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EDITORIAL

Applications of stable isotope analysis in mammalian ecology

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In this editorial, we provide a brief introduction and summarize the 10 research articles included in this Special Issue on *Applications of stable isotope analysis in mammalian ecology*. The first three articles report correction and discrimination factors that can be used to more accurately estimate the diets of extinct and extant mammals using stable isotope analysis. The remaining seven applied research articles use stable isotope analysis to address a variety of wildlife conservation and management questions from the oceans to the mountains.

Keywords: animals; carbon-13; diet; isotope ecology; mammals; nitrogen-15; sulfur-34

1. Introduction

Stable isotope analysis is an emerging method used to investigate the trophic structure of food webs, ecological niche of species, and diets of animals. Unlike the recent effort to promote the theoretical and analytical advances in stable isotope research in mammals (*Journal of Mammalogy*, Volume 93, Issue 3), we provide a collection of studies in this Special Issue to encourage the use of stable isotope analysis in applied mammalian ecology. The following articles have been solicited from mammalogists and stable isotope practitioners and serve to promote the use of this expanding field in mammalian ecology.

This collection, selected by the editor, provides methodological advances in stable isotope analysis and its applied use in mammalian research. For instance, this Special Issue provides correction factors that account for changes in atmospheric CO_2 over time for more accurate comparisons of isotopes measured in ancient and modern mammalian tissues and includes results from experiments that investigate the effects of size, age, sex, and tissue- and diet-types on stable isotope discrimination factors. The applied research that follows includes studies that use stable isotopes to address ecological questions used to inform wildlife conservation and management. These range from assessing wild carnivore interactions with humans to estimating the foraging ecology of endangered species to combining DNA metabarcoding of stomach contents with stable isotope analysis of consumer tissues to investigate relationships between population density and

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diet. As a collection, the papers presented in this Special Issue further our understanding of how to use stable isotope analysis in mammalian research.

2. Correction and discrimination factors in mammals diet

Bocherens et al. [1] examined the potential caveats for comparing stable isotope values from hair in modern species to those from fossil bone collagen in ancient species. Using data from a recent study that investigated the carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotope values in bone collagen from cave bears (*Ursus spelaeus*) from the European Late Pleistocene [2], the authors identified three corrections that are needed before comparing modern hair and ancient bone collagen: (a) calibrating tissues to the global decrease in $\delta^{13}C$ values in atmospheric CO_2 due to burning fossil fuels, (b) recognizing differences in discrimination factors between both diet and hair keratin and diet and bone collagen, and (c) accounting for different assimilation rates of isotopes from foods into bone collagen (long-term) and hair keratin (shorter-term). Bocherens et al. [1] incorporated these three corrections into their study and conclude that Romanian cave bears were likely not foraging on high trophic-level foods.

Hobson and Quirk [3] estimated the stable carbon ($\Delta^{13}C$) and nitrogen ($\Delta^{15}N$) isotope discrimination factors between diets and eight different tissues extracted from captive striped skunks (*Mephitis mephitis*) held on known diets for 7 months (adults) and 50 days (newborn young). The authors found that tissue type, age, and amount of preferred food (ration) had a significant effect on the $\Delta^{15}N$ values, whereas tissue type was the only significant factor affecting $\Delta^{13}C$ values. Despite these between-group differences, variation in isotope discrimination factors within experimental groups was fairly constrained. Their data are valuable for deciphering the diet of mid-size mammalian omnivores across age classes using stable isotopes. The authors also provide perspective on the importance of understanding the potential for variance around stable isotope discrimination factors and point out that probabilistic-based stable isotope mixing models can incorporate these variations when used to estimate diets of animals.

Kurle et al. [4] also examined factors driving differences in stable carbon and nitrogen isotope discrimination factors associated with multiple tissues from omnivorous rats held on four diets of comparable protein quality and quantity. Similar to Hobson and Quirk [3], Kurle et al. [4] also found that tissue type accounted for some of the variability observed in $\Delta^{13}C$ and $\Delta^{15}N$ values. Diet type also controlled differences in discrimination factors. For $\Delta^{13}C$, the carbon source mattered, whereas for $\Delta^{15}N$, the protein source mattered. Sex may play a role in driving $\Delta^{15}N$, but this was likely due to increased growth in males over the course of the study. The authors demonstrate that, when using stable isotopes to investigate mammalian omnivore foraging ecology, it is important to consider that: (a) the sources of dietary carbon affect $\Delta^{13}C$, (b) the dietary trophic level at which a mammalian omnivore is foraging affects $\Delta^{15}N$, and (c) the $\Delta^{15}N$ factors associated with tissues with fast isotope turnover rates are not affected by dietary sources.

