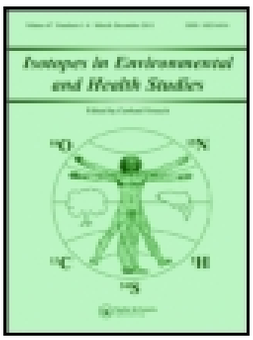

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Does faecal matter reflect location? An initial assessment of isotopic variability between consumed prey remains and faecal matter for wild jaguars*

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ABSTRACT

Faecal isotopic analysis may complement other non-invasive wildlife survey tools for monitoring landscape use by carnivores, such as motion-detecting cameras and non-invasive genetic sampling. We analysed carbon, nitrogen, and strontium isotopes in faecal matter produced by jaguars (*Panthera onca*) as well as bones from consumed prey at the Mountain Pine Ridge Forest Reserve (MPR) in Belize, Central America. The MPR is ideally suited for a spatial isotope study as vegetation and geology both vary considerably. The isotopic composition of faecal matter should reflect the habitat and geology where consumed prey lived. We used bone from consumed prey recovered from jaguar scats as a proxy for diet. Faecal matter and bone showed comparable spatial isotopic trends, suggesting that the isotopic composition of jaguar faeces can be used to detect foraging in different habitats (pine forest versus broadleaf forest) or on different geologies (Mesozoic carbonates; Palaeozoic granite, contact metamorphics, and metasediments). This result is reassuring as bones are not always present in carnivore scats. Studying landscape use by cryptic and wide-ranging carnivore species like jaguars remains challenging. Isotopic analysis of faecal matter complements the existing array of non-invasive spatial monitoring tools.

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1. Introduction

As anthropogenic habitat removal and degradation increasingly transform the surface of our planet, our need to monitor spatial ecology of threatened species has never been more pressing. This is particularly pertinent for rare, cryptic, and wide-ranging apex predators, such as big cats, which play key ecological roles in the ecosystems they inhabit [e.g. 1,2]. An increasing array of survey techniques is available to study carnivore spatial ecology, including monitoring animal signs, remote motion-sensing camera traps, radio

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and GPS telemetry, non-invasive genetic sampling, and stable isotope analysis [e.g. 3–9]. Those methods that allow researchers to track individuals non-invasively, such as the analysis of faecal matter, may be particularly advantageous.

Wultsch et al. [10–12] genetically identified species, sex, and individuals for faecal samples from multiple neotropical felid taxa in Belize to estimate genetic diversity, gene flow, and population densities. If paired with genetic analysis, isotope analysis of faecal matter may be a particularly powerful method for non-invasively tracking foraging patterns for solitary, cryptic, and rare species [13]. This has not been previously assessed. Here, we conducted a preliminary investigation of the degree to which carbon ($\delta^{13}C$), nitrogen ($\delta^{15}N$), and strontium ($^{87}Sr/^{86}Sr$) isotopes in faecal matter from genetically identified wild jaguars (*Panthera onca*) match isotopes in consumed prey remains, which should ultimately allow us to track individual landscape use. Specifically, carbon and nitrogen isotopes should differentiate foraging in moister, closed forest and drier, more open habitats [reviewed in 14], while strontium isotopes should track foraging in areas underlain by different geologies [e.g. 4,15–18]. Jaguars are ideally suited for this ground-truthing study since they are large-bodied, obligate carnivores and utilize a variety of habitats, including scrub, savannah, grassland, wetland, and agricultural land [e.g. 19–23].

1.1. Background on isotope values in faeces

The isotopic composition of carnivore faecal matter should be dominated by an individual's most recent meal [24,25]. On the basis of this assumption, researchers have previously used $\delta^{13}C$ and $\delta^{15}N$ values in faeces to investigate dietary variability within and among individuals [26–31], as well as habitat use for animals living in localities with a mixture of C_3 and C_4 biomes [e.g. 32–34]. However, because faecal matter is composed of undigested material, it may differ isotopically from consumed or assimilated diet, which could bias dietary estimates based on faeces [28,30,35,36]. We expect this to be a negligible issue for obligate carnivores, like jaguars, which almost exclusively consume animal matter, have simple digestive systems, and short gut retention times [e.g. 25,37]. Nevertheless, different animal tissues differ isotopically [reviewed in 14], and can vary considerably in their digestibility [e.g. 38–41]. Additionally, sloughed gut lining and microbes, as well as fractionation associated with digestion, may all result in an isotopic offset between consumed prey and faecal matter for predators [28,30,36,42,43].

One solution would be to only analyse undigested prey remains in faecal samples (e.g. nails, hooves, or bone fragments from consumed prey), and account for expected isotopic differences among tissues [e.g. 38,39,44]. However, the abundance of undigested prey remains in carnivore scats is variable (C. Wultsch personal observation). Faecal matter, or matrix, may therefore be a more widely available medium for isotope analysis [25,30,42].

In order to use faecal matter as a proxy for diet, we must first validate that it accurately reflects diet. Interest in applying strontium isotopes to ecological research is a relatively recent development. We are aware of just one study (on domestic pigs) that compared $^{87}Sr/^{86}Sr$ in diet and faecal matter [44]. On average, the offset in strontium isotopes between diet and faeces ($\Delta^{87}Sr/^{86}Sr_{diet-faeces}$) ranged from −0.000004 to 0.000051 for groups of pigs fed different diets that included varying amounts of marine-derived

protein; when all data were combined, average $^{87}Sr/^{86}Sr_{diet–faeces}$ was 0.000001. Conversely, researchers have spent over three decades investigating diet–faeces offset for carbon and nitrogen isotopes ($\Delta^{13}C_{diet–faeces}$ and $\Delta^{15}N_{diet–faeces}$) for a diversity of captive and wild mammals (Table 1). Overall, there are similar trends in $\Delta^{13}C_{diet–faeces}$ and $\Delta^{15}N_{diet–faeces}$ among taxa; faecal matter has lower $\delta^{13}C$ values and higher $\delta^{15}N$ values than bulk diet for most taxa. However, these trends are by no means universal and there is considerable variability in isotopic offset both within and among species. Some taxa even show the opposite trends, where faecal matter has higher $\delta^{13}C$ and lower $\delta^{15}N$ values than diet. This variability may reflect differences in digestive physiology, nutritional composition and digestibility of diet, or study design.

The majority of research investigating $\Delta^{13}C_{diet–faeces}$ and $\Delta^{15}N_{diet–faeces}$ has focused on large-bodied herbivores and small-bodied taxa like bats and rodents (Table 1). Very few researchers have examined $\Delta_{diet–faeces}$ in larger-bodied omnivores or carnivores. Montanari and Amato [42] analysed carbon and nitrogen isotope values in faecal matter from captive tigers (*Panthera tigris*) and snow leopards (*Panthera uncia*) as well as the premixed commercial feed that made up the majority of their diet. Their results are in stark contrast to the majority of taxa that have been studied to date (Table 1). On average, faecal matter for both species had higher $\delta^{13}C$ values than the commercial feed and $\delta^{13}C$ and $\delta^{15}N$ values in tiger faecal matter spanned nearly 6 ‰. In their paper, the authors speculated that this variability reflects differences in nutritional balance among individuals. Of course, these unexpected results may also stem from an isotopically variable diet; $\delta^{13}C$ values and $\delta^{15}N$ values for seven subsamples of feed ranged >7.5 ‰ and 1.75 ‰, respectively. The authors confirmed that feed samples were obtained from a few different batches during the same week (S. Montanari personal communication). However, they did not state if particular faecal samples and feed were collected at the same time.

