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# BULLETIN

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## **METHODS OF ASSESSING HEALTH AND DIET OF FLORIDA PANTHERS (*Puma concolor*) USING MUSEUM SPECIMENS**

### **PART I: Osteology as a Means of Assessing Florida Panther Health**

Laurie Wilkins, Julie M. Allen, Joan Coltrain, Shelly Flanagin, Terry D. Allen,  
& David L. Reed<sup>1</sup>

### **PART II. Stable Isotope Geochemistry: A Method to Evaluate the Diet of Florida Panthers (*Puma concolor*) Using Museum Specimens**

Julie M. Allen, Joan Coltrain, Laurie Wilkins, Shelly Flanagin, & David L. Reed

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## PART II: STABLE ISOTOPE GEOCHEMISTRY: A METHOD TO EVALUATE THE DIET OF FLORIDA PANTHER

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### ABSTRACT

Many of the methods used to determine the diet of large carnivores are costly, time-consuming and may not give a complete picture of diet. Here we discuss the potential for stable-isotope analysis to provide a more complete picture of long-term intake with a pilot study of Florida panthers. We use 20 Florida panthers (*Puma concolor*) from the Florida Museum of Natural History. Our results are consistent with a diet based primarily on deer and hog as other studies have suggested. However, male and female isotope values differ significantly suggesting distinctly different dietary intake. In addition, contrary to findings from studies employing other techniques, no differences are apparent in the isotope signatures of animals from northern versus southern areas. Although the isotope data presented here are preliminary, stable isotope analysis appears to be a useful tool for assessing differences in diet across demographic parameters and will be useful in dietary studies contributing to conservation efforts.

### INTRODUCTION

Determining the diet of an animal is costly and time-consuming, particularly with large carnivores that are difficult to track. Although many of the tools used to determine diet provide useful information, most cannot provide a complete picture of long-term dietary patterning. For example, the analysis of scat is biased toward indigestible dietary components. Furthermore, scat analysis examines the most recent meal but does not reveal seasonal or long-term dietary trends unless collection occurs over a long period of time. Another common method, kill-site analysis, requires that the home range of an animal be accurately determined then searched until a carcass is located. This method also tracks short-term dietary intake and is biased towards the carcasses of larger prey taxa that are easier to locate. Finally, observational studies require significant time depth, particularly when studying large nocturnal carnivores, since predation events are difficult to observe. Alternatively, stable isotope analysis does not require a multi-year study, but does provide a long-term dietary signal and can be used with other methods to give a more complete picture of carnivore diets. Here we present stable isotope analysis of museum speci-

mens as a potential source for information that can aid in management strategies of wild carnivores.

Stemming from the adage, 'you are what you eat,' the stable isotopes of carbon and nitrogen are incorporated into animal tissues in predictable ways representative of the isotopic signature of prey taxa (Hobson & Schwarcz 1986; Kelly 2000). Because tissues are created at different rates, an animal's isotope chemistry can track both long- and short-term dietary patterning. For example, hair grows much faster than bone, so examining the isotopic signature of a hair shaft will give a more recent signature of diet than bone collagen, which turns over very slowly providing an average of long-term intake. Thus stable carbon and nitrogen isotope values are commonly used to reconstruct the diets of large carnivores, such as seals and wolves, and other animals (Hobson et al. 1996; Szepanski et al. 1999; Urton & Hobson 2005, Kelly 2000). These values represent the ratio of the heavy to light isotopes for carbon and nitrogen ( $^{13}\text{C}/^{12}\text{C}$ ,  $^{15}\text{N}/^{14}\text{N}$ ) written in delta notation ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) as parts per million (‰) difference from an internationally recognized standard (PDB for carbon and atmospheric air for nitrogen). The standard is assigned by definition a value of 0‰ and computed as

follows:

Equation 1:

$$\delta^{13}\text{C} \text{ or } \delta^{15}\text{N} = R_{\text{sample}} - R_{\text{standard}} \times 1000 \text{ ‰}$$

where  $R = {}^{13}\text{C}/{}^{12}\text{C}$  or  ${}^{15}\text{N}/{}^{14}\text{N}$

