

The diet of the human groups buried in a late- and post-Medieval rural parish cemetery in Libkovice (Czech Republic)

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ABSTRACT: Libkovice is a village in the northwestern Czech Republic that was demolished at the end of the last century due to the expansion of a nearby mine. The former church cemetery has been a subject to bioarchaeological excavation and research, where some 850 burials from the 13th to the 19th Century have been discovered so far. With the application of stable isotope analysis, it has also been possible to uncover the dietary patterns of this exemplary rural Central European community, which was the aim of this study.

The materials analysed here consist of samples from long bones of 56 burials and 18 animal bones discovered in Libkovice during the 2019/21 excavations. It has been employed stable carbon ($\delta^{13}\text{C}$) isotope analysis to determine the average contributions of foods derived from the C3 plants.

Statistically significant differences were found between the analyzed fauna and human samples for nitrogen ($F=47.4$ $p<0.05$) and carbon ($F=19.18$ $p<0.05$). There were no statistically significant differences in the analyzed animal and human samples between the specific centuries. When considering the ages of various human individuals, the results indicated statistically significant differences in nitrogen isotopes ($F=7.71$ $p<0.05$) between children from the infants I group and older children together with adults from the Middle Ages, as well as between children from the infants I group and adults ($F=3.3$, $p<0.05$) from the modern times. The proportion of food from C3 plants that made up the diets of the studied population was on average 89%, and the potential proportion of freshwater fish in the diet could be higher than 80%.

The similarity between the chronologically diverse groups may indicate similar strategies for food acquisition. The results obtained for the population of Libkovice are very similar to the diets of the populations living in Central Europe broadly during the two periods.

KEY WORDS: human remains, isotopic analyses, rural communities, late- and post-Medieval period, Central Europe.

Original article

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Introduction

Libkovice (Usteckí Region, Most District) was a village in northwestern Czech Republic, which was occupied for approximately 800 years until the late 20th Century (Fig. 1).

At the end of the 20th Century, due to plans to expand a nearby open-pit lignite mine, the inhabitants of Libkovice were resettled, and the village was razed.

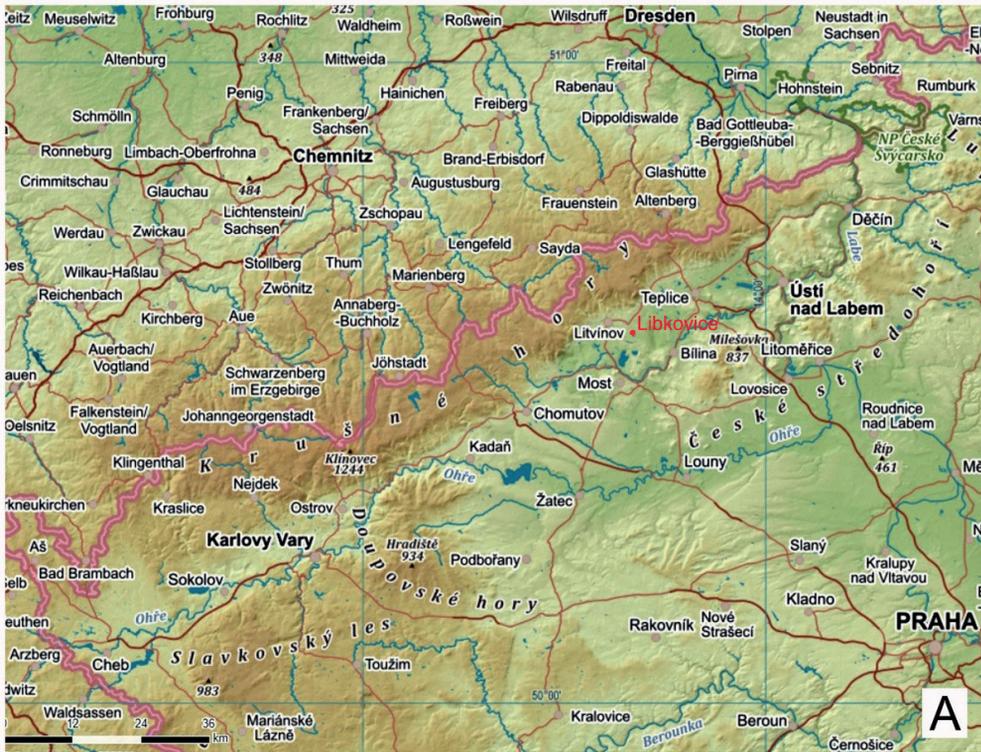


Fig. 1. Libkovice, Usteckí District: A – location of the village (red dot and caption) on the map of North-West Bohemia (source: mapy.cz), B – 19th century drawing of the St. Nicholas church (by Vařeka 2020), C – location of the village (red circle) on Petri's map from 1764 (by Kučera 2022)

The town's abandonment has created an opportunity for scholars to carry out a multi-faceted archaeological study of the entire area of the medieval and modern village, a region that until the 19th Century had a rural population (Biel 2021). Archaeological research carried out from 1995–1996 demonstrated that the central element of the layout of Libkovice was St. Nicholas Church since its foundation in the 13th Century (Vařeka 2020). Until around the mid-19th Century, there was a cemetery around the church, which was the burial place of successive generations of villagers. Comprehensive archaeological and anthropological research of this cemetery began in 2019, lasting until the end of 2022, which led to the discovery of over 850 burials in various states of preservation (Fig. 1). This research aimed to learn about funerary customs and changes in the management of the cemetery space throughout its use-life, as well as to learn about the biology, health, and demographics of the community buried there. Although the field research is ongoing, some preliminary results have already been published (e.g., pathology [Kwiatkowska et al. 2021]).

Isotopic studies are widely applied in biological anthropology and bioarchaeology and are applied broadly to various chronological periods. Isotopic analyses in anthropology are performed to study nutritional strategies of populations living in different historical periods, to reconstruct the process of weaning, or to describe migration phenomena.

Of particular use here, isotopic analysis can provide us with a direct insight into an individual's diet is possible (Schoeninger 2011), as the isotopic signature of the consumed food is reflect-

ed in predictable ways in the isotopic ratios in the body's tissues (Pate 1994; Schwarcz and Schoeninger 2012). Stable isotope analyses of human remains have been applied to a variety of cases with variable time depths. These include, but are not limited to, studies on the diet of Neanderthals (Richards and Trinkaus 2009), prehistoric hunter-gatherer populations (Pate 1998; Katzenberg et al. 2010), and Neolithic populations (Pearson et al. 2015), medieval populations (Reitsema et al. 2010; Tomczyk et al. 2021); and modern populations (Lamb et al. 2014; MacKinnon 2015; Tomczyk et al. 2020).

