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Diet of the Small Cave Bear *Ursus* (*Spelaearctos*) *rossicus* Borissak, 1930 (Mammalia, Carnivora, Ursidae) As Revealed by ^{13}C and ^{15}N Isotope Analyses in Bone Collagen

P. A. Kosintsev[a,*], K. Yu. Konovalova[a], and G. V. Simonova[b]

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Abstract—The ^{13}C and ^{15}N isotope contents in bone collagen were analyzed using bones of the small cave bear *Ursus* (*Spelaearctos*) *rossicus* Borissak, 1930 from localities in the Middle and Southern Urals. The bones date from the last interglacial (MIS 5) and glacial (MIS 3) periods. The bones were from males and females aged 3, 4, and >4 years. Sexual, geographical, and chronological differences in ^{13}C and ^{15}N contents were studied. Notable gender, geographic, and chronological differences were observed between samples. In the Middle Urals, females led a more predatory lifestyle than males during the interglacial period, and the trophic niches of males and females converged due to an increase in herbivory during the transition to the glacial period. In the Southern Urals, males led a more predatory lifestyle than in the Middle Urals during the interglacial period. The extent of changes in $\delta^{13}C$ and $\delta^{15}N$ values in the Southern Urals during the transition was found to correspond to differences between trophic levels.

Keywords: *Ursus rossicus*, small cave bear, Late Pleistocene, Urals, stable isotope, ^{13}C, ^{15}N, collagen, nutrition

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Collagen contents of the ^{13}C and ^{15}N isotopes in subfossil mammalian bones are broadly used to study the animal diets in the past [1, 2]. This is the main method to reconstruct the diet in the case of extinct species [3]. As for extinct species, one of the largest datasets on ^{13}C and ^{15}N contents in bone collagen has been obtained for the large cave bear *Ursus* (*Spelaearctos*) *spelaeus* s. l. of Western and Central Europe [4, 5]. Data on the ^{13}C and ^{15}N bone collagen contents of the small cave bear *U.* (*S.*) *rossicus* Borissak, 1930 are unavailable for Europe and originate from a single locality in the Urals [6, 7].

Based on morphological data and nuclear DNA analyses, the small cave bear *U.* (*S.*) *rossicus* Borissak, 1930 inhabited the Urals in the Late Pleistocene and greatly differed both morphologically and genetically from the large cave bear *U.* (S.) *spelaeus* s. l. [8, 9].

[a] *Institute of Plant and Animal Ecology, Ural Branch, Russian Academy of Sciences, Yekaterinburg, Russia*
[b] *Institute of Monitoring of Climatic and Ecological Systems, Siberian Branch, Russian Academy of Sciences, Tomsk, Russia*
**e-mail: kpa@ipae.uran.ru*

Small cave bear bones from eight new localities were tested for contents of the ^{13}C and ^{15}N isotopes in bone collagen. The localities of the Middle Urals include the caves Makhnevskaya Ledyanaya (59°26′ N 57°41′ E), Dvoynaya (59°06′ N 57°31′ E), Kizel (59°05′ N 57°36′ E), Viasher (59°05′ N 57°37′ E), Geologov 1 (58°46′ N 57°43′ E), and Dinamitnaya (58°41′ N 57°37′ E). Deposits of the Makhnevskaya Ledyanaya Cave date back to the Mikulin interglacial optimum (marine isotope stage (MIS) 5e, 130–115 thousand years ago) by the fauna composition and palynology data [10]. Deposits of the Dvoynaya Cave have been dated to the Mikulin interglacial (MIS 5, 130–76 thousand years ago) by the fauna composition. The following radiocarbon dates have been obtained for small cave bear bones from the Kizel Cave: >48500 BP, no.?; 46250 ± 700 BP, OxA-19565; 31870 ± 190 BP, OxA-16960; 36 390 ± 270 BP, OxA-16964; 39040 ± 330 BP, OxA-19566; 35330 ± 220 BP, OxA-19561; 35110 ± 230 BP, OxA-19562; 34610 ± 230 BP, OxA-19567; 32940 ± 190 BP, OxA-19564; and 32630 ± 180 BP, OxA–19563 [9, 11], corresponding to MIS 3 (57–29 thousand years ago). Samples from the caves Viasher, Geologov 1, and Dinamitnaya date back to MIS 3 (57–29 thousand years ago) according to the composition of the accompanying fauna.