#### BACKGROUND ON CARBON ( $\delta^{13}\text{C}\text{‰}$ )

Stable carbon isotope ratios in bone collagen monitor the ratio of carbon-13 to carbon-12 in the amino acid sequences that make up collagen fibrils, and are representative of diet for the following reasons. When  $\text{CO}_2$  is taken up during photosynthesis, metabolic processes alter or fractionate the ratio of  ${}^{13}\text{C}/{}^{12}\text{C}$ , depleting plant tissues in  ${}^{13}\text{C}$  relative to atmosphere ( $-7.7\text{‰}$ ). The degree of fractionation associated with photosynthesis covaries with the kinetic properties of carbon uptake and enzymatic processes of carbon fixation (Farquhar et al. 1989). In terrestrial plants, two photosynthetic pathways, the  $\text{C}_3$  or  $\text{C}_4$  pathway, have distinct carbon isotope signatures. Fractionation is contingent upon the photosynthetic pathway the plant uses to metabolize atmospheric  $\text{CO}_2$ . Cool season grasses, trees, tubers and most bushy plants utilize the  $\text{C}_3$  photosynthetic pathway which discriminates heavily against  ${}^{13}\text{C}$ , expressing a mean  $\delta^{13}\text{C}$  value of  $-26.7 \pm 2.7\text{‰}$  ( $n=370$ ) (Cerling et al. 1998). A small set of forbs and all warm-season grasses such as corn (*Zea mays*), common to regions where daytime growing-season temperature exceeds  $22^\circ\text{C}$  and precipitation exceeds 25 mm (Ehleringer et al. 1997), use  $\text{C}_4$  photosynthesis resulting in less discrimination against  ${}^{13}\text{C}$  and an average  $\delta^{13}\text{C}$  value of  $-12.5 \pm 1.1\text{‰}$  ( $n=455$ ) (Cerling et al. 1998).  $\delta^{13}\text{C}$  values are passed from producer to consumer leaving a diagnostic signature in both hard and soft tissues that does not covary with the skeletal element analyzed or sex of the sample independent of differences in feeding ecology (Hobson & Schwarcz 1986; Lovell et al. 1986). Fractionation between plants and herbivores is ca.  $5\text{‰}$  and isotopic enrichment between herbivores and each successive trophic level is about  $1\text{‰}$ .

Stable carbon isotope analysis is also used to estimate reliance on marine resources in diets lacking a  $\text{C}_4$  component. Kinetic processes governing bicarbonate ( $\text{HCO}_3^-$ ) formation in seawater fractionate marine bicarbonates approximately  $7\text{‰}$  relative to atmosphere, placing seawater  $\delta^{13}\text{C}$  values near  $0\text{‰}$  (Chisholm et al. 1982; Tauber 1981). Because submerged marine plants employ a  $\text{C}_3$  photosynthetic pathway and derive carbon primarily from seawater bicarbonates, they express mean

$\delta^{13}\text{C}$  values of  $-16$  to  $-18\text{‰}$ , approximately  $7\text{‰}$  more positive than terrestrial  $\text{C}_3$  plants giving them a distinctive marine label.

#### BACKGROUND ON NITROGEN ( $\delta^{15}\text{N}\text{‰}$ )

Nitrogen isotopes are also incorporated into the food web in predictable ways. Most terrestrial plant taxa obtain nitrogen from soil ammonium ( $\text{NH}_4^+$ ) or nitrate ( $\text{NO}_3^-$ ) and have mean  $\delta^{15}\text{N}$  values of  $3\text{--}6\text{‰}$  with a  $0\text{--}9\text{‰}$  range contingent upon temperature and aridity (Pate 1994). Independent of effective moisture, legumes and some epiphytes have symbionts that fix atmospheric nitrogen and accordingly express mean  $\delta^{15}\text{N}$  values of  $1\text{‰}$ , with a ca.  $-2$  to  $2\text{‰}$  range (Evans & Ehleringer 1994; Pate 1994). Herbivores in temperate climates typically exhibit  $\delta^{15}\text{N}$  values of  $6\text{--}9\text{‰}$  while arid-land species and non-obligate drinkers, those that recycle urea, reflect their water-conservation strategies in more positive  $\delta^{15}\text{N}$  values (Ambrose 1991).

Stable nitrogen isotope analysis follows from the understanding that  ${}^{15}\text{N}/{}^{14}\text{N}$  increases by approximately  $2\text{--}4\text{‰}$  with each increase in trophic level due primarily to fractionation during urea production, enriching the isotope signature of nitrogen available for protein synthesis (Schoeller 1999). For example, a carnivore eating a diet consisting mostly of herbivorous prey such as deer will exhibit systematically lower nitrogen isotope ratios than a carnivore in the same ecosystem subsisting primarily on omnivorous animals such as pigs and raccoons.

### METHODS

#### STUDY SYSTEM

The Florida panther (*Puma concolor*) is suitable for dietary studies using stable isotope analysis for several reasons. First, the Florida Museum of Natural History houses a large collection of panther, over 140 specimens, providing an excellent resource for bone tissue from animals representing various geographic ranges and temporal periods. In addition, Florida panther diet has been studied using scat and kill-site analysis providing a comparative database. Finally, Florida panthers are endangered, and an understanding of their predation strategies is central to successful management of a declining population.