More recently, Montanari [52] analysed $\delta^{13}C$ and $\delta^{15}N$ values in faeces from captive meerkats (*Suricata suricatta)* as well as samples of their various food items (whole chicks, mice, horse meat, carrots, apples, and dog biscuits). Her analysis of dietary items provided a good indication of the isotopic variability of consumed foods. However, because faeces were collected opportunistically from a common enclosure, it is not known when, exactly, each scat was produced, or by whom. The relative amount of food items provided to the meerkats varied from day to day, making it difficult to tease apart available diet from consumed diet. Faecal $\delta^{13}C_{faeces}$ and $\delta^{15}N_{faeces}$ values ranged 7.3 and 4.5 ‰, respectively. It is not possible to tease apart the degree to which these results may reflect preferential consumption of certain foods or physiological differences among individuals. Using the average isotopic composition of all possible dietary items, Montanari [52] estimated that there was no offset between faeces and diet and only a small offset for nitrogen based on average offset values (Table 1). However, offset values for individual faeces ranged from −4.2 to 3.2 ‰ for carbon and −4.3 to 0.2 ‰ for nitrogen. Given the isotopic variability in both diet and faecal isotope data, we would be hesitant to apply the average offset values in other contexts.

Reid and Koch [30] used a different approach. These authors estimated $\Delta^{13}C_{diet–faeces}$ and $\Delta^{15}N_{diet–faeces}$ values for coyotes (*Canis latrans*) using faeces–hair offsets calculated for four wild coyotes, combined with hair to dietary protein offsets that had previously been calculated for captive red foxes (*Vulpes vulpes*) and wolves (*Canis lupus*) [38,55].

Table 1. Published differences in $\delta^{13}C$ and $\delta^{15}N$ values between diet and faecal matter for mammals.

Taxon	Diet type	Wild or captive?	#Individuals/ samples	$\Delta^{13}C_{diet-faeces}$ (‰)	$\Delta^{15}N_{diet-faeces}$ (‰)	Source
Domestic goat (*Capra hircus*)	Herbivore/ browser	Captive	24/24[b]	0.5 to 3.9[a,b]		[36]
Alpaca (*Vicugna pacos*)	Herbivore/ grazer	Captive	4/	0.4 ± 0.4; 1.3 ± 0.2[a,c]		[45]
Aurochs (*Bos primigenius taurus*)	browser	Captive	4/	1.0 ± 0.2; 0.9 ± 0.2[a,c]		[45]
Domestic goat (*Capra hircus*)	Herbivore/ browser	Captive	4/	0.8 ± 0.1; 1.0 ± 0.4[a,c]		[45]
Llama (*Llama llama*)	Herbivore/ mixed	Captive	4/	0.4 ± 0.5 1.2 ± 0.4[a,c]		[45]
European rabbit (*Oryctolagus cuniculus*)	Herbivore/ mixed	Captive	4/	0.3 ± 0.1[a]		[45]
Horse (*Equus caballus*)	Herbivore/ grazer	Captive	4/	0.5 ± 0.4; 0.7 ± 0.2[a,c]		[45]
Llama (*Llama llama*)	Herbivore/ mixed	Captive	4/		−2.9 ± 0.3; −3.0 ± 0.4 [a,c]	[46]
Horse (*Equus caballus*)	Herbivore/ grazer	Captive	1/		−2.6; −3.3[a,c]	[46]
Domestic sheep (*Ovis aries*)	Herbivore/ grazer	Captive	4/4		−2.8 to −3.1	[47]
Wild boar (*Sus scrofa*)	Trophic omnivore	Captive	3/3		−1.2 ± 0.4	[48]
Aurochs (*Bos primigenius taurus*)	Herbivore/ grazer	Captive	4;4;6/4;4;6[d]		−1.4 ± 0.3; −0.6 ± 0.3; −0.4 ± 0.2[d]	[48]
Aurochs (*Bos primigenius taurus*)	Herbivore/ grazer	Captive	8/8		−1.9 ± 0.3[e]	[49]
Long-tailed vole (*Microtus longicaudus*)	Herbivore/ mixed	Captive	5/5	2.7[f]	−2.3[f]	[43]
Western jumping mouse (*Zapus princeps*)	Herbivore/ mixed	Captive	5/5	5.9[f]	−2.3[f]	[43]
Yellow-pine chipmunk (*Tamias amoenus*)	Trophic omnivore	Captive	5/5	3.0[f]	−1.4[f]	[43]
Deer mouse (*Peromyscus maniculatus*)	Trophic omnivore	Captive	5/5	3.2[f]	−2.2[f]	[43]
Meadow vole (*Microtus pennsylvanicus*)	Trophic omnivore	Captive	5/5	3.6[f]	−2.6[f]	[43]
Red-backed vole (*Myodes gapperi*)	Trophic omnivore	Captive	5/5	4.2[f]	−2.2[f]	[43]
Red-backed vole (*Myodes gapperi*)	Trophic omnivore	Captive	11;10;10/ 11;10;10[g]	−0.2 ± 0.4; 1.2 ± 0.3; 0.5 ± 0.4[g]	−1.3 ± 0.7; −1.2 ± 0.5; −1.8 ± 0.4[g]	[50]
Common chimpanzee (*Pan troglodytes*)	Trophic omnivore	Captive	5/33	0.9 ± 0.3; 2.1 ± 0.4[h]	−1.9 ± 0.4; −1.8 ± 0.4[h]	[51]
Meerkat (*Suricata suricatta*)	Trophic omnivore	Captive	7[i]/24	−0.1 ± 1.5	−1.5 ± 1.1	[52]
Greater mouse-eared bat (*Myotis myotis*)	Insectivore	Captive	3/15;21[j]	0.2 ± 1.1; 0.3 ± 0.8[j]	−1.8 ± 1.3; −2.3 ± 2.2[j]	[25]
Greater horseshoe bat (*Rhinolophus ferrumequinum*)	Insectivore	Captive	3/14;21[j]	0.2 ± 1.0; 0.1 ± 0.4[j]	−0.5 ± 0.5; −1.0 ± 0.5[j]	[25]
Tiger (*Panthera tigris*)	Carnivore	Captive	7/7	−1.2 ± 3.3	−1.5 ± 2.1	[42]
	Carnivore	Captive	10/10	−2.3 ± 3.6[k]	−2.4 ± 1.5[k]	[42]

(*Continued*)

Table 1. Continued.

Taxon	Diet type	Wild or captive?	#Individuals/ samples	$\Delta^{13}C_{diet-faeces}$ (‰)	$\Delta^{15}N_{diet-faeces}$ (‰)	Source
Snow leopard (*Panthera uncia*)						
Human (*Homo sapiens*)	Trophic omnivore	Captive	14;14;4/Not specified[k]	1.1 (0.8 to 1.5); 0.4 (0.2 to 0.6); 1.5 (1.3 to 1.7)[l]	0.4 (0.2 to 1.0); −0.4 (−0.6 to −0.1); −1.2 (−2.1 to −1.8)[l]	[53]
François' langur (*Trachypithecus francoisi*)	Herbivore/ mixed	Captive	1/82[m]	−1.3 ± 1.9	−1.0 ± 3.8	[54]
Mountain gorilla (*Gorilla beringei*)	Herbivore/ folivore	Wild	4/121	−0.3	−0.7	[29]
Common chimpanzee (*Pan troglodytes*)	Trophic omnivore	Wild	10/115	−1.3 ± 0.7[n]	−0.5 ± 0.7[n]	[31]
Coyote (*Canis latrans*)	Trophic omnivore	Wild	4	1.5 ± 1.6[o]	−2.3 ± 1.3[o]	[30]

[a]Some authors calculate apparent enrichment (ε^*) rather than simple offset (Δ), where $\varepsilon^* = [(10^3 + \delta^{13}C_{faeces})/(10^3 + \delta^{13}C_{diet}) - 1]*10^3$. The equations yield comparable results.
[b]Raw data not provided. These are average values for small groups of goats (3–6 individuals) that were fed diets with differing amounts of C_3 and C_4 foods. Offset values were similar for animals fed pure C_3 or C_4 diets (on the order of 0.5–0.7 ‰).
[c]Averages reported for two different diets: alfalfa (*Medicago sativa*), and Bermuda grass (*Cynodon dactylon*)
[d]Average ranges reported for groups fed three different diets.
[e]Average lumps pairs of animals fed different diets (pasture, pressed cake, hay, and sillage).
[f]Animals were wild-caught, provided with a transition diet (sunflower seeds, oats, and apples) for 2 days, and standard rodent chow diet for 7 days. Faecal samples were collected after 7 days.
[g]Three groups fed diets with differing levels of protein for 62 ± 12 days: high (26 %), medium (17 %); and low (14 %). Note that the authors called this species *Clethrionomys gapperi*; *Myodes* is now preferred.
[h]Diet–tissue offsets are provided using whole 'atomic' diet as well as just dietary protein.
[i]Samples were collected opportunistically from an enclosure containing seven individuals.
[j]Individuals fed two isotopically distinct diets for several days each.
[k]These values differ slightly from those reported by the authors due to rounding differences and correct propagation of error.
[l]Groups fed three different diets: Fish, Meat, and 'Vegetarian'. Raw data not provided; Median and interquartile range are reported.
[m]Only the adult female included; didn't include data from the infant in this study.
[n]Averages estimated using mean data provided for each individual.
[o]Diet was estimated using $\Delta_{scat-fur}$ for coyotes combined with $\Delta_{fur-diet}$ estimates for red foxes and wolves.