3. Use of stable isotope analysis in wildlife conservation and management

Hopkins et al. [5] measured the $\delta^{13}C$, $\delta^{15}N$, and sulfur isotope ($\delta^{34}S$) values in hair of threatened grizzly bears (*Ursus arctos*) from Banff National Park to determine if stable isotope analysis can be used as a tool to identify bears that forage along the Canadian Pacific Railway Canada. The authors found that bears killed by trains or captured along the rail had significantly higher $\delta^{15}N$ and $\delta^{34}S$ values compared to conspecifics sampled away from the rail, suggesting that rail-associated bears consumed more animal protein (e.g. train-killed ungulates) and sulfur pellets (directly or

indirectly via plants or animals), respectively. Results from this study indicate that stable isotope analysis could be used as a non-invasive, affordable, and efficient technique to monitor bears that forage on the railway in Banff and potentially other transportation networks worldwide.

Karamanlidis et al. [6] investigated foraging patterns of critically endangered Mediterranean monk seals (*Monachus monachus*) in Greece using stable isotope analysis of their hair. The authors compared the $\delta^{13}C$ and $\delta^{15}N$ values from seal hair to those from the muscle tissue of their known prey. As predicted, the $\delta^{13}C$ values for seals were similar to their coastal prey species and higher than their pelagic prey. The authors concluded that seals have a diverse diet comprising prey from multiple trophic levels found primarily along the coast. They recommend that marine resource managers use these results to reinforce the importance of protecting coastal resources in Greece for the benefit of Mediterranean monk seals.

Seamster et al. [7] used $\delta^{13}C$ and $\delta^{15}N$ values from the hair of small mammals extracted from the scats of coyotes (*Canis latrans*) to examine short-term vegetation changes in diet in central New Mexico in the USA. The authors used a mixing model [8] to assess the contribution of C_4 plants to the diets of the coyote prey over three seasons in both shrubland and grassland communities. They found that C_4 grasses increased in prey diets in the fall and were consumed less in shrubland areas. Seasonal variation in diet appears to be a response to increased live grass cover in the study area following rainfall events in both the summer and fall. These findings have implications for the availability of food resources for coyote mammalian prey in the face of continued woody plant encroachment within the refuge.

Soininen et al. [9] used the $\delta^{13}C$ and $\delta^{15}N$ values in consumer muscle combined with DNA metabarcoding of consumer stomach contents to assess the diets of five populations of small rodents in the Arctic. Their purpose was to examine the relationship between population density and diversity of diet within populations. The authors found little evidence suggesting that diet diversity expands in primary habitats as population densities of small rodents increase. Instead, they detected a density-driven increase of resource use in secondary habitats, leading to an increase in diet diversity within some populations. In conclusion, the authors suggest that the combined use of stable isotope analysis and DNA metabarcoding can be a powerful method for investigating the composition and temporal variation of animal diets (particularly terrestrial herbivores), but their concurrent use should be carefully assessed based on the specific study systems and questions.

Teunissen van Manen et al. [10] described the use of stable isotopes from fur to compare foraging ecology of wild and captive American black bears (*Ursus americanus*) that occupy remote areas vs campgrounds and picnic areas in a national park in the southwestern USA. The authors modeled the relationship between $\delta^{13}C$ and $\delta^{15}N$ values from bear hair and nine environmental variables. They found no significant relationships between $\delta^{13}C$ values and the variables they analyzed. In addition, the authors did not observe any differences in isotope values between wild bears sampled in 1980–2001 and those captured in campgrounds or picnic areas, suggesting nuisance bears do not forage for anthropogenic foods in the park. They did, however, find that $\delta^{15}N$ values were higher in bear hair when acorn (*Quercus* spp.) production was low and lower in bear hair when acorn production was high.

Walter [11] examined the $\delta^{13}C$ and $\delta^{15}N$ values in muscle and hoof tissue from Rocky Mountain elk (*Cervus elaphus*) to identify subpopulations that occupied either a C_4 plant-dominated landscape or a C_3 plant-dominated agricultural landscape in the Great Plains of the USA. Elk in the C_4-dominated landscape were partially confined within a national wildlife refuge, providing a unique opportunity to test for differences in the $\delta^{13}C$ and $\delta^{15}N$ values between these elk and two nearby subpopulations that occupied private lands dominated by C_3 agricultural plants. The differences in the $\delta^{13}C$ and δ^{15} N values across all years for elk foraging on C_3 agricultural plants points to a greater consumption of forages higher in percent nitrogen, suggesting the potential for higher nutritive value for elk foraging in agriculturally dominated landscapes compared to native landscapes. Walter [11] underscores the application of stable isotope analysis to identify

animal affinities to regions and for the delineation of mammalian subpopulations using terrestrial, vegetative isoscapes [12].

Warsen et al. [13] used stable isotope analysis of hair to investigate the diets of four sympatric carnivore populations occurring in the Adirondack Park in New York. The authors related isotopes in carnivore hair to relative measures of known behavior. They reported that carnivores with the highest human tolerance (red and gray foxes) exhibited the highest $\delta^{13}C$ values in their hair, reflecting corn-rich (C_4) diets comprising human-derived foods. Conversely, bobcats, with the lowest human tolerance, exhibited the lowest $\delta^{13}C$ values in their hair, indicating diets composed of animals that consumed C_3 plants in the northeast USA. Their research points to the utility of this non-invasive method for understanding the relative impacts people, and their foods, have on wildlife.

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