Based on previously published studies on the variability of the carbon isotopic ratios ($\delta^{13}\text{C}$), depending on which photosynthetic pathway is dominant in plants (Smith and Epstein 1971; van der Merwe and Vogel 1978), it is possible to determine the contributing proportions of C3 and C4 pathway plants in the diet based on the value of $\delta^{13}\text{C}$ bone collagen (e.g., Schwarcz and Schoeninger 1991).

The values of stable nitrogen isotopes ($\delta^{15}\text{N}$), on the other hand, demonstrate variation depending on the trophic level, which makes it possible to study the proportion of proteins of marine origin in the diet (Schoeninger et al. 1983; Choy and Richards 2009). The higher the trophic level, the greater the $\delta^{15}\text{N}$ differences among animal species. By comparing human nitrogen isotopic values with those of local herbivores (e.g., domesticated cattle), carnivores (e.g., dogs), or other omnivores (e.g., pigs), it is possible to determine the sources of protein in the human individuals' (Richards and Trinkaus 2009).

As such, this study aimed to reconstruct the diets and food preferences (e.g.,

% proportion of animal protein, C3 vs. C4 diet, and proportion of freshwater fish) of people buried in the Libkovice cemetery.

Material and Methods

Samples from long bones of 56 human skeletons (38 children and 18 female adults- selected randomly) discovered in the cemetery of the St. Nicholas Church in Libkovice (district Most, Czech Republic) were obtained for the study. The subjects were assigned to six age categories: 1) infans I (aged 0 to 7 years), 2) infans II (aged 7 to 12–14 years), 3) juvenis (aged 12–14 to 20–22 years), 4) adultus (aged 20–22 to 30–35 years), 5) matures (aged 30–35 to 50–55 years) and 6) senilis (over 55) (Malinowski and Strzałko 1989).

To build a local baseline, animal remains, including bone fragments from 18 domesticated animals (cattle, pig), from the same site were also analyzed. Both human and animal bones came from two historical periods, the Middle Ages (13th–15th Century) and modern times (16th–19th Century) (Tab. 1).

Isolation of collagen and measurement of stable isotopes

Bone collagen was isolated from cortical bone fragments using Bocherens' protocol (1997). Isotopic measurements were performed using a Costech ECS 4010 elemental analyzer coupled via a Thermo Scientific ConFlo IV to a continuous flow Thermo Scientific Delta V Advantage mass spectrometer at the Scottish Universities Environmental Research Center (SUERC). The values of the isotopic composition of nitrogen and carbon were

expressed in delta notation ($\delta^{15}\text{N}$, $\delta^{13}\text{C}$). The international laboratory standards used in the analyses were atmospheric nitrogen (AIR) and PeeDeeBelemnite (PDB).

Evaluation of diagenetic changes in collagen

To obtain reliable results from isotopic analyses, the C/N ratio was determined (DeNiro 1985; Ambrose and Norr 1993) and checked to ensure that it was within the acceptable range of 2.9 to 3.6 (van Klinken 1999). In addition, the percentage of carbon and nitrogen in collagen was determined for each sample. Acceptable values of these parameters were a minimum of 25% carbon and a minimum of 10% nitrogen in collagen (Ambrose 1990; Pate 1997; Van Klinken 1999; Pate et al. 2016).

Paleodiet reconstruction

A linear mixing model based on carbon isotopic values was used to determine the average proportion of foodstuffs from C3 plants (Dewar and Pfeiffer 2010; Pate et al. 2016). Experimental values from the populations from Miechow (southern Poland) were incorporated into the model (Mnich et al. 2020). As a result, no isotopic correction was necessary. Mixing models proposed by Hedges and Reynard (2007) and Fraser et al. (2013) were used to reconstruct the percentage of animal protein in the diet. Isotopic enrichment between the different trophic levels described by contemporary models should lie in the range of 3‰ – 5‰ (DeNiro and Epstein 1981; Sponheimer et al. 2003; Robbins et al. 2005). In this study, the differences between the individual trophic levels were assumed to be 4‰, which is a result consistent with the literature (DeNiro and Epstein 1981; Sponheimer et al. 2003; Robbins et al. 2005).

Statistical methods

Statistical analyses were performed using Statgraphics Centurion 18 software. These analyses included cluster analysis using Ward's method, linear models reconstructing the percentage of animal protein in the diet, and the proportion of C3 vs C4 plant food products, as well as the parametric ANOVA test. All tests were performed assuming a 95% confidence level.

Results

The values obtained for the samples' collagen C/N ratio were in the range of 3.1–3.4 (mean 3.25) and did not exceed the limit of the norm for diagenetically unchanged samples (2.9–3.6). The percentage of carbon in all samples was over 41% and nitrogen was over 14%. Therefore, all analyzed samples of human and animal bone collagen were not diagenetically altered and presented reliable data (Tab. 1, Fig. 2).

Table 1. Human and animal skeletal material from Libkovice site subjected to carbon and nitrogen isotope analysis ($\delta^{15}\text{N}$ nitrogen isotope result, $\delta^{13}\text{C}$ – carbon isotope result, %N – percentage of nitrogen in the sample, %C – percentage of carbon in the sample, CNMolar – diagenesis index; C/N – ratio values of collagen in the sample