On average, faecal matter $\delta^{13}C$ values were 1.5 ± 1.6 ‰ lower and $\delta^{15}N$ values were 2.3 ± 1.3 ‰ higher than estimated diet (Table 1). The authors did not provide diet–faeces offset estimates for individual scats.

Here, we tested a third approach: We used bone from consumed prey recovered from faecal matter as a proxy for diet in wild jaguars. This method is advantageous because it allowed us to account for potential variability among individuals and meals. We compared isotope data in faecal matter with $\delta^{13}C$ and $\delta^{15}N$ values in bone collagen, and $\delta^{13}C$ and $^{87}Sr/^{86}Sr$ in bone bioapatite. The $\delta^{13}C$ value of bioapatite is influenced by all dietary components, including protein, structural carbohydrates, and lipids while that of bone collagen is biased towards dietary protein [reviewed in 56,57]. Because dietary macronutrients differ isotopically, these two bony tissues differ isotopically. We included both $\delta^{13}C_{collagen}$ and $\delta^{13}C_{apatite}$ because they are complementary and may be useful for tracking the degree to which different dietary components are integrated into faecal matter.

2. Materials and methods

2.1. Study area

Our study took place at the Mountain Pine Ridge Forest Reserve (MPR) in central Belize (Figure 1). MPR encompasses ca. 430 km^2 and is the oldest protected area in Belize. It is located within the rain-shadow of the Maya Mountains. The dry season, which is characterized by monthly rainfall <150 mm, generally lasts from February to May and is frequently accompanied by forest fires (Johnson and Chaffey 1973). The rainy season lasts from June through January, and monthly rainfall peaks in September and October. Climate data are currently only available for Baldy Beacon, which is located in eastern MPR and is the reserve's highest point at 1017 m above sea level. Between 2005 and 2016, average annual rainfall was 2122 mm and average temperatures ranged from 17.4 to 25.1 °C (data provided by the National Meteorological Service of Belize).

MPR is well suited for a spatial isotope study because vegetation and geology vary considerably across the reserve (Figure 1). Lowland and submontane pine forest is prevalent in northwestern and central MPR [58,59]. Patches of submontane broadleaf forest occur in eastern MPR and lowland broadleaf forest is found in western MPR and in patches along its southeastern edge. The primary geologies are Silurian granite in western and northwestern MPR, Late Palaeozoic clay-rich sediments in southern and eastern MPR [60,61]. Mesozoic carbonates and contact metamorphics outcrop in western and central MPR, respectively (Figure 1). These differences in vegetation and geology should lead to spatial isotopic variability across MPR. Pine forest, which often has a sparse canopy, a more open understory and is fire prone, should have higher δ^{13}C and δ^{15}N values than broadleaf forest, which is moister and has a more closed canopy. Carbonates would be

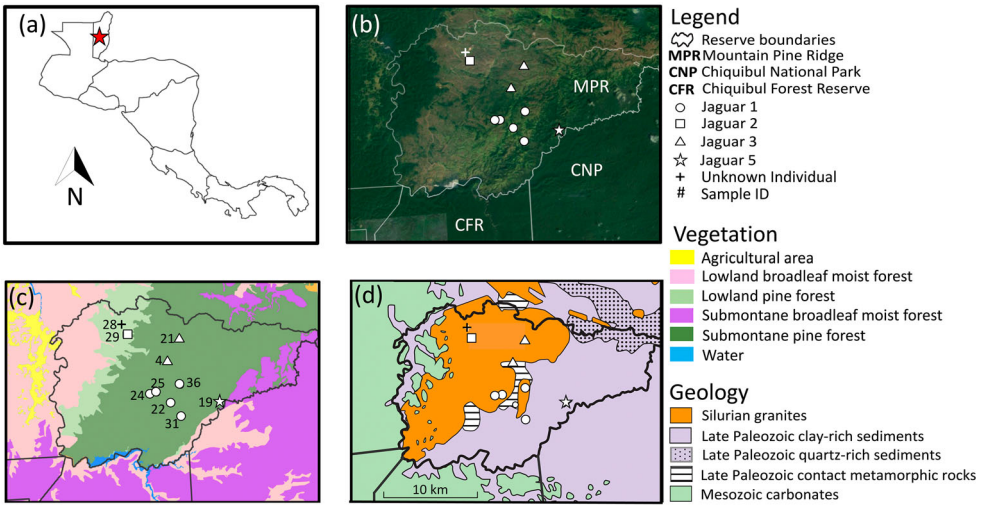

Figure 1. Maps showing (a) the location of the study area, (b) current vegetation cover, (c) vegetation types, and (d) underlying geology at Mountain Pine Ridge Forest Reserve and the surrounding region in Belize, Central America. Locations for faecal samples produced by individual jaguars are provided using a unique symbol for each individual. Satellite map from Google Earth, vegetation map from [58], and geology map adapted from J Cornec (unpubl).

expected to have relatively low $^{87}Sr/^{86}Sr$, granites should have high $^{87}Sr/^{86}Sr$, and clay-rich sediments and metamorphics should have intermediate ratios (reviewed in [18]).

2.2. Faecal sample collection and analysis

Jaguar faecal samples were collected opportunistically at MPR using a professionally trained scat-detecting dog from June to August 2007 for a conservation genetics and population density study focused on jaguars and four other neotropical felids [10–12]. Scats were detected during opportunistic searches that included repeated sampling along roads, trails and various landscape features (e.g. streams, ridgelines) roughly every 10 days. We recorded the geographic location of each scat via a handheld GPS unit. All scats were recovered from lowland or submontane pine forest located on granite, clay-rich sediments, or contact metamorphics (Table 2; Figure 1). We assessed the relative age of each sample based on its condition (colour, presence of mould and degree of degradation) and the amount of time that had passed since the last search had occurred. An aliquot of each scat was stored in DET buffer for genetic analysis [11]; the remaining scat was dried in the field and stored frozen after shipping to the United States for additional analysis. Genetic identification of species, sex, and individual for all faecal samples was previously conducted at the Laboratory for Ecological, Evolutionary and Conservation Genetics at the University of Idaho [10]. We selected 10 jaguar scats that contained bone chunks for isotopic analysis. These were genetically assigned to four males ('Jaguar 1', 'Jaguar 2', 'Jaguar 3', 'Jaguar 5') and one unidentified individual (sex unknown). Faecal matter was homogenized using an agate mortar and pestle (after removing bone, hair, and nail), and weighed for isotopic analysis. A single bone chunk from each faecal sample was cleaned using ultrapure water and dried. Separate portions of each bone sample were processed and chemically

Table 2. Information for individual jaguar scats. Vegetation and geology estimated based on sample location and available maps (see Figure 1).