Maehr et al. (1990) demonstrated using scat and kill site analysis that the main prey of panthers inhabiting the public lands of southwest Florida, including Florida Panther National Wildlife Refuge (FPNWR), Big Cypress National Park (BCNP), Big Cypress Seminole Indian Refuge (BCSIR), and Fakahatchee Strand State Preserve (FSSP), was wild hog (*Sus scrofa*) and white-

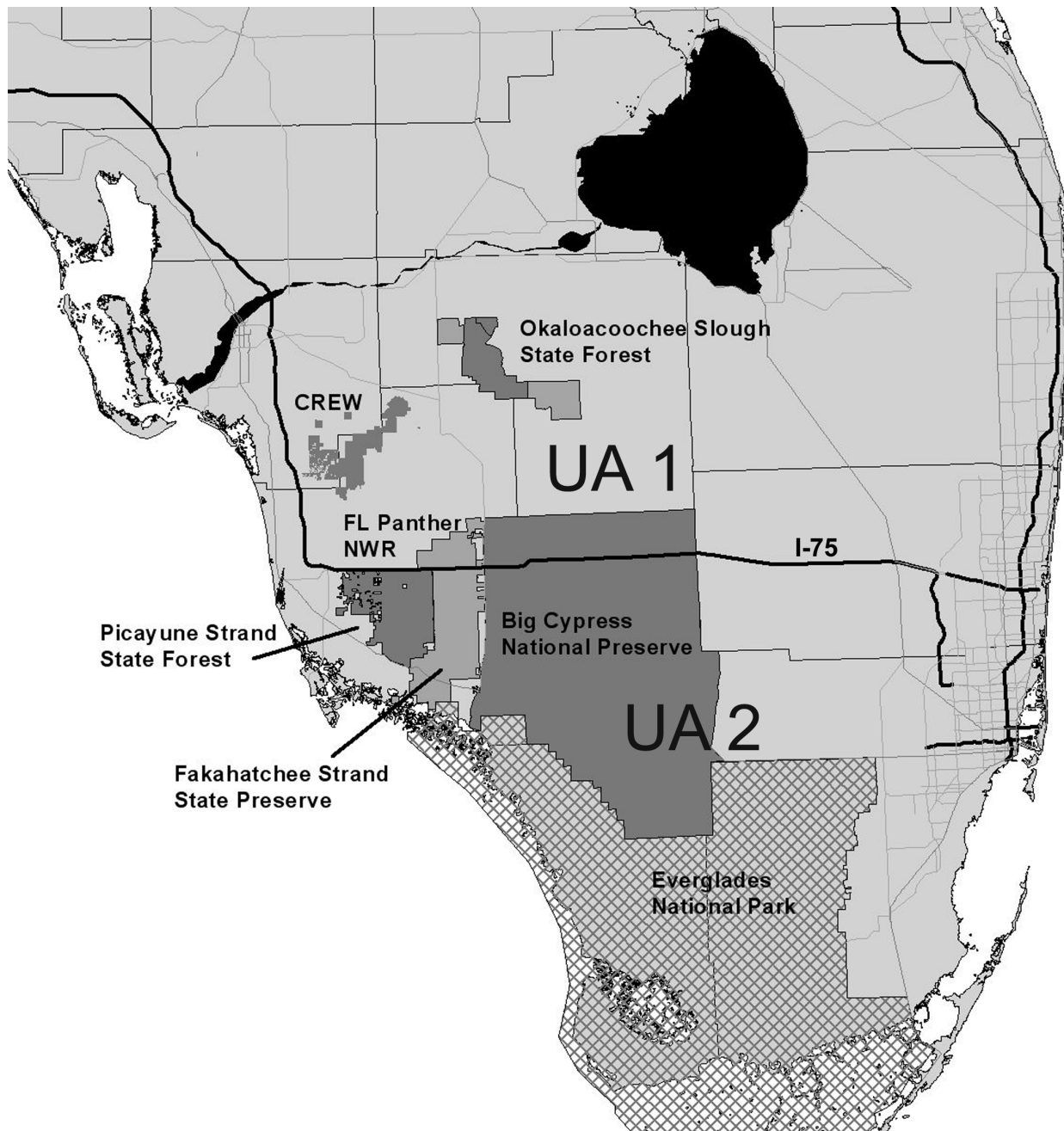


Figure 1: Map of Southern Florida showing the range of the Florida panther and the northern and southern major use areas. The northern use area includes Florida Panther National Wildlife Refuge (FPNWR), Big Cypress Seminole Indian Refuge (BCSIR) not shown, and northern Big Cypress National Park (nBCNP). The southern use area includes southern Big Cypress National Park (sBCNP), Fakahatchee Strand State Preserve (FSSP) and Everglades National Park (ENP). Northern and southern BCNP are separated by I-75. Map was provided by D. Land from FWC.

tailed deer (*Odocoileus virginianus*), followed by raccoon (*Procyon lotor*), armadillo (*Dasypus novemcinctus*), and rabbit (*Sylvilagus spp.*) in order of their abundance in the diet. However, frequencies of both large and small prey varied considerably between the northern and southern portions of BCNP (nBCNP and sBCNP; Figure 1), which crosses a soil boundary.