Sample ID	Sample Type	Age/Species	Gender	Dating	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	%N	%C	CN Molar
1013a	femur/humerus	Infans I (18 months)		13-15 th century	15,1	-19,8	14,9	43,4	3,4
1013b	femur/humerus	3-4 years		13-15 th century	12,9	-20,2	14,9	42,9	3,4
1049	femur	Infans I (5-6 months)		13-15 th century	14,4	-19,8	14,6	41,4	3,3
1087	femur	5-6 years		13-15 th century	12,9	-19,9	14,0	40,0	3,3
1089	tibia	4 years		13-15 th century	13,3	-19,8	14,6	42,1	3,4
1093	femur	9-10 years		13-15 th century	13,8	-19,8	14,7	41,8	3,3
1117	ulna	5-6 years		13-15 th century	13,4	-19,6	14,6	41,6	3,3
1163	femur	Infans I (18-24 months)		13-15 th century	14,5	-19,9	14,7	43,1	3,4
1191	femur/humerus	Infans I ((24 months)		13-15 th century	14,6	-20,1	14,9	42,8	3,4
1193	femur	10-12 years		13-15 th century	13,8	-19,5	14,4	41,7	3,4
1259	femur	10-11 years		13-15 th century	12,4	-20,2	15,0	42,5	3,3
1299	femur	(Infans I) 0-12 months		13-15 th century	16,0	-19,5	14,5	41,0	3,3
1307	femur	6-7 years		13-15 th century	13,1	-19,3	15,0	41,4	3,2
1309	femur	8-9 years	M	13-15 th century	12,9	-19,9	14,5	40,6	3,3
1343	femur	Infans I (7-12 months)		13-15 th century	15,6	-19,6	15,2	43,8	3,4

Sample ID	Sample Type	Age/Species	Gender	Dating	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	%N	%C	CN Molar
1349	femur	Infans I (0-1 months)		13-15 th century	14,6	-19,1	15,3	43,2	3,3
1353	femur	Infans I (6 months)		13-15 th century	12,0	-19,9	15,3	42,6	3,2
1369	tibia	13-14 years	M	13-15 th century	11,1	-19,2	15,3	43,2	3,3
1371	femur	12-13 years		13-15 th century	11,4	-19,8	15,1	42,7	3,3
1379	femur	7-8 years		13-15 th century	13,3	-20,2	15,5	43,1	3,2
1179	clavicle	Maturus	F	13-15 th century	11,4	-20,2	15,7	42,3	3,1
1401	tooth	Adultus	F	13-15 th century	10,6	-19,5	15,7	42,2	3,1
1493	humerus	Maturus	F	13-15 th century	13,1	-19,3	15,8	42,3	3,1
1553	fibula	Maturus	F	13-15 th century	12,4	-20,0	15,6	43,1	3,2
1561	clavicle	Maturus	F	13-15 th century	11,8	-20,0	15,7	42,2	3,1
1563	humerus	Adultus	F	13-15 th century	12,0	-20,0	15,5	42,6	3,2
1579	ulna	Maturus	F	13-15 th century	12,2	-20,0	16,0	42,2	3,1
1593	femur	Maturus	F	13-15 th century	13,6	-19,7	15,4	42,9	3,3
1601	femur	Adultus	F	13-15 th century	13,7	-19,3	15,3	42,1	3,2
1107	femur	Infans I/ Infans II		16-19 century	9,0	-20,6	14,0	40,8	3,4
1051	femur	0-3 months		16-19 century	12,6	-19,8	14,9	43,2	3,4
1025	femur	4-5 years		16-19 century	12,2	-20,0	14,4	40,9	3,3
1139	femur	Infans		16-19 century	15,5	-19,1	15,6	43,7	3,3
1145	tibia	9-10 years		16-19 century	11,3	-20,1	14,1	39,7	3,3
1189	femur	14-15 years		16-19 century	13,6	-19,6	14,0	40,7	3,4
1219	humerus	Juvenis		16-19 century	11,7	-19,8	14,0	40,7	3,4
1237a	humerus	8-9 years		16-19 century	13,4	-19,7	14,2	40,4	3,3
1237b	femur	7-8 years		16-19 century	13,3	-19,7	14,3	40,7	3,3
1247	femur	11-12 years		16-19 century	12,0	-19,4	14,1	40,4	3,3
1301	femur	2-3 years		16-19 century	14,1	-19,6	15,4	43,3	3,3
1315	femur	3,5-4,5 years		16-19 century	12,6	-20,1	14,6	42,3	3,4
1329	humerus	8-9 years		16-19 century	12,8	-20,3	15,1	42,4	3,3
1391	femur	13-14 years		16-19 century	12,8	-19,8	15,2	43,0	3,3
1393	humerus	6-8 months		16-19 century	15,7	-19,6	15,4	43,6	3,3
1405	femur	Infans II		16-19 century	13,9	-20,0	14,7	41,0	3,3
1411	femur	Infans II/Juvenis		16-19 century	12,0	-19,8	15,7	43,7	3,2
1443	femur	Juvenis		16-19 century	14,1	-19,8	15,2	42,4	3,3
1069	humerus	Adultus	F	16-19 century	12,7	-19,7	15,7	42,8	3,2
1101	ulna	Adultus	F	16-19 century	12,4	-19,7	15,3	43,7	3,3

Sample ID	Sample Type	Age/Species	Gender	Dating	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	%N	%C	CN Molar
1169	ulna	Maturus	F	16-19 century	13,4	-19,8	14,6	40,3	3,2
1197	ulna	Adultus	F	16-19 century	12,0	-20,1	13,6	38,5	3,3
1227	femur	Adultus	F	16-19 century	11,8	-19,7	15,2	42,6	3,3
1243	radius	Adultus	F	16-19 century	10,8	-20,2	15,6	41,7	3,1
1317	humerus	Adultus	F	16-19 century	12,1	-19,9	15,4	41,6	3,2
1383	ulna	Adultus	F	16-19 century	10,5	-20,0	15,2	41,9	3,2
1395	ulna	Adultus	F	16-19 century	13,1	-19,8	15,5	42,7	3,2
Q07	mandible/ tooth	Pig		16-19 century	9,5	-21,1	15,7	41,7	3,1
Q18	mandible/ tooth	Bovines		16-19 century	7,8	-20,8	13,9	38,6	3,2
Q27	tooth	Bovines		16-19 century	8,4	-21,4	15,0	40,1	3,1
Q29	mandible	Pig		16-19 century	6,1	-20,7	13,8	37,7	3,2
Q30	long bone	Bovines		13-15 th century	7,1	-20,9	14,9	39,9	3,1
Q32	tooth	Bovines		13-15 th century	9,2	-21,1	15,2	42,0	3,2
Q33	long bone	Bovines		13-15 th century	6,3	-20,9	14,9	39,9	3,1
Q39	mandible	Pig		13-15 th century	8,0	-20,0	14,5	39,8	3,2
Q40/41	tooth	Bovines		13-15 th century	7,3	-20,9	15,5	42,3	3,2
Q47/56	long bone	Bovines		13-15 th century	7,7	-20,2	14,8	41,2	3,2
Q48/57	tooth	Bovines		13-15 th century	7,8	-20,5	15,5	41,8	3,1
Q66	tooth	Bovines		13-15 th century	7,1	-20,1	15,2	40,7	3,1