Scat ID	Individual	Region	Vegetation	Geology	Date collected	Age class
4	Jaguar 3	Central MPR	Submontane pine forest	Contact metamorphics	6/27/07	1–2 days
19	Jaguar 5	Southeast MPR	Submontane pine forest	Late Palaeozoic clay-rich seds.	7/9/07	>2 days
21	Jaguar 3	North Central MPR	Submontane pine forest	Silurian granite	7/10/07	<24 h
22	Jaguar 1	Central MPR	Submontane pine forest	Contact metamorphics	7/13/07	1–2 days
24	Jaguar 1	Central MPR	Submontane pine forest	Silurian granite	7/15/07	<24 h
25	Jaguar 1	Central MPR	Submontane pine forest	Silurian granite	7/15/07	>2 days
28	Unknown Jaguar	Northwest MPR	Lowland pine forest	Silurian granite	7/18/07	>2 days
29	Jaguar 2	Northwest MPR	Lowland pine forest	Silurian granite	7/18/07	1–2 days
31	Jaguar 1	Southeast MPR	Submontane pine forest	Late Palaeozoic clay-rich seds.	7/19/07	>2 days
36	Jaguar 1	Central MPR	Submontane pine forest	Silurian granite	7/19/07	>2 days

pre-treated to isolate the mineral (bioapatite) and protein (collagen) components. For bioapatite, roughly 20 mg of each bone sample were powdered using a Dremel rotary tool equipped with a dental drill bit. Samples were then pre-treated following Crowley and Wheatley [62]. Samples were first soaked in a 30 % hydrogen peroxide solution at room temperature for 72 h (the solution was refreshed between 24 and 48 h) to remove organics. Samples were agitated regularly and lids were left loose to let evolved gas escape. They were rinsed 5× with ultrapure water and then soaked in 1 M calcium-buffered acetic acid for 24 h at 4 °C to remove non-lattice bound carbonates. Samples were again rinsed 5× with ultrapure water and lyophilized. For bone collagen, 50–100 mg of each sample were removed using tin snips. Samples were chemically pre-treated following Crowley et al. [62]. They were soaked in 0.5 M hydrochloric acid at 4 °C until they were soft and pliable (roughly 5 days), and rinsed 5× using ultrapure water. Next, samples were repeatedly sonicated in petroleum ether until all visible lipids were removed. Samples were again rinsed 5× with ultrapure water, lyophilized, and weighed for isotope analysis.

We analysed carbon and nitrogen isotopes in collagen and faecal matter at the University of Cincinnati Stable Isotope Biogeochemistry Facility on a Costech Elemental Analyser connected to a Thermo Scientific Delta V IRMS via a Costech Conflo IV interface. We weighed ca. 0.4 mg of bone collagen and ca. 3 mg of dried faecal matter into tin boats prior to analysis. We corrected data for linearity and drift using caffeine. Variability in scale was normalized using caffeine and USGS 41. Accuracy (based on independent references glycine and gelatine) was 0.16 ‰ for carbon and 0.26 ‰ for nitrogen. Precision (based on all four reference materials) was 0.13 ‰ for carbon and 0.17 ‰ for nitrogen. The average difference in $\delta^{13}C$ and $\delta^{15}N$ values between runs for two samples run in duplicate was 0.47 ‰ and 0.37 ‰, respectively.

Carbon isotopes in bone bioapatite were analysed at the Stable Isotope Mass Spec Laboratory in the Department of Geological Sciences, University of Florida on a Finnigan-MAT 252 isotope ratio mass spectrometer equipped with a Thermo Scientific Kiel III carbonate device. Approximately 1 mg of bioapatite was dissolved in five drops of 100 % phosphoric acid at 70 °C for five minutes, and the evolved CO_2 was analysed. Precision, based on 10 NBS-19 replicates, was 0.022 ‰. The difference in $\delta^{13}C$ values between runs for a single sample run in duplicate was 0.13 ‰.

Strontium in bioapatite and faecal matter was extracted and analysed in the Department of Geology Isotope Geochemistry Laboratory at the University of Illinois, Urbana-Champaign. Between 3 and 5 mg of bone and 12–16 mg of dried faecal matter were first dissolved in nitric acid. Bone was dissolved in 0.5 mL of 3 M nitric acid while faecal samples were dissolved in 1 mL of 8 M nitric acid. Samples were heated in Teflon beakers on a hot plate at 127 °C for ∼1.5 h and then filtered through Teflon mini-columns containing Eichrom Sr-specific resin. They were then sequentially rinsed with 0.05, 3, and 8 M nitric acid to selectively remove cations from the resin to isolate strontium. Strontium was eluted from the resin with 3 mL of ultrapure water and 1 mL of 0.05 M nitric acid into 4 mL autosampler vials. Samples were analysed on a Nu Plasma high resolution multicollector inductively coupled plasma mass spectrometer. Data were corrected for drift using the international standard NBS 987. Two internal laboratory references ('Coral' and 'E&A') were used to monitor accuracy and precision. Analytical uncertainty for all samples was ±0.00005.

2.3. Data analysis

We created three age classes for faecal samples based on sample condition (<24 h, 1–2 days, >2 days). Considering our search schedule, it is highly unlikely that any samples were >10 days old. We then calculated the isotopic offset (Δ) between bone and faecal matter (e.g. $\Delta^{13}C_{collagen–faeces} = \delta^{13}C_{collagen} – \delta^{13}C_{faeces}$) and compared $\Delta^{13}C_{apatite–faeces}$, $\Delta^{13}C_{collagen–faeces}$, $\Delta^{15}N_{collagen–faeces}$, $\Delta^{87}Sr/^{86}Sr_{apatite–faeces}$, and atomic C:N ratios among age classes. We compared $\delta^{13}C$ and $\delta^{15}N$ values for bone and faeces between vegetation types (lowland pine forest, and submontane pine forest [58]), and $^{87}Sr/^{86}Sr$ data among geologies. We also examined spatial isotopic variability among regions where samples were recovered (northwest MPR, north-central MPR, central MPR, and southeast MPR), which should reflect spatial variability in both habitat and geology where scats were recovered (Figure 1). We used Bartlett tests to assess homoscedasticity. Because sample sizes were small, we used non-parametric Wilcoxon rank sum or Kruskal–Wallis tests to compare groups. Where relevant, we also used post-hoc Steel–Dwass All Pairs tests. We also ran separate discriminant function analyses (DFA) for bone collagen and faeces to assess the confidence with which we can discriminate regions at MPR using carbon, nitrogen, and strontium isotopes. All analyses were performed in JMP Pro 13.0.0 and statistical significance was set at $\alpha = 0.05$.

3. Results

Atomic C:N of faecal matter ranged from 4.2 to 7.3 % and was comparable for relatively fresh and older scats (Figure 2). Median atomic C:N did not differ among groups ($\chi^2 = 3.02$, df = 2, $P = 0.22$). Atomic C:N was significantly more variable for samples thought to be >2 days old (Bartlett $P = 0.034$); both the smallest and largest values belonged to this group.

We found considerable isotopic variability among both bone and faecal samples (Supplementary Table 3; Figures 2–4). On average, $\delta^{13}C_{faeces}$ values (−23.3 ± 1.4 ‰) were significantly lower than those in apatite (−14.7 ± 2.9 ‰), but variances did not differ (Bartlett $P > 0.05$). Faecal $\delta^{13}C$ values were also significantly lower and less variable than those in collagen (−18.7 ± 3.7 ‰; Figure 3). However, for one sample (#19 from Jaguar 5) $\delta^{13}C_{collagen}$ was lower than $\delta^{13}C_{faeces}$ (Supplementary Table 3; Figure 3). Average $\Delta^{13}C_{apatite–faeces}$ was 8.0 ± 2.7 ‰ (range = 4.5 to 13.0 ‰), while $\Delta^{13}C_{collagen–faeces}$ is 4.0 ± 3.2 ‰ (range = −2.5 to 8.2 ‰). Conversely, $\delta^{15}N_{faeces}$ values (10.0 ± 2.0 ‰) were significantly higher than those in collagen (7.2 ± 2.2 ‰; Figure 3); variance was similar between the two materials (Bartlett $P > 0.05$). Average $\Delta^{15}N_{collagen–faeces}$ was −2.8 ± 2.5 ‰ (range = −7.0 to 1.3 ‰). Lastly, $^{87}Sr/^{86}Sr$ for apatite (0.72882 ± 0.01309) and faecal matter (0.72667 ± 0.01030) were statistically indistinguishable; average $\Delta^{87}Sr/^{86}Sr_{apatite–faeces}$ was 0.00216 ± 0.00673 (range = −0.00620 ± 0.01797; Supplementary Table 3; Figure 3).