The soils of nBCNP are thought to be better drained and more fertile (Leighty et al. 1954). Here, hogs dominated panther diet making up 58.7% of the biomass consumed, whereas in sBCNP hogs made up merely 22.7% (Maehr et al. 1990, Table 1). In contrast, smaller species (raccoon, armadillo, and rabbit) were present in higher frequencies in the diets of panthers from sBCNP

Table 1: Percent biomass of prey animals consumed by Florida panthers. Values were taken from Maehr et al. (1990), and Dalrymple and Bass (1996) calculated from scat and kill-site analysis.

	Percent Biomass consumed				Study
	Hog	Deer	Small Mammals	Other	
North Big Cypress National Park	58.7	27.0	9.4	4.9	Maehr et al., 1990
South Big Cypress National Park	22.7	43.4	31.8	2.0	Maehr et al., 1990
Everglades National Park	0.7	78.4	18.1	2.4	Dalrymple and Bass 1996

(Maehr et al. 1990). Shortemeyer (1994) reported that south of I-75 deer and hog densities rarely exceed one animal per 100 acres. In Everglades National Park (ENP), deer represented 78.4% of biomass consumed, in part because no hogs are present, (Dalrymple and Bass 1996) but a 67% decrease in deer densities since 1959 has been reported (Fleming et al. 1994) suggesting that deer are more difficult to hunt in ENP. Consequently, the biomass of smaller prey in ENP was 18.2% higher than that of nBCNP (Table 1).

Veterinary studies based on physical condition, body weight, reproductive status, and hematologic and serum values conducted between 1986 and 1990 identified a north/south "health cline." Panthers utilizing land north of State Road 84, including private ranches, the FPNWR, and the Bear Island unit of the BCNP, were in better condition than those in the ENP or FSSP (Roelke 1990). The panthers in sBCNP were also found to be in poorer physical condition, have larger home ranges and lower reproductive output than panthers in the north (Maher et al., 1989a; Maher et al. 1990). McCown (1991) found that panthers in the cypress-dominated southern portion of sBCNP were smaller and less productive than in the Bear Island unit located in nBCNP. Differences in panther diet leading to a north to south gradient of declining health has been attributed to a shift from reliance on hog in the north to deer and smaller prey in the south (Beier et al. 2003). In the most southern part of ENP panther mean body size is small which is thought to be due to low deer densities. One danger the panthers face when feeding on small more aquatic based prey is the potential for mercury ingestion. (Maehr et al. 1990; Iriate et al. 1990; Roelke 1990; Dalrymple & Bass 1996).

Here we report the bone collagen stable carbon and nitrogen isotopes ratios of 20 Florida panthers and 25 prey individuals. Panthers were chosen across the north/south diet cline to determine if their stable isotope chemistry reflects the apparent dietary differences discussed above. The northern use area included FPNWR, nBCNP and FPSIR; the southern use area was comprised of sBCNP, FSSP and ENP (Figure 1). Only adult

panthers were selected for study, and we chose animals with the greatest amount of natural history information in order to relate isotope values to known home ranges and diets. All of the animals were radio-collared and tracked by FWC.

Stable isotope values were also obtained from bone collagen of the five primary prey taxa of Florida panthers gathered from three sources; the FLMNH mammal collection, actual prey remains in panther scats from ENP, and samples provided by Deborah Jansen from panther scats/kill sites in the BCNP. We sampled 10 white-tailed deer (*Odocoileus virginianus*), 11 raccoon (*Procyon lotor*), three rabbit (*Sylvilagus palustris*), two armadillo (*Dasypus novemcinctus*), and two feral hogs (*Sus scrofa*).

#### LAB PROCEDURE

Approximately 500 mg of cortical bone was cleaned of surface contaminants then demineralized whole in 0.6N HCl at 4° C. Sterile ddH<sub>2</sub>O was used throughout and glassware was sterilized by heating to 550° C for 3h. The resultant collagen was then rinsed to neutrality, treated with 5% KOH to remove organic contaminants, rinsed to neutrality, and lyophilized. Approximately 100 mg of lyophilized collagen was gelatinized in 5 ml of water (pH 3) for 24 hours at 120° C. Water-soluble and -insoluble phases were separated by filtration and the former was lyophilized and weighed to determine the collagen yield.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values were determined by flash combustion to produce CO<sub>2</sub> and N<sub>2</sub> and measured against the appropriate reference gas on a Finnigan Delta Plus mass spectrometer with Carlo Erba EA118 CHN interface. Stable isotope measurements and weight percent C and N values were obtained from single sample combustion. All samples met well-established preservation criteria (Ambrose 1990). Analytical precision is  $\pm 0.1\text{‰}$  for carbon and  $\pm 0.2\text{‰}$  for nitrogen.