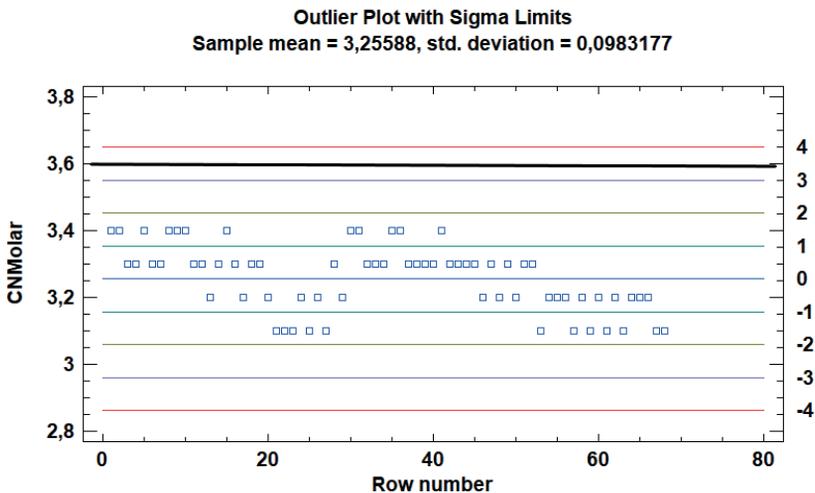


Fig. 2. Values of the diagenetic index C/N of all analysed bone samples. The black line shows the limit for post-mortem non-diagenetic samples (C/N=3.6) (The row number – the number of the next sample)

Reconstruction of diet

A comparison was made between carbon and nitrogen isotopic values obtained for human and animal samples from the two analyzed periods (the Middle Ages and modern times). The mean value of $\delta^{15}\text{N}$ for all human samples (without distinguishing the individuals by their ages) from the Middle Ages was 13.17‰ (SD=1.34), and in the case of modern samples, the mean was 12.53‰

(SD=1.52). The results obtained for animal samples in both periods were, respectively, 7.56‰ (SD=0.85) and 7.43‰ (SD= 1.19) (Fig. 3).

For carbon isotopes, the mean value of $\delta^{13}\text{C}$ for medieval human samples was -19.76‰ (SD=0.32), and for modern times samples -19.88‰ (SD=0.37). The results for animal samples were -20.57‰ (SD=0.43) and 20.97‰ (SD=0.38), respectively (Fig. 4).

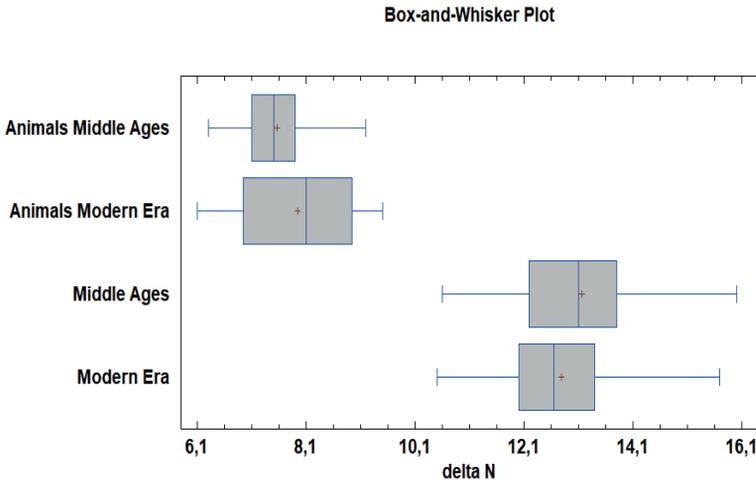


Fig. 3. Variation of nitrogen isotope levels in humans and animals in Libkovice (delta N – nitrogen isotope ratio)

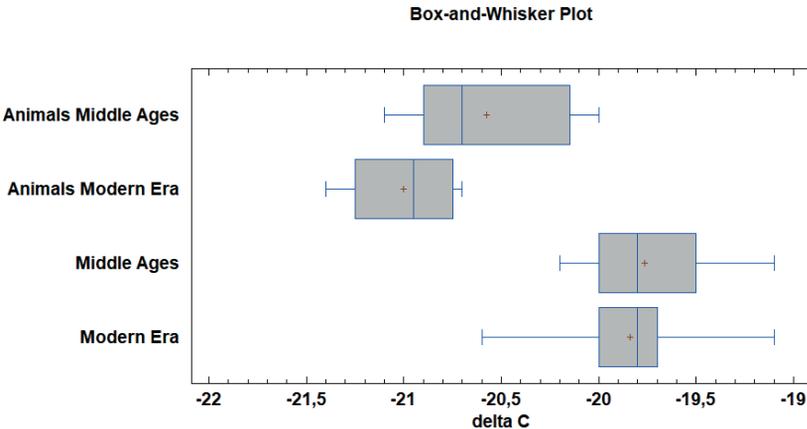


Fig. 4. Variation of carbon isotope levels in humans and animals in Libkovice (delta C – carbon isotope ratio)

Both nitrogen and carbon isotopes showed statistically significant differences between the analyzed fauna and human samples. For nitrogen it was $F=47.4$; $p<0.05$, and for carbon $F=19.18$; $p<0.05$. There were no statistically significant differences between the study periods, both for the analyzed animals and humans.

The diversity of trophic networks was analyzed via cluster analysis based on Ward's model (Fig. 5). The human samples formed a homogeneous cluster within which all individuals from medieval and modern times fit. The second cluster was formed by animal bones that differed in nitrogen and carbon levels. In the case of nitrogen isotopes, the differences were more than 4‰, indicating a trophic level shift.

At the border of the clusters was one individual (individual no. 4) from the infants I/II group who died in modern times. Such low values of both nitrogen

and carbon isotopes suggest a completely different diet compared to the other individuals, that is, their diet included a lower proportion of proteins of animal origin.

The results of the isotopic analyses of samples from the Middle Ages, considering the age distribution, indicate statistically significant differences for nitrogen isotopes ($F=7.71$ $p<0.05$) (Fig. 6).