There were no significant differences in median isotopic offsets among various-aged scats (Kruskal Wallis $P > 0.05$ for all comparisons; Figure 2). Samples thought to be >2 days old had apparently more variable offsets than other groups, but this was not significant (Bartlett $P > 0.05$ for all comparisons). There were also no significant differences in median carbon and nitrogen isotope values for faeces, carbon and nitrogen isotope values for bones, or carbon and nitrogen isotopic offsets among habitats (Bartlett $P > 0.05$ and

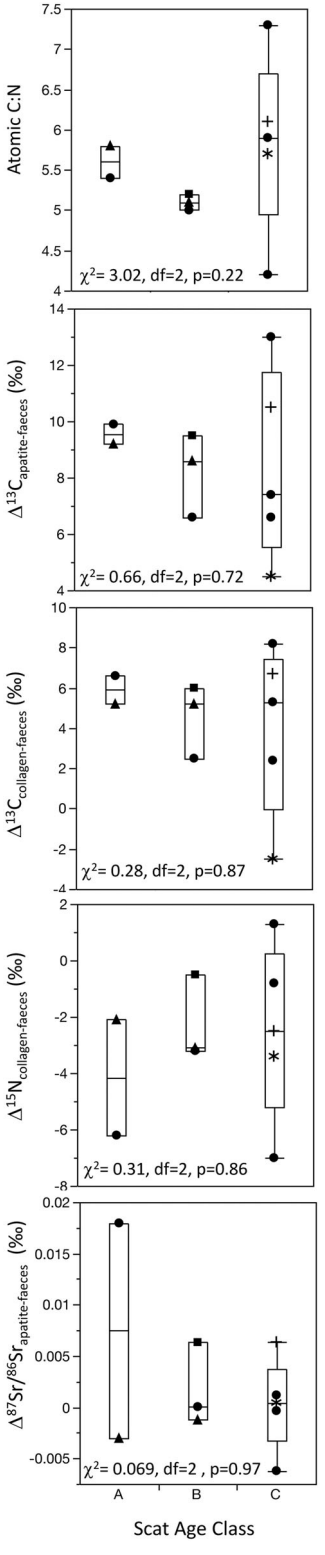

Figure 2. Box and whisker plots comparing atomic C:N, and isotopic offset ($\Delta_{bone-faeces}$) among faecal age classes: $A \leq 24$ h; $B = 1–2$ days; $C \geq 2$ days. Boxes represent 25 % and 75 % quartiles, and whiskers contain 1.5 times the interquartile range. Symbols for individual jaguars correspond with Figure 1.

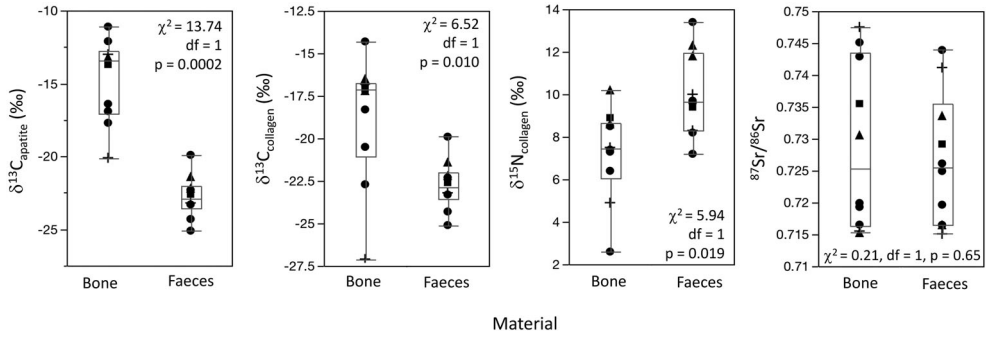

Figure 3. Box and whisker plots comparing isotope data for bone and faecal matter. Boxes represent 25 % and 75 % quartiles, and whiskers contain 1.5 times the interquartile range. Symbols for individual jaguars correspond with Figure 1.

Kruskal–Wallis $P > 0.05$ for all comparisons; Figure 4). Samples from submontane pine forest had larger isotopic ranges than those from lowland forest (Supplementary Figure 1), but this may simply reflect the larger sample size available for submontane pine forest ($n = 8$ versus $n = 2$). Similarly, there were no significant differences in median $^{87}Sr/^{86}Sr_{apatite}$, $^{87}Sr/^{86}Sr_{faeces}$, or $\Delta^{87}Sr/^{86}Sr_{apatite–faeces}$ among geologies (Kruskal–Wallis $P > 0.05$ for all comparisons; Supplementary Figure 2). Variances contrasted considerably among groups, but these apparent differences were only significant for $^{87}Sr/^{86}Sr_{faeces}$; scats collected on contact metamorphics had less variable $^{87}Sr/^{86}Sr_{faeces}$ than those collected on Late Palaeozoic clay-rich sediments or Silurian granite (Bartlett $P = 0.0027$).

Comparing different regions within MPR, we found that variances were equal among all groups (including individual materials as well as isotopic offset between bone and faeces) with the exception of $\Delta^{87}Sr/^{86}Sr_{apatite–faeces}$ (Bartlett $P = 0.0024$); $\Delta^{87}Sr/^{86}Sr_{apatite–faeces}$ was more variable for faeces collected in central MPR than other regions (Figure 6). Median isotope data were statistically indistinguishable among regions (Kruskal–Wallis $P > 0.05$ for all comparisons; Figure 6). However, there were apparent spatial trends in the data, and overall, these were similar for bone and faeces (Figure 6). Carbon and nitrogen isotope values tended to be lower for southeast MPR, higher for northern and northwestern MPR, and most variable for central MPR. An exception to this was sample #31, which was recovered in southeastern MPR and produced by Jaguar 1. The $\delta^{13}C_{apatite}$ value for this sample was considerably higher than $\delta^{13}C_{faeces}$. Strontium isotope data were quite variable for all regions of MPR where more than one data point was available (Figure 4). However, values <0.72 were only observed in central and southeastern MPR. Overall, strontium isotope data for bone and faeces were quite similar. One sample (#24), which was recovered from central MPR and produced by Jaguar 1, had considerably higher $^{87}Sr/^{86}Sr_{apatite}$ than $^{87}Sr/^{86}Sr_{faeces}$ (Table 2; Figure 4).

Because samples from northwestern and north-central MPR had indistinguishable $\delta^{13}C$, $\delta^{15}N$, and negligible apparent differences in $^{87}Sr/^{86}Sr$, we combined samples from these two regions in our discriminant function analyses. The DFA for bone collagen classified 100 % of the samples correctly (Figure 5; Supplementary Tables 1 and 2). The DFA for faeces was not significant (Supplementary Table 1). However, it still did reasonably well at correctly classifying region. Only one sample from Central MPR (#36) was misclassified as southeastern MPR (Figure 5; Supplementary Table 2).