#### RESULTS AND DISCUSSION

Stable isotope values for panthers and their prey are reported in Table 2. Mean carbon and nitrogen isotope

values for panthers in the northern and southern use areas are not significantly different ( $\delta^{13}\text{C}\text{‰}$ , t-test,  $t = -1.7$ ,  $df = 11$ ,  $p = 0.10$ ;  $\delta^{15}\text{N}\text{‰}$ , t-test,  $t = -0.97$ ,  $df = 13$ ,  $p = 0.35$ ; Fig. 2A). However, animals in the north have a wider range of  $\delta^{13}\text{C}\text{‰}$  and  $\delta^{15}\text{N}\text{‰}$  values indicative of a more varied diet.

When sorted by sex, male and female panthers differ significantly in carbon (t-test,  $t = -3.7$ ,  $df = 17$ ,  $p = 0.002$ ), and nitrogen isotope values ( $t = -4.4$ ,  $df = 15$ ,  $p = 0.0005$ ; Fig. 2B) indicating likely reliance upon a different suite or combination of prey taxa. Also, the range of male panther isotope values is greater than that of females suggesting a more varied diet in males.

### CARBON ( $\delta^{13}\text{C}\text{‰}$ ) RESULTS

Carbon ( $\delta^{13}\text{C}\text{‰}$ ) isotope signatures increase by 1‰ with each increase in trophic levels above the primary consumer.  $\delta^{13}\text{C}\text{‰}$  values of deer are consistent with a high  $\text{C}_3$  browsing diet and ranged from -24.1‰ to -20.4‰ with an average of  $-21.9 \pm 1.2\text{‰}$ . Feral hog  $\delta^{13}\text{C}$  values are nested within the range for deer making them indistinguishable isotopically, at -23.2‰ and -21.6‰ with an average of  $-22.4 \pm 1.1\text{‰}$ . Raccoon  $\delta^{13}\text{C}\text{‰}$  values range from -23.9‰ to -16.7‰ with an average

of  $-19.9 \pm 2.4\text{‰}$  indicating a wide range of dietary elements including some with a marine component. Rabbit  $\delta^{13}\text{C}\text{‰}$  values are -21.6‰ to -20.4‰, consistent with a diet of  $\text{C}_3$  plant foods. The two armadillos sampled have widely varying  $\delta^{13}\text{C}\text{‰}$  isotope values of -19.9‰ and -15.7‰ suggesting that isotopic diversity is likely high within this species, and that a larger sample of armadillos, as well as raccoon, is needed (Table 2, Fig. 3). Armadillo prey items (invertebrates) may feed on a wide variety of plant material, generating considerable variation in  $\delta^{13}\text{C}\text{‰}$  values.

Male panther  $\delta^{13}\text{C}\text{‰}$  isotope values range from -20.1‰ to -15.6‰ with an average of  $-18.6 \pm 1.5\text{‰}$  and females from -21.0‰ to -19.1‰ with an average of  $-20.3 \pm 0.7\text{‰}$  (Table 2, Fig. 3). Three males from the northern use area are outside the range of deer and hog values suggesting a large portion of their diet included other prey taxa, possibly raccoon and armadillo. Two Florida panthers (FP 15 and FP 16) were tracked by Dalrymple and Bass (1996) and have carbon numbers of -19.6‰ and -19.9‰, respectively. Both animals subsisted primarily on deer and hog but it was noted that FP 16 also killed eight raccoons during the tracking.

Table 2: Stable carbon and nitrogen isotope values for (A) Florida panthers and (B) their prey species. Florida panthers are listed by Florida Museum of Natural History identification number (UFID) as well as a FP number assigned by the Florida Fish and Wildlife Conservation Commission. The prey taxa are listed by the species, county collected, and UFID for animals sampled from the Florida Museum of Natural History mammal collection.

A. Florida Panthers					
UFID	Panther	Sex	$\delta^{13}\text{C}\text{‰}$	$\delta^{15}\text{N}\text{‰}$	Use Area
18798	3	F	-21.0	7.9	S
30935	49	F	-20.7	8.0	N
24267	8	F	-20.9	8.1	S
24563	15	F	-19.6	8.1	S
27148	31	F	-19.1	8.1	N
30960	32	F	-21.0	8.2	N
30393	23	F	-20.2	8.3	S
29621	36	F	-20.2	8.5	S
24931	37	M	-20.1	8.3	N
20777	2	M	-19.9	8.4	S
24315	25	M	-18.6	8.4	N
29263	51	M	-18.4	8.4	S
19096	1	M	-19.4	8.5	S
22529	4	M	-18.3	8.7	S
22409	7	M	-20.1	8.9	S
26939	26	M	-18.5	9.0	N
29821	16	M	-19.9	9.3	S
29819	63	M	-17.7	9.3	N
26844	34	M	-16.4	9.3	N
29262	45	M	-15.6	9.8	N

Table 2 continued:

