/10.1002/ece3.8077>.
- Ficetola, G. F., J. Pansu, A. Bonin, et al. 2015. "Replication Levels, False Presences and the Estimation of the Presence/Absence From eDNA Metabarcoding Data." *Molecular Ecology Resources* 15: 543–556. <https://doi.org/10.1111/1755-0998.12338>.
- Freund, D. R., T. D. Gable, S. M. Johnson-Bice, A. T. Homkes, S. K. Windels, and J. K. Bump. 2023. "The Ethology of Wolves Foraging on Freshwater Fish in a Boreal Ecosystem." *Royal Society Open Science* 10: 230210.
- Fuhrmann, R. T. 1998. "Distribution, Morphology, and Habitat Use of the Red Fox in the Northern Yellowstone Ecosystem." Doctoral dissertation, Montana State University-Bozeman, College of Letters and Science.
- Fuhrmann, R. T. 2002. "Tracking Down Yellowstone's Red Fox: Skis, Satellites, and Historical Sightings." *Yellowstone Science* 10: 8–15.
- Gilman, S. E., M. C. Urban, J. Tewksbury, G. W. Gilchrist, and R. D. Holt. 2010. "A Framework for Community Interactions Under Climate Change." *Trends in Ecology & Evolution* 25: 325–331.
- Gotelli, N. J., and R. K. Colwell. 2001. "Quantifying Biodiversity: Procedures and Pitfalls in the Measurement and Comparison of Species Richness." *Ecology Letters* 4: 379–391. <https://doi.org/10.1046/j.1461-0248.2001.00230.x>.
- Gotelli, N. J., E. M. Hart, and A. M. Ellison. 2015. "EcoSimR: Null Model Analysis for Ecological Data." R Package Version 0.1.0, 10.
- Grinnell, J., J. S. Dixon, and J. M. Linsdale. 1937. *Fur-Bearing Mammals of California: Their Natural History, Systematic Status, and Relations to Man*. Volume II. University of California Press.
- Hodges, K. E. 1999. "Ecology of Snowshoe Hares in Southern Boreal and Montane Forests [Chapter 7]." In *Ecology and Conservation of Lynx in the United States*. Gen. Tech. Rep. RMRS-GTR-30WWW, edited by L. F. Ruggiero, K. B. Aubry, S. W. Buskirk, et al., 163–206. US Department of Agriculture, Forest Service, Rocky Mountain Research Station.
- Holling, C. S. 1965. "The Functional Response of Predators to Prey Density and Its Role in Mimicry and Population Regulation." *Memoirs of the Entomological Society of Canada* 97: 5–60. <https://doi.org/10.4039/entm9745fv>.
- Jensen, A. J., E. A. Saldo, Z. G. Chapman, J. W. Butfiloski, and D. S. Jachowski. 2023. "Risk From a Top Predator and Forest Structure Influence Scavenging by Smaller Carnivores." *Ecosphere* 14: e4596. <https://doi.org/10.1002/ecs2.4596>.
- Johnston, A. N., J. E. Bruggeman, A. T. Beers, E. A. Beever, R. G. Christophersen, and J. I. Ransom. 2019. "Ecological Consequences of Anomalies in Atmospheric Moisture and Snowpack." *Ecology* 100: e02638. <https://doi.org/10.1002/ecy.2638>.
- Knowles, N., M. D. Dettinger, and D. R. Cayan. 2006. "Trends in Snowfall Versus Rainfall in the Western United States." *Journal of Climate* 19: 4545–4559. <https://doi.org/10.1175/JCLI3850.1>.

- Lamb, P. D., E. Hunter, J. K. Pinnegar, S. Creer, R. G. Davies, and M. I. Taylor. 2019. "How Quantitative Is Metabarcoding: A Meta-Analytical Approach." *Molecular Ecology* 28: 420–430. <https://doi.org/10.1111/mec.14920>.