They occur between the infants I group and older children together with adults. For the individuals from modern times, statistically significant differences were observed between infants I and adults ($F=3.31$, $p<0.05$).

The observed differences may indicate the effect of breastfeeding, which analyzes will be presented later in the other study. There were no statistically significant differences in carbon isotopes considering the age of individuals in the two historical periods analyzed.

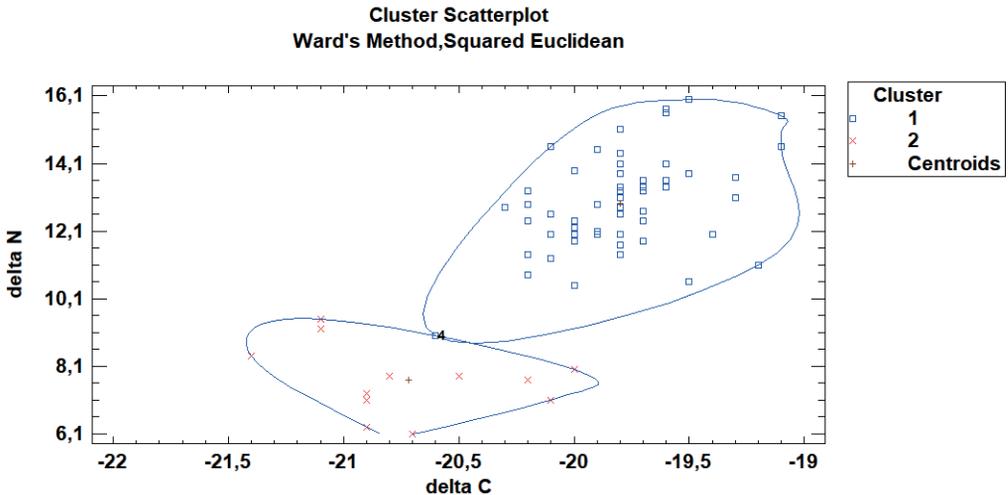


Fig. 5. Isotopic diversity of human and animal samples analysed. Cluster 1 – human specimens, cluster 2 – animal specimens

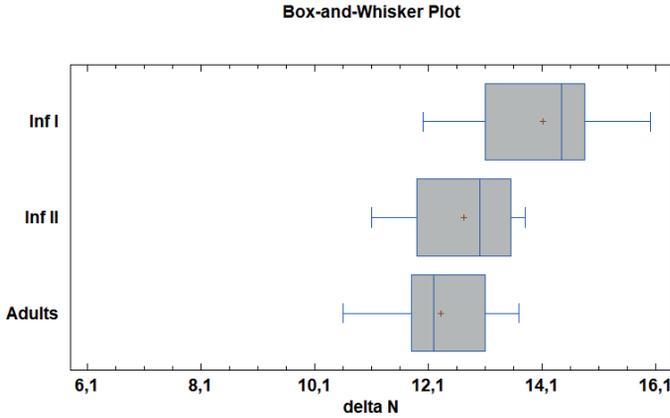


Fig. 6. Variation of nitrogen isotope levels in human samples from the Middle Ages by age in Libkovic. 1 – infans I; 2 – infans II; 3 – adults

The isotopic data obtained from the animal collagen samples do not differ from known isotope levels in medieval land animals from Poland (Reitsema et al. 2013; Krajewska 2015; Reitsema et al. 2017; Tomczyk et al. 2020). Isotopic values of carbon and nitrogen from the studied animal samples were typical for herbivores eating C3 plants (Polet and Katzenberg 2003).

C3 vs. C4 diets

To estimate the proportion of nutrients characteristic of the C3 photosynthetic pathway compared to products of C4 origin, a detailed analysis of isotopic vari-

ation was used based on the model proposed by Dewar and Pfeiffer (2010).

Considering the high probability of consumption of C4 products by individuals analyzed from the historical periods, a model was applied to the products of the C3 vs. C4 photosynthetic pathways (Pospieszny et al. 2020; Monk et al. 2020). Data from Mnich and colleagues were used to determine the carbon isotopic values within C4 plants (2020). The reconstruction was based only on adult bone samples due to the potential impact of breastfeeding on children and juveniles (Table 2).

Table 2. Estimated proportion of C3 food in the diet of studied adults (females) representing the medieval and modern periods from Libkovic based on stable carbon isotope data

Sample	n	Isotopic data		mean C3 vs. C4-based plant and animal component (%)	Range C3 (%)
		mean $\delta^{13}\text{C}$ (‰)	range (‰)		
13-15th century	9	-19,8	-20,2; -19,3	88,4 C3 vs. 8,6 C4	91,4 – 85,0
16-19th century	9	-19,9	-20,2; -19,7	89,2 C3 vs. 10,8 C4	91,4 – 87,9

Lower limit -21,4‰ (C3- based terrestrial diet, experimental data for cattle)
Upper limit -7,4‰ (C4- based terrestrial diet experimental data lead out from millet. Mnich et al. 2020)

The analysis showed that within the study group, the proportion of C3 food was dominant, averaging 89%. In addition, the low inter-individual variability indicates that both the groups representing the Middle Ages and the modern times probably consumed food of similar quality with respect to the carbon isotopes studied. The results show that, with a high probability, the studied individuals were a socially homogeneous group which predominantly consumed food made from the C3 plants, regardless of the historical period in which they lived.

The proportion of animal protein in the diet in the light of the models

We propose two scenarios to verify the percentage of animal protein in the diets of the individuals studied, taking advantage of the fact that the $\delta^{15}\text{N}$ value increases by 4‰ on average with each successive trophic level (Minagawa and Wada 1984; Schoeninger and DeNiro 1984; Sealy et al. 1987; Fraser et al. 2013). In the first model, a 100% plant-based diet was represented by the mean value obtained for herbivores. In this case, the starting point was the mean ratio of nitrogen isotopes obtained for all animals analyzed (medieval cattle = 7.5‰, modern times cattle = 8,1‰). Due to the fractionation between trophic levels (Δ plant-herbivorous fraction \approx fractionation 4‰), the predicted nitrogen level of vegetation consumed by herbivores was reconstructed, equaling 3.5‰ for both periods studied.