B. Prey Species			
	County	$\delta^{13}\text{C}\text{‰}$	$\delta^{15}\text{N}\text{‰}$
<i>Odocoileus virginianus</i> (White-tailed Deer)			
	Dade	-20.9	3.7
UFID 24907	Collier	-24.1	4.2
	Dade	-20.4	4.8
UFID 9512	Collier	-22.2	4.9
UFID 22583	Collier	-22.7	5.2
UFID 22582	Collier	-23.1	5.8
UFID 9508	Collier	-21.0	6.2
	Collier	-20.4	6.2
	Dade	-21.9	6.4
	Collier	-22.0	8.8
<b>Average</b>		<b>-21.9</b>	<b>5.6</b>
<i>Procyon lotor</i> (Raccoon)			
UFID 25949	Dade	-21.7	6.5
	Dade	-23.0	7.2
UFID 22569	Dade	-21.0	7.6
UFID 10192	Collier	-17.8	8.2
	Collier	-20.2	8.5
UFID 24012	Collier	-16.7	8.5
	Collier	-17.7	8.6
	Dade	-19.4	8.8
	Dade	-20.1	9.1
UFID 10089	Collier	-17.0	9.2
UFID 4611	Broward	-23.9	10.7
<b>Average</b>		<b>-19.9</b>	<b>8.4</b>
<i>Sus scrofa</i> (Feral Hog)			
	Dade	-21.6	4.8
	Dade	-23.2	6.5
<b>Average</b>		<b>-22.4</b>	<b>5.7</b>
<i>Sylvilagus palustris</i> (Rabbit)			
	Dade	-20.8	1.2
	Dade	-20.4	1.8
	Dade	-21.6	2.9
<b>Average</b>		<b>-20.9</b>	<b>2.0</b>
<i>Dasypus novemcinctus</i> (Armadillo)			
	Dade	-19.9	7.1
	Collier	-15.7	7.2
<b>Average</b>		<b>-17.8</b>	<b>7.15</b>



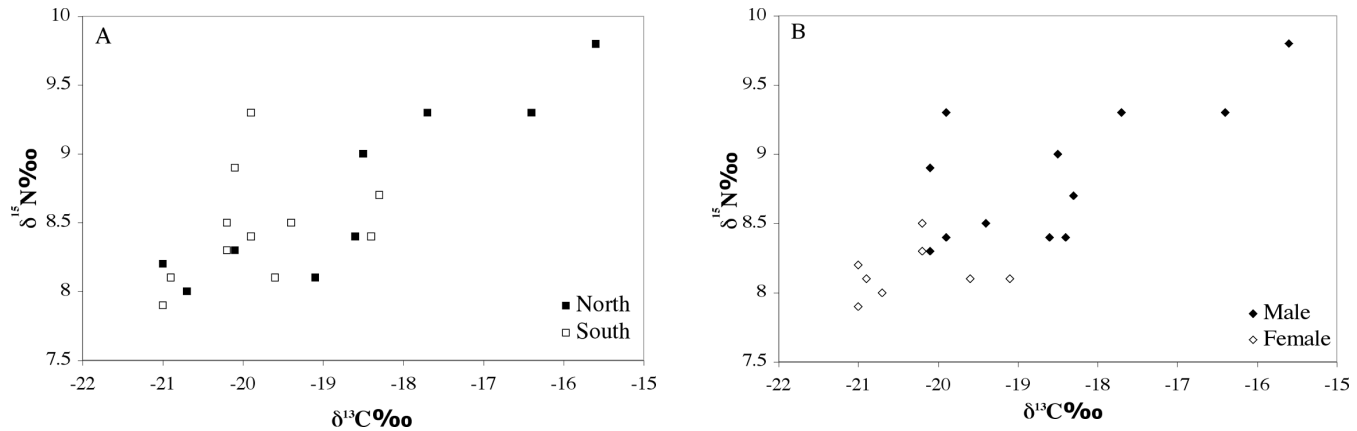


Figure 2: Stable carbon ( $\delta^{13}\text{C}\text{‰}$ ) and nitrogen ( $\delta^{15}\text{N}\text{‰}$ ) isotope ratios for 20 Florida panthers from the Florida Museum of Natural History. There was no significant difference between animals in the north ( $n = 9$ ) and southern ( $n = 11$ ) use areas (A). A significant difference was found between male ( $n = 12$ ) and female ( $n = 8$ ) panthers (B).

### NITROGEN ( $\delta^{15}\text{N}\text{‰}$ ) RESULTS

When nitrogen isotope signatures ( $\delta^{15}\text{N}\text{‰}$ ) are plotted (Fig. 3), panthers have higher  $\delta^{15}\text{N}\text{‰}$  signatures than most of their prey taxa as expected. Nitrogen isotope signatures increase 2-4‰ with each step up the food web, and values for animals at each trophic level met this expectation. Rabbits from the Everglades have the lowest nitrogen values, with a range of 1.2‰ – 2.9‰,  $n=3$ , perhaps due to the use of agricultural fertilizers (SWFMD 1992). Fertilizers are depleted in  $^{15}\text{N}$  with  $\delta^{15}\text{N}\text{‰}$  values ranging from -0.5 to 0.3‰ (Schwertl et al. 2005). It is also possible, that vegetation in the diets of the sampled rabbits was depleted by high levels of nitrogen-fixing bacteria in agricultural runoff (Craft and Richardson 1998). If so, further sampling of rabbits may show distinct differences between rabbit isotope values in various micro-environmental zones within southern Florida.