- Lee, M. A., G. Burger, E. R. Green, and P. W. Kooij. 2021. "Relationships Between Resource Availability and Elevation Vary Between Metrics Creating Gradients of Nutritional Complexity." *Oecologia* 195: 213–223. <https://doi.org/10.1007/s00442-020-04824-4>.
- Levi, T., and C. C. Wilmers. 2012. "Wolves-Coyotes-Foxes: A Cascade Among Carnivores." *Ecology* 93: 921–929.
- Levin, S. A., and R. T. Paine. 1974. "Disturbance, Patch Formation, and Community Structure." *Proceedings of the National Academy of Sciences of the United States of America* 71: 2744–2747. <https://doi.org/10.1073/pnas.71.7.2744>.
- Levins, R. 1979. "Coexistence in a Variable Environment." *American Naturalist* 114: 765–783. <https://doi.org/10.1086/283527>.
- Lewis, J. C., K. L. Sallee, and R. T. Golightly. 1993. *Introduced Red Fox in California*. State of California, the Resources Agency, Department of Fish and Game, Wildlife Management Division.
- Logan, J. A., W. W. Macfarlane, and L. Willcox. 2010. "Whitebark Pine Vulnerability to Climate-Driven Mountain Pine Beetle Disturbance in the Greater Yellowstone Ecosystem." *Ecological Applications* 20: 895–902. <https://doi.org/10.1890/09-0655.1>.
- Macarthur, R., and R. Levins. 1967. "The Limiting Similarity, Convergence, and Divergence of Coexisting Species." *American Naturalist* 101: 377–385. <https://doi.org/10.1086/282505>.
- Macfarlane, W. W., B. Howell, J. A. Logan, A. L. Smith, C. C. Rasmussen, and R. E. Spangler. 2023. "Climate Change-Driven Cumulative Mountain Pine Beetle-Caused Whitebark Pine Mortality in the Greater Yellowstone Ecosystem." *Forests* 14: 2361. <https://doi.org/10.3390/f14122361>.
- Major, J. T., and J. A. Sherburne. 1987. "Interspecific Relationships of Coyotes, Bobcats, and Red Foxes in Western Maine." *Journal of Wildlife Management* 51: 606–616. <https://doi.org/10.2307/3801278>.
- Martin, M. 2011. "Cutadapt Removes Adapter Sequences From High-Throughput Sequencing Reads." *EMBnet.Journal* 17: 10. <https://doi.org/10.14806/ej.17.1.200>.
- Masters, H. M., and C. R. Maher. 2022. "Diet Reveals Potential for Competition and Coexistence Among Coyotes (*Canis latrans*), Red Foxes, (*Vulpes vulpes*) and Gray Foxes (*Urocyon cinereoargenteus*)." *Canadian Journal of Zoology* 100: 90–97. <https://doi.org/10.1139/cjz-2021-0103>.
- Matías, L., and A. S. Jump. 2014. "Impacts of Predicted Climate Change on Recruitment at the Geographical Limits of Scots Pine." *Journal of Experimental Botany* 65: 299–310. <https://doi.org/10.1093/jxb/ert376>.
- McLaren, T. H., D. F. Tomback, N. Grevstad, et al. 2023. "Clark's Nutcracker Forest Community Visitation: Whitebark Pine Maintains a Keystone Seed Disperser." *Ecology and Evolution* 13: e10813.
- Meyer, M. D., M. R. Slaton, S. E. Gross, R. J. Butz, and C. Clark. 2023. "Ecological Integrity of Whitebark Pine Ecosystems in California's National Forests." *Canadian Journal of Forest Research* 53: 328–342.
- Millar, C. I., R. D. Westfall, D. L. Delany, M. J. Bokach, A. L. Flint, and L. E. Flint. 2012. "Forest Mortality in High-Elevation Whitebark Pine (*Pinus albicaulis*) Forests of Eastern California, USA; Influence of Environmental Context, Bark Beetles, Climatic Water Deficit, and Warming." *Canadian Journal of Forest Research* 42: 749–765. <https://doi.org/10.1139/x2012-031>.