Table 1. Measurements of $\delta^{13}C$ and $\delta^{15}N$ in skeletal bone collagen in small cave bear *U.* (*S.*) *rossicus* bones from the Urals

Time period	Gender[1]	*n*	$\delta^{13}C$, ‰			$\delta^{15}N$, ‰		
			min	max	M ± sd	min	max	M ± sd
Middle Urals								
MIS 5 130–76 TYA[2]	♂	4	–24.6	–22.6	–23.3 ± 0.89	6.1	7.1	6.6 ± 0.44
	♀	7	–23.9	–22.4	–22.9 ± 0.51	5.8	10.1	7.6 ± 1.37
	♂,♀	11[3]	–24.6	–22.4	–23.1 ± 0.66	5.8	10.1	7.3 ± 1.20
MIS 3 57–29 TYA	♂	8	–23.5	–22.5	–22.9 ± 0.39	1.8	6.3	5.2 ± 1.69
	♀	7	–24.3	–22.4	–23.2 ± 0.63	3.1	8.3	5.4 ± 2.01
	♂,♀	15[3]	–24.3	–22.4	–23.1 ± 0.51	1.8	8.3	5.3 ± 1.78
Southern Urals								
MIS 5 130–76 TYA	♂	4	–23.5	–22.3	–22.8 ± 0.54	5.6	12.5	7,9 ± 3.14
	♀	1	23.1			10.2		
	♂,♀	5[3]	–23.5	–22.3	–22.9 ± 0.48	5.6	12.5	8.4 ± 2.90
MIS 3 57–29 TYA	?	16[4]	–25.6	–19.3	–22.3 ± 1.93	3.2	8.1	5.4 ± 1.34
	?	5[5]	–21.8	–22.8	–21.3 ± 0.46	3.1	8.8	4.9 ± 2.32
	?	21[3]	–25.6	–19.3	–22.1 ± 1.74	3.1	8.8	5.2 ± 1.57

[1]♂, male; ♀, female; ?, gender not determined.
[2]TYA, thousand years ago.
[3]Pooled sample.
[4]Silaev, Parshukova, Gimranov, et al., 2020.
[5]Gimranov, Bocherens, Kavcik–Graumann et al., 2022.

In the Southern Urals, bones from two caves, Barsuchiy Dol (55°09′ N 57°15′ E) and Sikiyaz–Tamak 22 (55°11′ N 58°36′ E) were used to obtain new isotope data. Deposits date to the Mikulin interglacial optimum in the former [12] and the total Mikulin interglacial in the latter according to the fauna composition. Published isotope data were used in the case of the Imanay Cave (53°02′ N, 56°26′ E) [6, 7], where deposits date back to MIS 3 [7]. Bear deaths during winter hibernation led to bone accumulation in all caves [13]; i.e., the remains are of the same taphonomic type. Thus, two, interglacial (MIS 5) and glacial (MIS 3), climatic periods of the Late Pleistocene were characterized with respect to the ^{13}C and ^{15}N isotope contents in small cave bear bone collagen in the Middle and Southern Urals.

The analysis included 4 sculls, 2 ribs, 2 humeri, 1 radius, 1 ulna, 7 femori, and 14 tibiae. The genders and ages of the bears to which the bones belonged were inferred from the bone sizes, epiphyseal fusion, and the state of dentition [14–17]. Both males and females of ages 3+ and 4+ (subadults) or >4+ (adults) years were present in the samples (Table 1). All bears were sexually mature and were subsequently pooled into a single sample. All bones were from different individuals. The gender has not been specified in the data from the Imanay Cave [6, 7].

The carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) isotope compositions in bone collagen were determined by isotope–ratio mass spectrometry, using a DELTA V Advantage instrument (Thermo Fisher Scientific, Germany) with a Flash 2000 element analyzer (the instruments were provided by the Tomsk Regional Collective Use Center (Siberian Branch, Russian Academy of Sciences). A standard protocol was followed. The Vienna Pee Dee Belemnite (VPDB) was used as an international standard reference for carbon isotope measurements. Gaseous atmospheric N_2 was used as an international standard for nitrogen isotope measurements. The laboratory working gases CO_2 and N_2 were calibrated using IAEA-600 caffeine as an international reference substance. The absolute measurement error estimated using three consecutive measurements did not exceed ±0.2‰ for $\delta^{13}C$ and ±0.4‰ for $\delta^{15}N$.

The carbon and nitrogen isotope compositions in small cave bear bone collagen are summarized in Table 1.