Deer  $\delta^{15}\text{N}\text{‰}$  isotope signatures are highly vari-

able ranging from 3.7‰ – 8.8‰ with an average of  $5.6 \pm 1.4\text{‰}$  ( $n=10$ ); the lowest value (3.7‰) is from a Dade County specimen. Feral hog  $\delta^{15}\text{N}\text{‰}$  values again fell within the range of deer at 4.8‰ and 6.5‰ with an average of  $5.7 \pm 1.2\text{‰}$  ( $n=2$ ) making their influence on panther  $\delta^{15}\text{N}\text{‰}$  indistinguishable isotopically from that of deer. Armadillo  $\delta^{15}\text{N}\text{‰}$  values are 7.1‰ and 7.2‰ ( $n=2$ ) slightly higher than deer (5.6‰) as expected given their insectivorous diet (Taber 1945). Raccoon  $\delta^{15}\text{N}\text{‰}$  values range from 6.5‰ – 10.7‰ with an average of  $8.4 \pm 1.1\text{‰}$  ( $n=11$ ) representing a varied omnivorous diet (Whitney 1931). Enriched raccoon  $\delta^{15}\text{N}\text{‰}$  values suggest the input of marine resources such as sea turtle eggs, crustaceans, bivalves and other marine invertebrates.

Male panther  $\delta^{15}\text{N}\text{‰}$  values range from 8.3‰ – 9.8‰ ( $n=12$ ) with an average of  $8.9 \pm 0.5\text{‰}$  and female panther  $\delta^{15}\text{N}\text{‰}$  values from 7.9‰ – 8.5‰ ( $n=8$ ) with

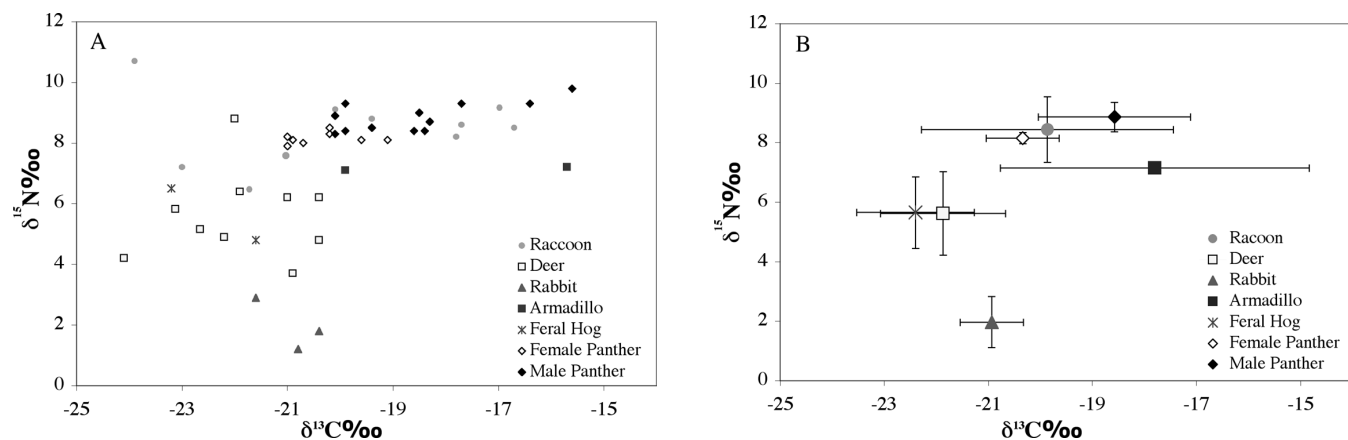


Figure 3: Stable carbon ( $\delta^{13}\text{C}\text{‰}$ ) and nitrogen ( $\delta^{15}\text{N}\text{‰}$ ) isotope ratios for 20 Florida panthers and common prey species. Part A includes all data points and B the averages; bars represent one standard deviation.

an average of  $8.1 \pm 0.2\%$ . These values are consistent with a diet primarily based on deer and hogs, as reported in earlier dietary studies (Maehr et al. 1990; Dalrymple & Bass 1996). Everglades panthers 15 and 16 (UF24563 and UF29821) were tracked by Dalrymple and Bass (1996) who reported that FP 15 ate 14 deer, one marsh rabbit and one opossum during the follows; FP 15 had a nitrogen value of 8.1‰. FP 16 had the most varied diet in the Dalrymple and Bass study, seven deer, five hogs, 11 alligators, eight raccoons, one marsh rabbit, one otter, one bobcat and exhibits a nitrogen isotope signature of 9.3‰. This nitrogen value is indicative of a diet high in high-trophic-level prey taxa, such as alligator, raccoon and bobcat, in keeping with the findings of Dalrymple and Bass (1996). Bone collagen isotope values average dietary intake over several years, perhaps the lifetime of an adult panther, and tracking is likely to capture predation events over a narrower window of time. These results then suggest that FL panther 15 had a diet high in trophic level taxa for a long period of time.