- Murray, D. L., and S. Boutin. 1991. "The Influence of Snow on Lynx and Coyote Movements: Does Morphology Affect Behavior?" *Oecologia* 88: 463–469. <https://doi.org/10.1007/BF00317707>.
- Neale, J. C. C., and B. N. Sacks. 2001a. "Food Habits and Space Use of Gray Foxes in Relation to Sympatric Coyotes and Bobcats." *Canadian Journal of Zoology* 79: 1794–1800. <https://doi.org/10.1139/z01-140>.
- Neale, J. C. C., and B. N. Sacks. 2001b. "Resource Utilization and Interspecific Relations of Sympatric Bobcats and Coyotes." *Oikos* 94: 236–249. <https://doi.org/10.1034/j.1600-0706.2001.940204.x>.
- Nesmith, J. C. B., M. Wright, E. S. Jules, and S. T. McKinney. 2019. "Whitebark and Foxtail Pine in Yosemite, Sequoia, and Kings Canyon National Parks: Initial Assessment of Stand Structure and Condition." *Forests* 10: 35. <https://doi.org/10.3390/f10010035>.
- Newsome, T. M., and W. J. Ripple. 2015. "A Continental Scale Trophic Cascade From Wolves Through Coyotes to Foxes." *Journal of Animal Ecology* 84: 49–59. <https://doi.org/10.1111/1365-2656.12258>.
- Niittynen, P., R. K. Heikkinen, and M. Luoto. 2018. "Snow Cover Is a Neglected Driver of Arctic Biodiversity Loss." *Nature Climate Change* 8: 997–1001. <https://doi.org/10.1038/s41558-018-0311-x>.
- O'Donoghue, M., S. Boutin, C. J. Krebs, G. Zuleta, D. L. Murray, and E. J. Hofer. 1998. "Functional Responses of Coyotes and Lynx to the Snowshoe Hare Cycle." *Ecology* 79: 1193–1208.
- Oksanen, J., G. L. Simpson, F. G. Blanchet, et al. 2024. "Vegan: Community Ecology Package (Version 2.6-8) [Computer Software]." <https://cran.r-project.org/web/packages/vegan/index.html>.
- Olsen, R. L., K. Øverbø, and B. Myrnes. 1991. "Alkaline Phosphatase From the Hepatopancreas of Shrimp (*Pandalus borealis*): A Dimeric Enzyme With Catalytically Active Subunits." *Comparative Biochemistry and Physiology Part B: Comparative Biochemistry* 99: 755–761. [https://doi.org/10.1016/0305-0491\(91\)90139-5](https://doi.org/10.1016/0305-0491(91)90139-5).
- Palomares, F., and T. M. Caro. 1999. "Interspecific Killing Among Mammalian Carnivores." *American Naturalist* 153: 492–508. <https://doi.org/10.1086/303189>.
- Parker, A. J. 1991. "Forest/Environment Relationships in Lassen Volcanic National Park, California, USA." *Journal of Biogeography* 18: 543. <https://doi.org/10.2307/2845690>.
- Parnesan, C. 2006. "Ecological and Evolutionary Responses to Recent Climate Change." *Annual Review of Ecology, Evolution, and Systematics* 37: 637–669. <https://doi.org/10.1146/annurev.ecolsys.37.091305.110100>.
- Peers, M. J. L., Y. N. Majchrzak, A. K. Menzies, et al. 2020. "Climate Change Increases Predation Risk for a Keystone Species of the Boreal Forest." *Nature Climate Change* 10: 1149–1153. <https://doi.org/10.1038/s41558-020-00908-4>.