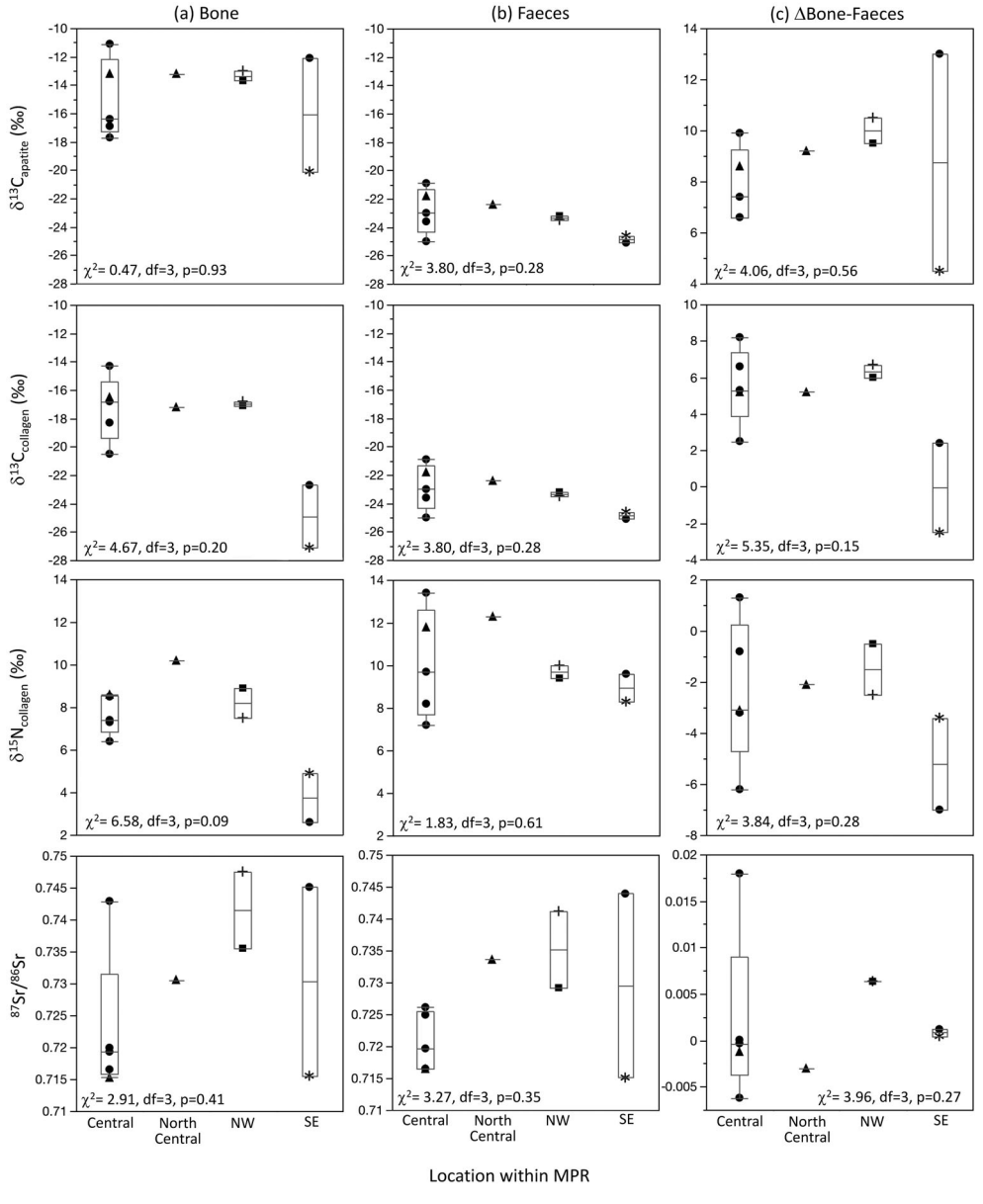

Figure 4. Box and whisker plots comparing isotope data for (a) bone, (b) faecal matter, and (c) $\Delta_{bone-faeces}$ among regions at MPR. Boxes represent 25 % and 75 % quartiles, and whiskers contain 1.5 times the interquartile range. Symbols for individual jaguars correspond with Figure 1.

4. Discussion

We set out to establish the isotopic differences between bone from consumed prey and jaguar faecal matter, and to test the ability of carbon, nitrogen, and strontium isotopes in faecal matter for tracking landscape use of wild jaguars. This work builds on previous studies that estimated diet–faeces offsets for wild coyotes, and captive tigers, snow leopards, and meerkats [30,42,52]. Overall, our data suggest that isotope values in faecal

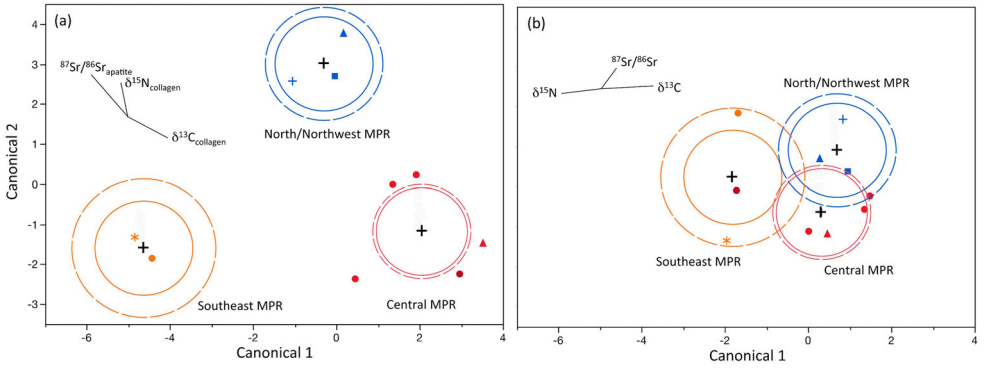

Figure 5. Discriminant function analysis of regions at MPR using (a) bone, or (b) faecal matter. Samples from northwestern and north-central MPR were combined since they have indistinguishable $\delta^{13}C$, $\delta^{15}N$, and very minor apparent differences in $^{87}Sr/^{86}Sr$ (Figure 4). Symbols for individual jaguars correspond with Figure 1. Fifty and 95 % confidence contours are shown for each group are indicated with a star symbol. The relevant contribution of each isotope to each canonical axis is indicated in the upper left-hand corner of each panel. Canonical details and score summaries are provided in Supplementary Tables 1 and 2.

matter can be used to track jaguar landscape use. The combination of $\delta^{13}C_{collagen}$, $\delta^{15}N_{collagen}$, and $^{87}Sr/^{86}Sr_{apatite}$ is highly effective at distinguishing bone samples from different regions of MPR (Figure 5). Carbon, nitrogen, and strontium in faecal matter are less effective at distinguishing regions. Nevertheless, trends are similar to bone, and only one sample was misclassified in the DFA model (Supplementary Table 2).

All scats were found in pine forest (Figure 1). However, those samples that were near large tracts of broadleaf forest (i.e. in southeastern MPR; Figure 1) had some of the lowest faecal and bone $\delta^{13}C$ and $\delta^{15}N$ values (Table 2; Figure 4). These results are consistent with jaguars foraging along the southeastern border of MPR, or perhaps down into the adjacent Chiquibul National Park (Figure 1). They could also indicate foraging in riparian gallery forests within MPR. Strontium isotope data appear to reflect geology, as expected. They are quite variable, but broadly consistent with what we might expect for animals foraging on Palaeozoic granite and metasediments [reviewed in 63–66]. Strontium is most variable in central MPR, but this is not surprising considering that multiple geologies are exposed in this region of the protected area (Figure 1).

Those few examples where isotope values in bone and faecal matter show differing trends (e.g. $\delta^{13}C$ for scats #28, 31 and 36, $\delta^{15}N$ for scats #24 and 25, $^{87}Sr/^{86}Sr$ for sample #24) may indicate movement across different habitats or geologies between meals. All of these samples were recovered from central MPR (Figure 1). Jaguars tend to defecate along the edges of their individual home ranges to advertise their presence and mark their territories [e.g. 67]. Based on camera trap grids and non-invasive genetic sampling, we know that several males co-occurred in central MPR at the time of this study [10,12,68], and isotope data corroborate these findings. However, it is not possible to deduce much about the landscape use of individual jaguars based on such a small sample size. Reported home ranges for male jaguars vary considerably. Within Belize, they range from 28–48 to 150–250 km^2 [19,67]. This most likely reflects abundance and distribution of prey, vegetation structure, access to water, and the distribution of

females' home ranges [e.g. 19,23]. Average daily movement is typically on the order of 2.5 km (reviewed in [23]) but daily movements >18 km have been observed for individuals in Brazil [19]. Regular movement of individuals on the order of 2–10 km within MPR would be consistent with this previous work. An expanded study with a larger sample size will enable investigation of spatial partitioning among individuals more fully.