The objective of this work was to compare the $\delta^{13}C$ and $\delta^{15}N$ values in bone collagen for small cave bear males and females, the interglacial and glacial periods of the Late Pleistocene in the Middle and Southern Urals.

The $\delta^{13}C$ and $\delta^{15}N$ mean values differed between male and female samples from the Middle Urals by 0.6 and 1.0‰, respectively, in the interglacial period and 0.3 and 0.2‰, respectively, in the glacial period (Table 1). Differences in $\delta^{13}C$ and $\delta^{15}N$ means

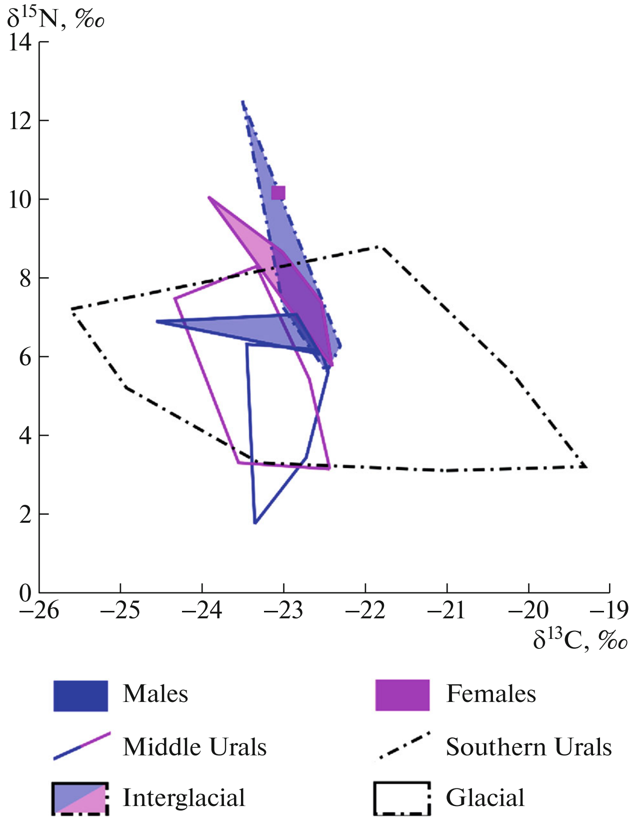

Fig. 1. Distribution of bone collagen $\delta^{13}C$ and $\delta^{15}N$ (‰) of small cave bear *U.* (*S.*) *rossicus* males (blue) and females (pink) of the Middle and Southern Urals.

between the interglacial and glacial periods were, respectively, 0.4 and 1.4‰ in males and 0.3 and 2.2‰ in females. Pooled samples of males and females did not differ in $\delta^{13}C$, while their $\delta^{15}N$ values differed by 2.0‰. In the Southern Urals, differences in $\delta^{13}C$ and $\delta^{15}N$ could be estimated only for pooled samples of males and females of the interglacial and glacial peridos (Table 1). The differences were 0.8‰ for $\delta^{13}C$ and 3.2‰ for $\delta^{15}N$ (Table 1). In the interglacial period, geographical differences in $\delta^{13}C$ and $\delta^{15}N$ were, respectively, 0.5 and 1.3‰ between males from the Middle and Southern Urals and 0.2 and 1.1‰ between the pooled samples of males and females. In the glacial period, differences in $\delta^{13}C$ and $\delta^{15}N$ were 1.0 and 0.1‰, respectively, between the pooled samples of small cave bear males and females from the Middle and Southern Urals.

Differences in $\delta^{13}C$ between all samples were relatively low, not exceeding 1.0‰. Differences in $\delta^{15}N$ between the samples were greater. Gender differences in $\delta^{15}N$ did not exceed 1.0‰ in the Middle Urals in the interglacial and glacial periods. Geographical differences in $\delta^{15}N$ between males were 1.3‰ in the interglacial period. Chronological differences in $\delta^{15}N$ between bears of the interglacial period and those of the glacial period were greater, ranging from 1.4‰ between males of the Middle Urals to 3.2‰ between bears of the Southern Urals (Table 1).

When the distribution of $\delta^{13}C$ and $\delta^{15}N$ values was analyzed, almost no overlap was observed between males and females of the Middle Urals in the interglacial period and between interglacial and glacial males and females (Fig. 1). There was virtually no overlap between the distributions of males from the Middle and Southern Urals in the interglacial. Values of males and females from the glacial of the Middle Urals overlapped substantially and fell nearly totally within the range of values obtained for bears of the glacial of the Southern Urals (Fig. 1). It should be noted that a great variation in $\delta^{13}C$ and $\delta^{15}N$ was observed in Southern Ural bears of the glacial period, and their distribution covered the majority of values obtained for Ural bears of both glacial and interglacial periods (Fig. 1).