- Perrine, J. D. 2005. "Ecology of Red Fox (*Vulpes vulpes*) in the Lassen Peak Region of California, United States of America." Dissertation, University of California, Berkeley.
- Perrine, J. D., L. A. Campbell, and G. A. Green. 2010. "Sierra Nevada Red Fox (*Vulpes vulpes necator*): A Conservation Assessment." R5-FR-010. US Department of Agriculture.
- Peterson, M., M. Baglieri, K. Mahon, et al. 2021. "The Diet of Coyotes and Red Foxes in Southern New York." *Urban Ecosystems* 24: 1–10. <https://doi.org/10.1007/s11252-020-01010-5>.
- Pianka, E. R. 1973. "The Structure of Lizard Communities." *Annual Review of Ecology and Systematics* 4: 53–74. <https://doi.org/10.1146/annurev.es.04.110173.000413>.
- Piggott, M. P. 2004. "Effect of Sample Age and Season of Collection on the Reliability of Microsatellite Genotyping of Faecal DNA." *Wildlife Research* 31: 485–493.
- Quinn, C. B., P. B. Alden, and B. N. Sacks. 2019. "Noninvasive Sampling Reveals Short-Term Genetic Rescue in an Insular Red Fox Population." *Journal of Heredity* 110: 559–576. <https://doi.org/10.1093/jhered/esz024>.
- Quinn, C. B., S. Preckler-Quisquater, J. A. Akins, et al. 2022. "Contrasting Genetic Trajectories of Endangered and Expanding Red Fox Populations in the Western U.S." *Heredity* 129: 123–136.

- Quinn, C. B., S. Preckler-Quisquater, M. R. Buchalski, and B. N. Sacks. 2024. "Whole Genomes Inform Genetic Rescue Strategy for Montane Red Foxes in North America." *Molecular Biology and Evolution* 41: msae193. <https://doi.org/10.1093/molbev/msae193>.
- Rahbek, C. 1995. "The Elevational Gradient of Species Richness: A Uniform Pattern?" *Ecography* 18: 200–205. <https://doi.org/10.1111/j.1600-0587.1995.tb00341.x>.
- Riaz, T., W. Shehzad, A. Viari, F. Pompanon, P. Taberlet, and E. Coissac. 2011. "Ecoprimers: Inference of New DNA Barcode Markers From Whole Genome Sequence Analysis." *Nucleic Acids Research* 39: e145. <https://doi.org/10.1093/nar/gkr732>.
- Rödger, D., T. Schmitt, P. Gros, W. Ulrich, and J. C. Habel. 2021. "Climate Change Drives Mountain Butterflies Towards the Summits." *Scientific Reports* 11: 14382.
- Schlater, S. M., J. M. Ringenberg, N. Bickford, and D. H. Ranglack. 2021. "White-Tailed Jackrabbits: A Review and Call for Research." *Southwestern Naturalist* 65: 161–172. <https://doi.org/10.1894/0038-4909-65.2.161>.
- Schoenherr, A. A. 1992. *A Natural History of California*. Univ of California Press.
- Shehzad, W., T. Riaz, M. A. Nawaz, et al. 2012. "Carnivore Diet Analysis Based on Next-Generation Sequencing: Application to the Leopard Cat (*Prionailurus bengalensis*) in Pakistan." *Molecular Ecology* 21: 1951–1965. <https://doi.org/10.1111/j.1365-294X.2011.05424.x>.
- Shirk, A. J., S. A. Cushman, K. M. Waring, et al. 2018. "Southwestern White Pine (*Pinus strobiformis*) Species Distribution Models Project a Large Range Shift and Contraction due to Regional Climatic Changes." *Forest Ecology and Management* 411: 176–186. <https://doi.org/10.1016/j.foreco.2018.01.025>.
- Shulgina, T., A. Gershunov, B. J. Hatchett, et al. 2023. "Observed and Projected Changes in Snow Accumulation and Snowline in California's Snowy Mountains." *Climate Dynamics* 61: 4809–4824. <https://doi.org/10.1007/s00382-023-06776-w>.