An animal-specific enrichment of +4‰ was also added to the $\delta^{15}\text{N}$ value to determine the value of the endpoint $\delta^{15}\text{N}$ for the linear model. The endpoint of the model for the Middle Ages was $\delta^{15}\text{N} = 11.5\text{‰}$, while for modern times

was $\delta^{15}\text{N} = 12.1\text{‰}$. The mean value of $\delta^{15}\text{N}$ for the medieval group was 12.31‰, while that of the modern population was 12.09‰.

The observed levels of $\delta^{15}\text{N}$ in the individuals studied here exceeded the assumed isotopic variability in herbivores (considering the 4‰ fractionation of isotopes) from Libkovice. The first scenario suggests that the fraction of animal protein in the diet of the medieval and modern groups probably came mostly from sources other than herbivore and omnivore meat. Similar results were obtained using the pig model. Therefore, a second scenario based on data from prehistoric freshwater fish was proposed, focusing on predatory freshwater fish such as Northern Pike (*Esox Lucius*) and Zander (*Sander lucioperca*) (Robson et al. 2016). Therefore, data from the work of Tomczyk and colleagues (2020b) were adopted as the endpoint. This point is the average of the literature data describing a potential diet consisting of 100% freshwater carnivorous fish protein. This made it possible to conclude that the hypothetical, predicted value of nitrogen isotopes for people consuming only freshwater fish was more likely. This model showed that the potential proportion of predatory freshwater fish in the diet could have been over 80%. Pike is a widely used supplementary fish in carp ponds. It allows higher production per hectare of pond and plays an important role in clearing the pond of insects, larvae and tadpoles, as well as undesirable species that come in with waters from natural reservoirs and compete with carp for food.

The observed similarity between the analyzed chronologically diverse groups is high, which may indicate similar food acquisition strategies despite the passage of multiple centuries.

Discussion

Population-based comparative analyzes were used to better understand the nutritional strategies of the analyzed community of Libkovice. It is interesting to compare the studied group against selected groups from the Middle Ages and modern times (Fig. 7).

The isotopic values of adults were compared with the $^{13}\text{C}/^{12}\text{C}$ and $^{15}\text{N}/^{14}\text{N}$ ratio data from several groups of modern animals from different ecosystems with different isotopic values (Fig. 7). A similar dispersion of the $\delta^{15}\text{N}$ values was observed for individuals from the Middle Ages and the modern times. The isotopic shifts recorded in the human samples from Libkovice (regardless of the period) against the results on animals from the same location prove that for both periods studied, cattle and pig meat were a component of the diet of the inhabitants of the area. The fact that the nitrogen isotopic ratios of the inhabitants of Libkovice are relatively high suggests that the predatory freshwater fish were also an important component of the local diet. This data calls into question the source and availability of freshwater fish for the inhabitants of Libkovice. Since its founding, the village was run by the Cistercian Order, located approx. 5 km north of the monastery in Osek (Vařeka 2020). Complexes of fishponds built in the late Middle Ages were part of the monastic property, the nearest of which was located 3–4 km east of Libkovice, on the border with the village of Liptice (Liptiz). The ponds can be seen on archival plans, e.g., on the map of Saxony published in 1764 by Issak Jacob von Petri (Kučera 2022).

Special attention should be paid to two ponds located directly in the village, included in the Ordonan Survey Map of 1842. The long tradition of small water reservoirs

and a developed water management represented by mills in Libkovice is shown by a 1240 privilege issued by Slavko, abbot of the nearby Cistercian monastery in Osek and Prussian bishop (“villam Lubcowitz ... cum duobus molendinis ... Predictus vero Wazlaus curiam, ... piscinulam domi predicte ...” Friedrich and Kristen 1962).

The results of the isotopic analyses were plotted on a graph presenting data available for populations from Poland and other European countries from various periods (Fig. 7 and 8). The Medieval and modern populations of Libkovice were characterized by isotopic values most similar to the Polish population from Radom from the 16th–17th Centuries and the 18th–19th Centuries (Fig. 7 and 8). Similar values were also recorded in studies of a Medieval group of individuals from the village of Kaldus (Fig. 7), in the Kuyavian-Pomeranian Voivodeship. The results obtained from Solt-Tételhegy in Hungary on a medieval population (mean value of $\delta^{13}\text{C}$ for enamel – 11.1‰, for dentin – 17.4‰; mean value of bone apatite $\delta^{13}\text{C}$ – 10‰, mean value of bone collagen $\delta^{13}\text{C}$ – 17.1‰) also suggest that C3 plants were the predominant type consumed.

The $\delta^{15}\text{N}$ values of dentin and bone further indicated that animal protein constituted a moderate part of the diet of the study group. Despite signs of status differences indicated by burial location, the stable nitrogen isotope values suggest that individuals had relatively egalitarian access to animal protein (Gugora et al. 2018).

The similarity to the populations from Radom and Kaldus (Poland) can be attributed to the exploitation of protein resources from inland water reservoirs, such as rivers and lakes (Reitsema et al. 2017; Tomczyk et al. 2020). The monks arrived in nearby Osek (Osseg

in German) in 1197, and Libkovice was included in their estates (Vařeka 2020).

The inter-population comparison as well as the Cistercian fishponds in the vicinity of Libkovice, support the results of our isotopic analyses. The isotopic data obtained from the animal collagen samples do not differ from known isotopic levels in Medieval land animals from Poland (Reitsema et al. 2013; Krajewska 2015; Reitsema et al. 2017; Tomczyk et al. 2020). The carbon and nitrogen isotopic values of the animal samples tested were typical of C3 plant-eating animals (Polet and Katzenberg 2003). It should be emphasized that the state of preservation

of the bone material was very good, and none of the analyzed samples exceeded critical diagenetic values.

The assumptions regarding a diet enriched with protein derived from fish are confirmed by the model approaches presented in the literature (Schulting 2018). According to Schulting (2018), the typical composition of a diet containing a variety of C3 and animal protein, derived mainly from land animals, is assumed for values of $\delta^{13}\text{C}$ below -18‰ and $\delta^{15}\text{N}$ below 12‰ . In this study, the mean values obtained for nitrogen isotopes were higher, indicating the influence of protein from higher trophic levels.