Isotopic offsets between consumed prey and jaguar faecal matter (Supplementary Table 3; Figure 3) were broadly consistent with previously reported diet–faeces offsets for other mammals (Table 1), but differed somewhat from those previously reported for specific carnivore species. Average $\Delta^{13}C_{collagen-faeces}$ for jaguar scats (4.0 ± 3.3 ‰) was slightly larger than previously estimated $\Delta^{13}C_{diet-faeces}$ for wild coyotes, considerably larger than $\Delta^{13}C_{diet-faeces}$ estimates for captive meerkats, and the reverse direction of $\Delta^{13}C_{diet-faeces}$ estimates for captive snow leopards and tigers (Table 1). These offsets were also more variable than those estimated for coyotes and meerkats, but comparably variable to those recorded for snow leopards and tigers. Average $\Delta^{13}C_{apatite-faeces}$ for jaguar scats (8.0 ± 2.7 ‰) was larger than $\Delta^{13}C_{collagen-faeces}$, but similarly variable. Average $\Delta^{15}N_{collagen-faeces}$ (-2.8 ± 2.5 ‰) was comparable to estimates for wild coyotes and captive snow leopards, but larger than that reported for tigers or meerkats (Table 1). These values were more variable than those reported for coyotes, meerkats, and snow leopards, but variance was similar to that recorded for tigers. Lastly, $\Delta^{87}Sr/^{86}Sr_{apatite-faeces}$ (0.00216 ± 0.00673) was larger and more variable than the $\Delta^{87}Sr/^{86}Sr_{diet-faeces}$ previously reported for groups of captive pigs fed differing diets (−0.000004 to 0.000051 [44]).

There are several potential explanations for the differences in our results and those previously reported results for mammalian omnivores and carnivores. These include the nutritional composition and digestibility of diet, isotopic differences between digested flesh and undigested prey remains in faecal matter, isotopic variability among meals, study design, and sample degradation. First, differences in the digestibility of premixed cat feed and whole prey could readily explain at least some of the differences we observed between wild jaguars and captive cats [e.g. 41,69]. Diet itself appears to influence the isotopic offset between diet and faeces (Table 1). Data obtained for a particular species may not be applicable to that same species if fed a different diet. For example, remarkably different $\Delta^{13}C_{diet-faeces}$ and $\Delta^{15}N_{diet-faeces}$ values have been obtained for chimpanzees (*Pan troglodytes*) in captive versus wild settings [31,51; Table 1]. Likewise, very different values were obtained for wild red-backed voles (*Myodes gapperi*) that were fed a controlled diet for seven days [43] versus 62 ± 12 days [50].

Second, because the isotopic composition of bone collagen and muscle tissue from consumed prey may differ slightly (<2.5 ‰ for carbon; <1 ‰ for nitrogen; reviewed in [62]), our estimate of dietary protein based on bone collagen may differ from the animal flesh that jaguars actually ingested and assimilated. This could help explain why collagen–faeces offsets (particularly $\Delta^{13}C_{collagen-faeces}$) for jaguars tend to be larger than previously reported diet–faeces offsets for carnivores and omnivores, while estimates for nitrogen are more comparable (Table 1).

Third, it is not unusual to find remains from more than one prey species in a given faecal sample (C. Wultsch personal observation). We do not yet have information about prey species represented in the faecal samples from MPR, but analysis is currently underway. If faecal matter reflects more than one prey species, this could bias our results. Certainly,

it would be challenging to use average calculated offsets to estimate dietary intake (for example, we would caution against using these values to compare diets among individuals). However, this challenge is not unusual to our study. Indeed, the ranges in $\delta^{13}C_{faeces}$ (−25.1 to −19.9 ‰) and $\delta^{15}N_{faeces}$ (7.2 to 13.4 ‰) for jaguar scats were similar to those previously reported for captive snow leopards (−24.9 to −18.9 ‰) and tigers (8.2 to 14.3 ‰) that were given a commercial feed [42]. The potential for multiple prey items to contribute to faecal matter is probably not of appreciable concern if we are only interested in landscape use. With few exceptions, isotope data for jaguar faecal matter and bone from consumed prey showed similar trends among regions at MPR, and these trends were broadly consistent with the vegetation and geology where scats were collected (Table 2).

Fourth, study design may also explain some of the observed differences. For example, it is possible that differences in treatment affected faecal $\delta^{13}C$ or $\delta^{15}N$ values. We did not chemically treat faecal matter in our study. Other researchers have also used dried, homogenized faecal matter to calculate diet–faeces offsets for captive tigers, snow leopards and meerkats [42,52]. Conversely, Reid and Koch [30] cleaned faecal matter from coyotes with ultrapure water and dilute hydrochloric acid to remove potential carbonate contaminants. They verified that this did not affect $\delta^{15}N$ values, but it may have had an impact on $\delta^{13}C$ values. This could help explain why average $\Delta^{13}C_{collagen–faeces}$ for jaguar scats was slightly larger than estimated $\Delta^{13}C_{diet–faeces}$ for coyotes, but it cannot explain the differences between our data and estimates for meerkats, snow leopards, or tigers (Table 1). Instead, as mentioned above, calculated $\Delta^{13}C_{diet–faeces}$ and $\Delta^{15}N_{diet–faeces}$ values for captive snow leopards, tigers, and meerkats may be problematic given that the authors were not able to disentangle available diet from diet consumed by each individual prior to defecation. Diet–faeces offsets for coyotes may also be somewhat uncertain considering they are based on a combination of (1) $\Delta^{13}C_{hair–faeces}$ and $\Delta^{15}N_{hair–faeces}$ for just four wild coyotes, and (2) published $\Delta^{13}C_{hair–diet}$ and $\Delta^{15}N_{hair–diet}$ for captive foxes and wolves [38,55].

Sample degradation is a less likely explanation for our results (Figure 2). Based on the sample collection schedule, all scats included in this study should have been less than two weeks old. Atom %C:N for the jaguar scats (4.2 to 7.3 %) was well within the ranges previously reported for captive snow leopards (3.0 to 14.2 %), tigers (4.9 to 9.8 %), and meerkats (4.5 to 11.2 %) [42,52] and there were no isotopic trends with scat age (Figure 2). Collagen yield and atom %C:N for bones (3.3 to 3.5 %) were also consistent with unaltered collagen [70] and there was nothing unusual about bone apatite samples; all yielded similar amounts of carbonate during analysis (J. Curtis personal communication). However, the $\delta^{13}C$ values for collagen and apatite in bone from consumed prey showed somewhat differing trends, which is unexpected. Although these two portions of bone reflect different dietary components, their isotope values should co-vary [56]. Plotting $\delta^{13}C_{apatite}$ versus $\delta^{13}C_{collagen}$ (Figure 6), we observed a linear relationship between $\delta^{13}C_{apatite}$ and $\delta^{13}C_{collagen}$ values for all scats ($R^2 = 0.36$, $F_{1,9} = 4.42$, $P = 0.069$). However, bones from three scats (#25, #31, and #36), which were all produced by Jaguar 1 were somewhat unusual. Excluding these three samples, the relationship between $\delta^{13}C_{apatite}$ versus $\delta^{13}C_{collagen}$ improved dramatically ($R^2 = 0.98$, $F_{1,6} = 309.84$, $P < 0.001$; Figure 6). The offset between $\delta^{13}C_{apatite}$ and $\delta^{13}C_{collagen}$ ($\Delta^{13}C_{apatite–collagen}$) for these three bones was also rather unusual: bones in scats #25 and #36 (both recovered from central MPR) had either very small or negative $\Delta^{13}C_{apatite–collagen}$ while offset for sample #31 (recovered from southeastern MPR) was the largest in our dataset (10.6 ‰; Supplementary Table 3).