- Sierra Nevada Red Fox Conservation Advisory Team [SCAT]. 2022. "A Conservation Strategy for the Sierra Nevada Red Fox." California Department of Fish and Wildlife, Sacramento, USA. https://www.dfw.state.or.us/wildlife/management_plans/docs/A%20Conservation%20Strategy%20for%20the%20Sierra%20Nevada%20Red%20Fox_2022.pdf.
- Smith, A. T. 2020. "Conservation Status of American Pikas (*Ochotona princeps*)." *Journal of Mammalogy* 101: 1466–1488. <https://doi.org/10.1093/jmammal/gyaa110>.
- Smith, J. A., A. C. Thomas, T. Levi, Y. Wang, and C. C. Wilmers. 2018. "Human Activity Reduces Niche Partitioning Among Three Widespread Mesocarnivores." *Oikos* 127: 890–901. <https://doi.org/10.1111/oik.04592>.
- Spencer, E. E., T. M. Newsome, and C. R. Dickman. 2017. "Prey Selection and Dietary Flexibility of Three Species of Mammalian Predator During an Irruption of Non-Cyclic Prey." *Royal Society Open Science* 4: 170317. <https://doi.org/10.1098/rsos.170317>.
- Steinmetz, R., N. Seuaturien, P. Intanajitjuy, P. Inrueang, and K. Prempre. 2021. "The Effects of Prey Depletion on Dietary Niches of Sympatric Apex Predators in Southeast Asia." *Integrative Zoology* 16: 19–32. <https://doi.org/10.1111/1749-4877.12461>.
- Stewart, J. A. E., J. D. Perrine, L. B. Nichols, et al. 2015. "Revisiting the Past to Foretell the Future: Summer Temperature and Habitat Area Predict Pika Extirpations in California." *Journal of Biogeography* 42: 880–890. <https://doi.org/10.1111/jbi.12466>.
- Stewart, J. A. E., D. H. Wright, and K. A. Heckman. 2017. "Apparent Climate-Mediated Loss and Fragmentation of Core Habitat of the American Pika in the Northern Sierra Nevada, California, USA." *PLoS One* 12: e0181834. <https://doi.org/10.1371/journal.pone.0181834>.
- Storm, G. L., R. D. Andrews, R. L. Phillips, R. A. Bishop, D. B. Siniff, and J. R. Tester. 1976. "Morphology, Reproduction, Dispersal, and Mortality of Midwestern Red Fox Populations." *Wildlife Monographs* 49: 3–82.
- Sultaire, S. M., J. N. Pauli, K. J. Martin, M. W. Meyer, M. Notaro, and B. Zuckerberg. 2016. "Climate Change Surpasses Land-Use Change in the Contracting Range Boundary of a Winter-Adapted Mammal." *Proceedings of the Royal Society B: Biological Sciences* 283: 20153104. <https://doi.org/10.1098/rspb.2015.3104>.
- Taberlet, P., E. Coissac, F. Pompanon, et al. 2007. "Power and Limitations of the Chloroplast trnL (Uaa) Intron for Plant DNA Barcoding." *Nucleic Acids Research* 35: e14. <https://doi.org/10.1093/nar/gkl938>.
- Todd, A. W., and L. B. Keith. 1983. "Coyote Demography During a Snowshoe Hare Decline in Alberta." *Journal of Wildlife Management* 47: 394–404. <https://doi.org/10.2307/3808512>.
- U.S. Fish and Wildlife Service. 2022. <https://www.federalregister.gov/documents/2022/12/15/2022-27087/endangered-and-threatened-wildlife-and-plants-threatened-species-status-with-section-4d-rule-for>.