The above differences in the distribution of $\delta^{13}C$ and $\delta^{15}N$ point to differences in diet between gender, chronological, and geographical groups of small cave bears in the Urals. Females had higher $\delta^{15}N$ as compared with males in the Middle Urals of the interglacial period. South Ural males showed higher $\delta^{15}N$ values than Middle Ural males did (Fig. 1). A decrease in

$\delta^{15}N$ was observed upon a transition from the interglacial to the glacial in males and females of the Middle Urals. The same was detected in bears of the Southern Urals (Fig. 1). Thus, changes in $\delta^{15}N$ were mostly responsible for the gender, chronological, and geographical differences.

Protein consumption, or the portion of meat in the diet in our case, determines the $\delta^{15}N$ value to a great extent [18, 19]. The above differences in $\delta^{15}N$ reflect different degrees of carnivory in gender, chronological, and geographical groups of small cave bears of the Urals. In the interglacial period, females led a more predatory lifestyle than males in the Middle Urals and males of the Southern Urals had a higher degree of carnivory than males of the Middle Urals did. A trophic shift to a greater degree of herbivory occurred in bears of the Middle and Southern Urals upon a transition to the glacial period. The trophic shift involved both males and females in the Middle Urals. A similar trend was detectable in the Southern Urals.

As mentioned above, differences in mean $\delta^{13}C$ did not exceed 1.0‰ between gender, chronological, and geographical groups. Differences in mean $\delta^{15}N$ did not exceed 2.2‰ in the majority of cases and was only observed to be 3.2‰ in a single case (Table 1). Samples are assigned to different trophic levels when a difference in $\delta^{13}C$ is 0–2‰ and a difference in $\delta^{15}N$ is 3–5‰ [3]. The differences in mean $\delta^{13}C$ and mean $\delta^{15}N$ were found to be 0.8 and 3.2‰, respectively, between interglacial and glacial pooled samples of males and females of the Southern Urals (Table 1). Thus, the differences between the samples correspond to differences between different trophic levels. The small cave bear population of the Southern Urals possibly passed from a higher to a lower trophic level upon the transition from the interglacial to the glacial period.

The $\delta^{13}C$ and $\delta^{15}N$ contents in bone collagen display considerable differences between gender, geographical, and chronological samples of small cave bears of the Middle and Southern Urals. In the Middle Urals, females led a more predatory lifestyle than males during the interglacial period, and the trophic niches of males and females converged due to an increase in herbivory during the transition to the glacial period. Males showed geographical differences in diet in the interglacial period; i.e., males of the Southern Urals led a more predatory lifestyle than in the Middle Urals. A common shift to a greater degree of herbivory occurred in both males and females in the Middle and Southern Urals upon the transition from the interglacial to the glacial period. The extent of changes in $\delta^{13}C$ and $\delta^{15}N$ values in the Southern Urals during the transition was found to correspond to differences between trophic levels.

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ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This work does not contain any studies involving human and animal subjects.

CONFLICT OF INTEREST

The authors of this work declare that they have no conflicts of interest.