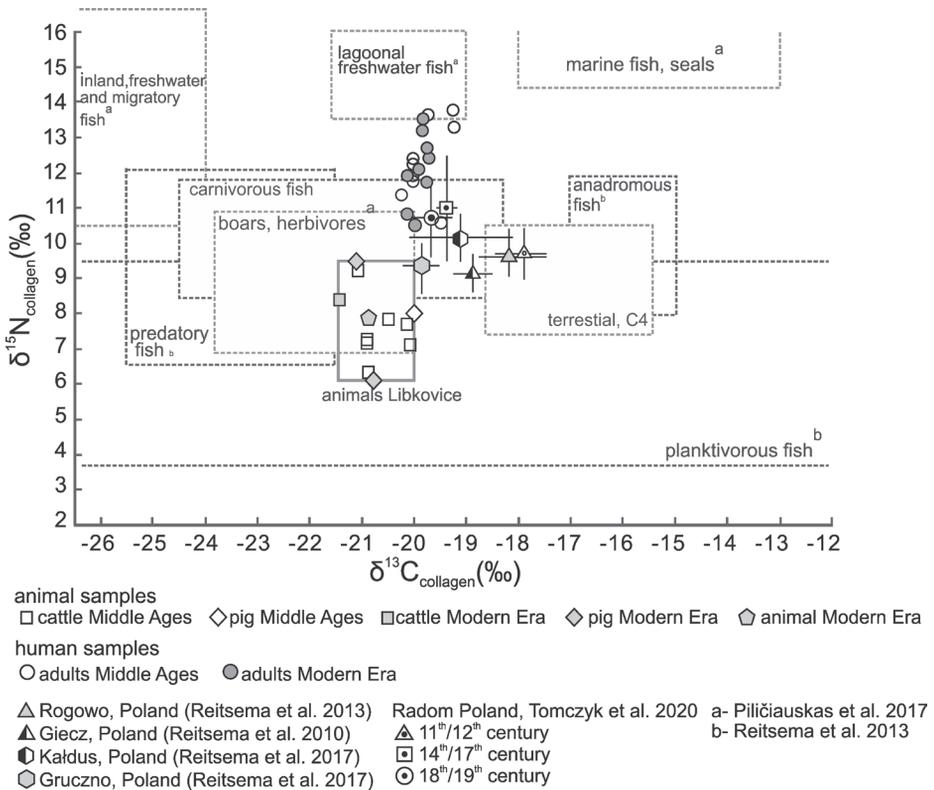


Fig. 7. C and N isotopic proportions of individuals buried at the Libkovice site compared with the environmental background of the site, isotopic ranges of different animal groups and chronologically similar populations

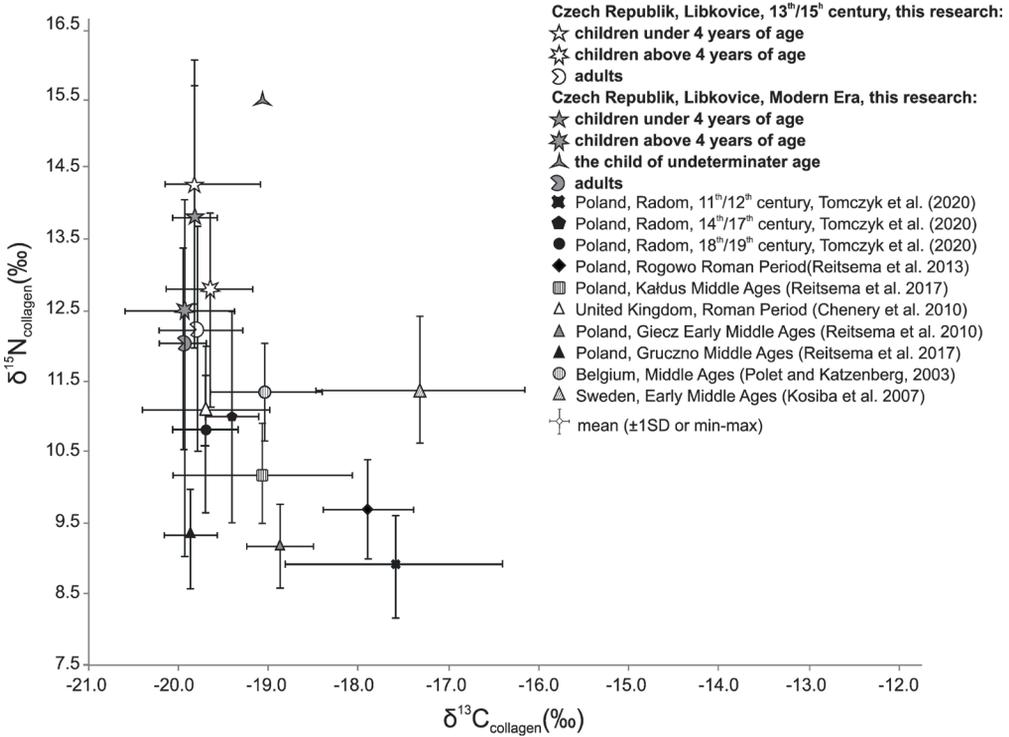


Fig. 8. Carbon and nitrogen isotopic values of adults and children from the Libkovice site in comparison with other selected European populations

Conclusions

The results of analyzes of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ stable isotopes obtained from skeletal remains excavated from the St. Nicholas Church cemetery in Libkovice indicate that the proportions of the studied isotopes differentiate well between the trophic networks characteristic of the analyzed historical fauna. The isotopic levels of the animals significantly differ from the human samples by more than 4‰.

The reconstruction of the diet indicates a general dominance of the C3 plants in the diet, reaching up to 90%, with practically imperceptible differences between the studied periods (Middle Ages vs. modern times). In addition, the

low inter-individual variability indicates that individuals representing both historical periods consumed food of similar quality, with respect to the carbon isotopes studied.

Based on the results, it can be concluded with high confidence that the individuals studied were a socially homogenous group and predominantly consumed foods from C3 plants, with a small proportion of higher carbon isotope products (e.g., sugar from sugarcane, millet, etc.). It is unlikely that they consumed the meat of animals feeding on C4 plants, such as millet, for example, since the animals' $\delta^{13}\text{C}$ values are too low, thus excluding feeding on C4 plants. The variation in the percentage of the

C3 food components indicates that the populations' diets were similar over the periods under study.

The reconstruction of the percentage of animal protein in the diet showed that by using a model that included the consumption of freshwater fish, the proportion of the diet based on protein derived from aquatic organisms could have been high and reached over 80%. Certainly, however, the remaining protein was obtained from herbivores/omnivores feeding on C3 plants. Significant similarities were observed between the analyzed, chronologically diverse groups, which may indicate that these populations maintained their food acquisition strategies and culinary practices, despite the passage of centuries, as verified by comparative population analyses.