- U.S. Fish and Wildlife Service Region 8 Sacramento, CA. 2018. "Species Status Assessment Report for the Sierra Nevada Distinct Population Segment of the Sierra Nevada Red Fox." <https://www.sierraforestlegacy.org/Resources/Conservation/SierraNevadaWildlife/SierraNevadaRedFox/USFWS-SpeciesStatusReport-SNRedFox2018.pdf>.
- Ugland, K. I., J. S. Gray, and K. E. Ellingsen. 2003. "The Species–Accumulation Curve and Estimation of Species Richness." *Journal of Animal Ecology* 72: 888–897. <https://doi.org/10.1046/j.1365-2656.2003.00748.x>.
- Urban, M. C. 2018. "Escalator to Extinction." *Proceedings of the National Academy of Sciences of the United States of America* 115: 11871–11873. <https://doi.org/10.1073/pnas.1817416115>.
- Urban, M. C., J. J. Tewksbury, and K. S. Sheldon. 2012. "On a Collision Course: Competition and Dispersal Differences Create No-Analogue Communities and Cause Extinctions During Climate Change." *Proceedings of the Royal Society B: Biological Sciences* 279: 2072–2080.
- Van Etten, K. W., K. R. Wilson, and R. L. Crabtree. 2007. "Habitat Use of Red Foxes in Yellowstone National Park Based on Snow Tracking and Telemetry." *Journal of Mammalogy* 88: 1498–1507. <https://doi.org/10.1644/07-MAMM-A-076.1>.
- Vestheim, H., and S. N. Jarman. 2008. "Blocking Primers to Enhance PCR Amplification of Rare Sequences in Mixed Samples – A Case Study on Prey DNA in Antarctic Krill Stomachs." *Frontiers in Zoology* 5: 12. <https://doi.org/10.1186/1742-9994-5-12>.
- Voigt, D. R. 1987. "Red Fox. Chapter 30." In *Wild Furbearer Management and Conservation in North America*, edited by M. Novak, 379–382. Ministry of Natural Resources; Ontario Trappers Association.
- Ward, J. N., J. W. Hinton, K. L. Johannsen, M. L. Karlin, K. V. Miller, and M. J. Chamberlain. 2018. "Home Range Size, Vegetation Density, and Season Influences Prey Use by Coyotes (*Canis latrans*)." *PLoS One* 13: e0203703. <https://doi.org/10.1371/journal.pone.0203703>.
- Webster, S. C., M. J. Chamberlain, J. W. Hinton, and J. C. Beasley. 2021. "Isotope Analysis Reveals Dietary Overlap Among Sympatric Canids." *Journal of Mammalogy* 102: 1222–1234. <https://doi.org/10.1093/jmammal/gyab077>.
- White, C. Q., J. P. Bush, and B. N. Sacks. 2023. "Deer Dietary Responses to Wildfire: Optimal Foraging, Individual Specialization, or Opportunism?" *Molecular Ecology* 32: 6953–6968. <https://doi.org/10.1111/mec.17185>.
- Yandow, L. H., A. D. Chalfoun, and D. F. Doak. 2015. "Climate Tolerances and Habitat Requirements Jointly Shape the Elevational Distribution of the American Pika (*Ochotona princeps*), With Implications for Climate Change Effects." *PLoS One* 10: e0131082. <https://doi.org/10.1371/journal.pone.0131082>.

Zhang, J. L., and K. P. Ma. 2014. "Spaa: An R Package for Computing Species Association and Niche Overlap." *Research Progress of Biodiversity Conservation in China* 10: 165–174.

Zimova, M., A. P. K. Sirén, J. J. Nowak, et al. 2020. "Local Climate Determines Vulnerability to Camouflage Mismatch in Snowshoe Hares." *Global Ecology and Biogeography* 29: 503–515. <https://doi.org/10.1111/geb.13049>.

Supporting Information

Additional supporting information can be found online in the Supporting Information section. **Data S1:** mec70087-sup-0001-Supinfo.pdf.