REFERENCES

1. DeNiro, M.J., Postmortem preservation and alteration of in vivo bone collagen isotope ratios in relation to palaeodietary reconstruction, *Nature*, 1985, vol. 317, no. 31, pp. 806–809.
2. DeNiro, M.J. and Epstein, S., Influence of diet on the distribution of carbon isotopes in animals, *Geochim. Cosmochim. Acta*, 1978, vol. 42, no. 5, pp. 495–506.
3. Bocherens, H. and Drucker, D., Trophic level isotopic enrichment of carbon and nitrogen in bone collagen: Case studies from recent and ancient terrestrial ecosystems, *Int. J. Osteoarchaeol.*, 2003, vol. 13, no. 1/2, pp. 46–53.
4. Bocherens, H., Isotopic insights on cave bear palaeodiet, *Hist. Biol.*, 2019, vol. 31, no. 4, pp. 410–421.
5. Kosintsev, P.A., Simonova, G.V., and Konovalova, K.Yu., The first data on the nutrition of the ural cave bear (Mammalia, Carnivora, Ursidae, *Ursus* (*spelaearctos*) *kanivetz* Verestchagin, 1973) based on the results of the analysis of isotopes ^{13}C and ^{15}N, *Dokl. Biol. Sci.*, 2023, vol. 510, no. 1, pp. 160–162. https://doi.org/10.1134/S0012496623700357
6. Silaev, V.I., Parshukova, M.N., Gimranov, D.O., et al., Mineralogical and geochemical features of cave fossilization of bone detritus on the example of Imanay cave (South Urals), *Vestn. Permsk. Univ.: Geol.*, 2020, vol. 19, no. 4, pp. 323–358. https://doi.org/10.17072/psu.geol.19.4.323
7. Gimranov, D., Bocherens, H., Kavcik–Graumann, N., et al., The cave bears from Imanay Cave (Southern Urals, Russia), *Hist. Biol.*, 2022, pp. 1–9.
8. Vereschagin, N. and Baryshnikov, G., Small cave bear *Ursus* (*Spelearctos*) *rossicus uralensis* from the Kizel Cave in the Ural (Russia), *Geoloski Zbornik*, 2000, no. 15, pp. 53–66.
9. Barlow, A., Paijmans, J.L.A., Federica, A., et al., Middle Pleistocene genome calibrates a revised evolutionary history of extinct cave bears, *Curr. Biol.*, 2021, vol. 31, no. 8, pp. 1771–1779.

10. Fadeeva, T.V., Kosintsev, P.A., Lapteva, E.G., et al., Makhnevskaya Ledyanaya Cave (Middle Urals, Russia): Biostratigraphical reconstruction, *Quat. Int.*, 2020, vol. 546, pp.135–151.

11. Pacher, M. and Stuart, A.J., Extinction chronology and palaeobiology of the cave bear (*Ursus spelaeus*), *Boreas*, 2008, vol. 38, pp. 189–206.

12. Kosintsev, P.A., Yakovlev, A.G., Plasteeva, N.A., et al., Mammalian fauna of the Late Pleistocene from the Barsuchiy Dol cave (Southern Urals), *Russ. J. Theriol.*, 2022, vol. 21, no. 2, pp. 180–191.

13. Gimranov, D.O. and Kosintsev, P.A., Cave Bears (*Ursus spelaeus* sensu lato) of the Urals, *Paleontol. J.*, 2022, vol. 1, pp. 97–106. https://doi.org/10.31857/S0031031X22010068

14. Vorob'ev, A.A., Dimensions of long tubular bones of a large cave bear of the Middle Urals, *Materialy konferentsii molodykh uchenykh "Sovremennye problemy populyatsionnoi, istoricheskoi i prikladnoi ekologii"* (Proc. Conf. Young Sci. "Current Problems in Population, Historical, and Applied Ecology") Yekaterinburg: Ekaterinburg, 2001, pp. 38–41.

15. Vorob'ev, A.A., Stages of postnatal ontogenesis of the skeleton of a large cave bear, *Materialy konferentsii molodykh uchenykh "Biota gornykh territorii: Istoriya i sovremennoe sostoyanie"* (Proc. Conf. Young Sci. "The Biota of Highlands: History and Current State"), Yekaterinburg: Akademkniga, 2002, pp. 22–28.

16. Kosintsev, P.A. and Vorob'ev, A.A., Biology of Large Cave Bear (*Ursus spelaeus* Ros. et Hein) in the Ural Mountains, in *Mamont i ego okruzhenie: 200 let izucheniya* (Mammut and Its Environment: 200 Years of Investigations), Rosanov, Yu.A., Ed., Moscow: GEOS, 2001, pp. 266–278

17. Fosse, P. and Cregut-Bonnoure, E., Ontogeny/growth of (sub)modern brown bear (*Ursus arctos*) skeleton: A guideline to appraise seasonality for cave bear (*Ursus spelaeus*) sites?, *Quat. Int.*, 2014, vols. 339–340, pp. 275–288

18. Chisholm, B.S., Variation in Diet Reconstructions Based on Stable Carbon Isotopic Evidence, in *The Chemistry of Prehistoric Human Bone*, Cambridge: Cambridge Univ. Press, 1989, pp. 10–37.

19. Bocherens, H., Billiou, D., Patou-Mathis, M., Bonjean, D., Otte, M., and Mariotti, A., Paleobiological implications of the isotopic signatures (^{13}C, ^{15}N) of fossil mammal collagen in Scladina Cave (Sclayn, Belgium), *Quat. Res.*, 1997, vol. 48, no. 3, pp. 370–380.

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