### CONCLUSIONS AND RECOMMENDATIONS

Our stable isotope analyses are consistent with the expectation that deer and feral hog are primary dietary elements. Our results have also revealed interesting differences in male and female panther diets, but further sampling of panthers and prey taxa is required to substantiate sex-based dietary patterning. The most likely prey base for female panthers is deer, hog and rabbits with an emphasis on deer and hog indicated by their nitrogen values. Male panthers had wider ranges of  $\delta^{13}\text{C}\%$  and  $\delta^{15}\text{N}\%$  isotope values indicating a more varied diet. It is possible that the more varied diet in males is related to the fact that males have larger home ranges (Maher et al. 2002; Jansen pers. com.). It is possible that males may wander out of suitable deer or hog habitat in defense of a large home range necessitating the addition of other prey species. Also, the simple act of moving across large geographic distances may cause male panthers to come into contact with a more diverse assemblage of prey species. Alternatively, it is possible that defending large home ranges makes it inefficient to remain at kill-sites of very large prey (deer or hog) as long as females. The pronounced differences in male and female panther isotope values should be investigated further.

In particular, male panthers in the north have carbon values that appear more representative of raccoon and armadillo values. A dietary cline has been reported with panthers in the north feeding primarily on feral hog whereas those in the south feed primarily on deer and small mammals (Beier et al. 2003). We found no differ-

ences isotopically between the diets of northern and southern panthers, and show no significant difference between deer and hog isotope signatures, thus cannot speak to this issue in greater detail. A larger sample of deer and hogs may reveal differences in their isotopic signatures, given the omnivorous diet of feral hogs. Three male panthers in the northern study area appear to have relied heavily on small mammals, a result not previously found in other studies. Two of the three, FP 45 (UF29262) and FP 63 (UF29819), were likely killed by other panthers. These results suggest heavy competition between panthers in the north for prey or mating opportunity. The third panther, FP 34 (UF26844), died of a bacterial infection. All three panthers had Harris lines and other osteopathologies, which are suggestive of overall health problems (Wilkins et al. 2007). It is possible that stable isotope chemistry can assist in identifying sick animals that were not able to hunt efficiently and were thus forced to eat smaller prey. Similarly, stable isotopes may reveal areas in which prey densities are low and panthers are competing heavily for food. Sampling many more animals in each area may help to differentiate between these two issues. Our results, although preliminary, suggest that stable isotopes may reveal important dietary differences among Florida panthers and prove to be a useful management tool.

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### EPILOGUE

Natural history museums are charged with the mission of archiving the world's biodiversity. As threats to the world's biodiversity increase it is becoming apparent that the only place to view or study certain organisms will be within natural history collections. Cooperation between natural history museums and the agencies that monitor threatened wildlife can facilitate the archival of specimens from seriously threatened species. In the case of Florida panthers, the Florida Museum of Natural History has cooperated with the Florida Fish and Wildlife Conservation Commission, The National Park Service, and US Fish and Wildlife Service to recover, process, and archive skeletal, genetic, and other material from every dead Florida panther recovered during the last 20 years. In so doing, the FLMNH now has an impressive collection of over 140 specimens of Florida panther.

Archival material such as that maintained in natural history collections provides a unique opportunity to examine a population over time and link health parameters and dietary shifts to life history parameters such as age, parentage, genetic profile and/or habitat. In the case of small, highly inbred populations like Florida panthers, it is important to note that we cannot foresee all of the health threats to the population without the benefit of having many specimens to examine. Similarly, we cannot foresee all of the tools and technologies that will be available to study these museum specimens. As such, it is important that we maintain a safely archived, permanent repository for Florida panthers and other critically endangered organisms.

In studying the osteology and osteopathology of over 140 individual Florida Panthers, Wilkins et al. (2007, Part I) were able to not only examine the correlates of bone abnormalities that have been well documented, but they also noticed new findings relating to systemic infections that have yet to be studied in detail. Allen et al. (2007, Part II) examined the diet of Florida panthers with stable isotope geochemistry, which has not yet been applied to Florida panthers. This study was able to show significant differences in the diet of male and female panthers, which had never been noted before in the literature. Both studies demonstrate the value of natural history collections to the conservation and maintenance of critically endangered species.