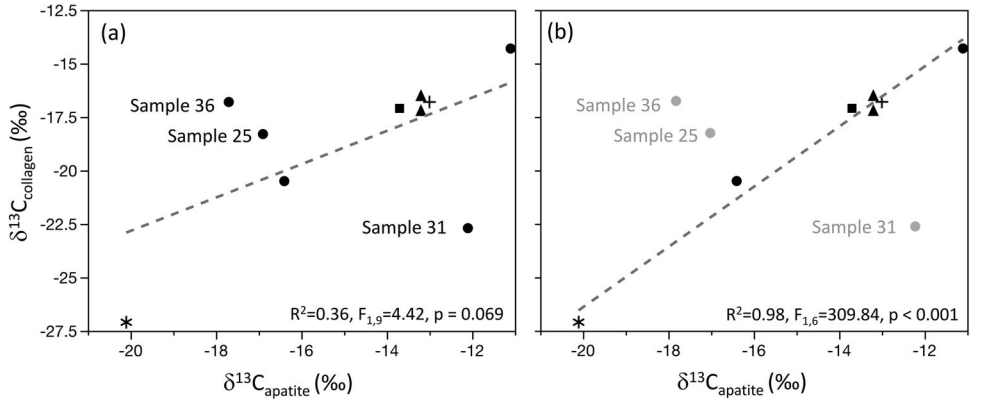

Figure 6. Bivariate plots showing the relationship between $\delta^{13}C_{apatite}$ and $\delta^{13}C_{collagen}$ values for bone from each scat. Samples with unusual $\Delta^{13}C_{apatite-collagen}$ are labelled. Excluding sample 31 considerably improves the relationship (b).

It is possible that passage through a jaguar's digestive tract had some influence on $\delta^{13}C_{apatite}$ or $\delta^{13}C_{collagen}$ values. There is a dearth of information about this topic in the literature. If digestion altered the isotopic composition of bone, then we would expect all samples to have been similarly influenced by this process. We might also expect unusual $\delta^{15}N$ values for affected samples, since this isotope is typically more susceptible to alteration [70]. Plotting $\delta^{15}N_{collagen}$ against $\delta^{13}C_{apatite}$ or $\delta^{13}C_{collagen}$, we observed positive trends (Figure 7), which is in line with expectations for prey feeding at different trophic levels or inhabiting different microenvironments at MPR. There was nothing particularly unusual about scats #25 or #36. This suggests that isotopic alteration during digestion is an unlikely explanation for the small $\Delta^{13}C_{apatite-collagen}$ values. Conversely, scat #31, had much higher $\delta^{13}C_{apatite}$ and lower $\delta^{13}C_{collagen}$ than expected based on its $\delta^{15}N_{collagen}$ values (Figure 7). Something may, indeed, be odd about this individual bone; yet atom %C:N for this sample was normal and there was nothing unusual about this sample during either collagen or apatite analysis. An investigation of the influence of digestion on the isotopic composition of bone is warranted but beyond the scope of the present study.

If alteration during digestion was not responsible for the particularly small or large $\Delta^{13}C_{apatite-collagen}$ we observed for a few specimens, then we may be observing the influence of taxon on $\Delta^{13}C_{apatite-collagen}$ for consumed prey. Typical $\Delta^{13}C_{apatite-collagen}$ are ca. 2 to 10 ‰ depending on an animal's diet [56,57,71,72]. Faunivores tend to have smaller $\Delta^{13}C_{apatite-collagen}$ while herbivores tend to have larger $\Delta^{13}C_{apatite-collagen}$, largely due to increased microbial fermentation of food in the gut [57]. However, small (<2 ‰) to negative $\Delta^{13}C_{apatite-collagen}$ has previously been reported for rodents and humans whose dietary protein and energy respectively came from C_3- and C_4-derived foods [e.g. 56,73,74]. Large (>10 ‰) $\Delta^{13}C_{apatite-collagen}$ has been reported for large-bodied ruminants and now-extinct ground sloths [56,57,72]. We speculate that the bones that produced the smallest $\Delta^{13}C_{apatite-collagen}$ values in scats #25 and #36 belonged to a trophic omnivore, such as an agouti or peccary, which conceivably could have eaten a mixture of C_3 and C_4 foods in the grassy understorey of MPR's pine forest. The bone in scat #31 could have been from a tapir that relied to some degree on microbial fermentation of foliage

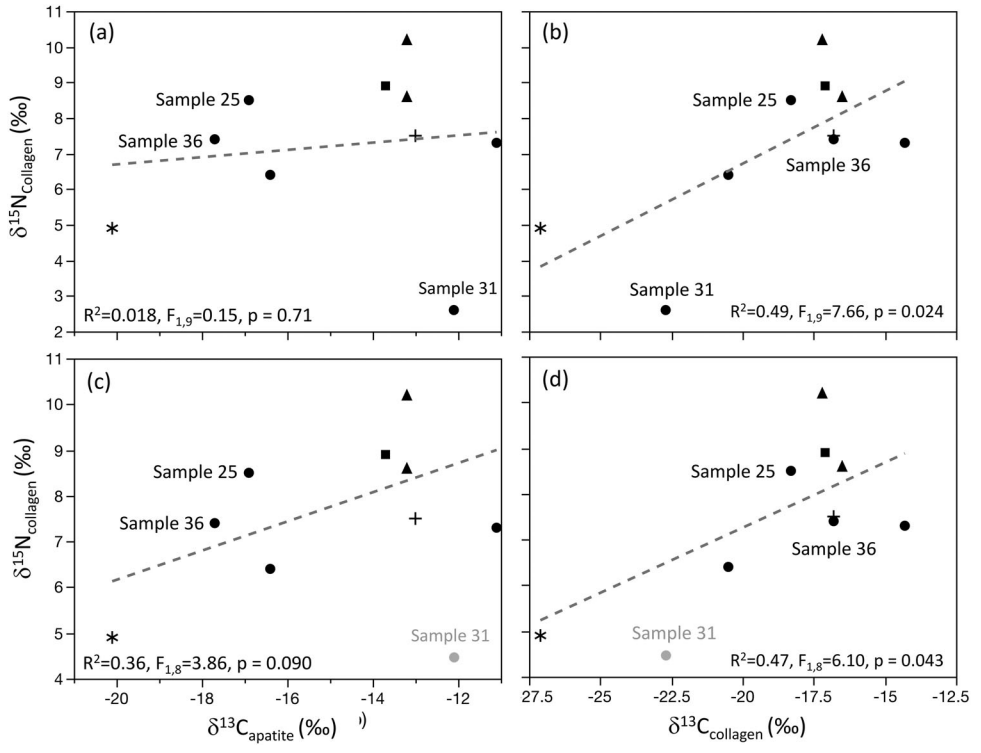

Figure 7. Bivariate plots showing the relationship between $\delta^{15}N_{collagen}$ and $\delta^{13}C_{apatite}$ (a and c) or $\delta^{15}N_{collagen}$ and $\delta^{13}C_{collagen}$ (b and d) for bone from each scat. Samples with unusual $\Delta^{13}C_{apatite-collagen}$ are labelled. Excluding sample 31 improves the relationship between $\delta_{15}N_{collagen}$ and $\delta^{13}C_{apatite}$ (c) but not $\delta^{15}N_{collagen}$ and $\delta^{13}C_{collagen}$ (d).

in its gut (elevated $\delta^{13}C_{apatite}$). Genetic analysis of consumed prey in faeces will provide additional context for interpreting these data.

In conclusion, the isotopic composition of jaguar faecal matter may not be a sound indicator of diet in C_3-dominated natural ecosystems. However, carbon, nitrogen, and strontium isotopes in jaguar faecal matter do reflect foraging in different habitats and on different geologies. This result is reassuring as prey bones are not always present in carnivore faecal samples. It is challenging to obtain landscape use data for solitary, cryptic carnivores like jaguars. Isotopic analysis of faecal matter extends the information that can be gathered from a single faecal sample and expands the existing array of tools that allow us to non-invasively track habitat use and potentially movement of individuals. We envision that a multidisciplinary approach combining genetics and isotopes will be particularly useful for monitoring use of forest fragments and human-altered landscapes (e.g. use of agricultural areas and wildlife movement corridors). Ultimately, the application of multi-disciplinary, non-invasive monitoring techniques has great potential to further improve conservation and management of jaguars and other elusive and threatened species worldwide.

Geolocation information

Mountain Pine Ridge Forest Reserve is located in central Belize (16 ° 57′ 54″ N, 88 ° 54′ 40″ W).

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Disclosure statement

No potential conflict of interest was reported by the authors.

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Data availability statement

All new data associated with this paper are provided in Tables 2 and Supplementary Table 3.

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