The dispersion of $\delta^{15}\text{N}$ values in individuals from the Middle Ages is similar to the values obtained for samples from the modern periods. In contrast, the isotopic shifts recorded in human samples from Libkovice, concerning the animal background data, prove that animal protein from cattle and pigs was one of the components, albeit a minor component, of the diet during the Middle Ages and modern times. Moreover, the nitrogen isotopic ratios of those buried in Libkovice were relatively high and demonstrated values about 3‰ above the ranges characteristic of predatory freshwater fish, which shows that predatory freshwater fish were an important component of the diet, in addition to terrestrial animal protein.

This research will be expanded in the future by broadening the source database by analyzing further samples from the cemetery at the St. Nicholas Church in Libkovice, as well as human remains from an early Medieval cemetery and

a cemetery from the 19th–20th Century, both of which are located in the vicinity of Libkovice but are not yet excavated. Ultimately, this will allow us to verify the presented research results and paint a more complete picture of foodways in this region over time. Eventually, we aim to conduct archaeobotanical and zooarchaeological analyses to supplement the isotopic data, which will make it possible to obtain a full reconstruction of the diet and food production in the Central European rural community from Libkovice.

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Conflict of interests

The authors certify that they have no affiliations with or involvement in any organization or entity with any financial interest or non-financial interest in the subject matter or materials discussed in this manuscript.

Authors' contribution

AK – Manager of funded project, originator; analysis of bone material, selection of research methods, preparation of introduction; PK – Supervision of the excavation work, preparation of the historical background of the work; BK – Substantive supervision of the work, preparation of the theoretical part of the work; JW – Excavation work, transportation of material from the Czech Republic, assistance in macroscopic analysis; AL-G – Preparation of material for isotopic analyses, isolation of bone collagen, preparation of discussion; KS – Preparation of material for isotopic analyses, isolation of bone collagen, preparation of discussion; substantive proofreading of the text.

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References

- Ambrose SH, Norr L. 1993. Experimental evidence for the relationship of the carbon isotope ratios of whole diet and dietary protein to those of bone collagen and carbonate. In: Grupe G, Lambert JB, editors. *Prehistoric human bone*. Berlin, Heidelberg: Springer. 1–37.
- Biel R. 2021. Libkowice. 800-letnia wieś, którą wkrótce pochłonie kopalnia węgla brunatnego. Available at: <https://archeologia.com.pl/> [Accessed 6 January 2023].
- Chenery C, Müldner G, Evans J, Eckardt H, Lewis M. 2010. Strontium and stable isotope evidence for diet and mobility in Roman Gloucester, UK. *J Archaeol Sci* 37(1):150–63. <https://doi.org/10.1016/j.jas.2009.09.025>
- Choy K, Richards MP. 2009. Stable isotope evidence of human diet at the Nukdo shell midden site, South Korea. *J Archaeol Sci* 3(67):1312–18. <https://doi.org/10.1016/j.jas.2009.01.004>
- DeNiro MJ. 1985. Postmortem preservation and alteration of palaeodietary reconstruction of in vivo bone collagen isotope ratios in relation. *Nature* 317:806–9. <https://doi.org/10.1038/317806a0>
- DeNiro MJ, Epstein S. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochim Cosmochim Acta* 4(53):341–51. [https://doi.org/10.1016/0016-7037\(81\)90244-1](https://doi.org/10.1016/0016-7037(81)90244-1)
- Dewar G, Pfeiffer S. 2010. Approaches to estimating marine protein in human collagen for radiocarbon date calibration. *Radiocarbon* 52(4):1611–25. <https://doi.org/10.1017/S0033822200056344>
- Fraser RA, Bogaard A, Schäfer M, Arbogast R, Heaton TH. 2013. Integrating botanical, faunal and human stable carbon and nitrogen isotope values to reconstruct land use and palaeodiet at LBK Vaihingen an der Enz, Baden-Württemberg. *World Archaeol* 45(3):492–517. <https://doi.org/10.1080/00438243.2013.820649>
- Friedrich G, Kristen Z. 1962. *Codex diplomaticus et epistolaris regni Bohemiae III/2*. No. 261. *Sumptibus Academiae Scientiarum Bohemoslovenicae*. Pragae.
- Gugora A, Dupras TL, Fóthi E. 2018. Pre-dating paprika: Reconstructing childhood and adulthood diet at medieval (13th century CE) Solt-Tételhegy, Hungary from stable carbon and nitrogen isotope analyses. *J Archaeol Sci Rep* 18:151–60. <https://doi.org/10.1016/j.jasrep.2017.12.036>
- Hedges RE, Reynard LM. 2007. Nitrogen isotopes and the trophic level of humans in archaeology. *J Archaeol Sci* 34(8):1240–51. <https://doi.org/10.1016/j.jas.2006.10.015>

- Katzenberg MA, Bazaliiskii VI, Goriunova OI, Savel'ev NA, Weber AW. 2010. Diet reconstruction of prehistoric hunter-gatherers in the Lake Baikal region. Prehistoric Hunter-gatherers of the Baikal Region, Siberia. Philadelphia: University of Pennsylvania Museum of Archaeology and Anthropology.
- Krajewska M. 2015. Dziecko i jego rozwój biologiczny oraz uwarunkowania stanu zdrowia w populacjach ludzkich z okresu średniowiecza oraz czasów nowożytnych. Thesis, UMK Toruń.
- Kwiatkowska B, Bisiecka A, Pawelec Ł, Witek A, Witan J, Nowakowski D, Konczewski P, Biel R, Król K, Martewicz L, Lissek P, Vařeka P, Lipowicz A. 2021. Differential diagnosis of a calcified cyst found in an 18th century female burial site at St. Nicholas Church cemetery (Libkovice, Czechia). PLoS ONE 16(7):e0254173. <https://doi.org/10.1371/journal.pone.0254173>
- Kučera Z. 2022. Historický atlas Euroregionu Elbe/ Labe. Euroregion Elbe/Labe. ISBN: 978-80-11-00449-1.
- Lamb AL, Evans JE, Buckley R, Appleby J. 2014. Multi-isotope analysis demonstrates significant lifestyle changes in King Richard III. J Archaeol Sci. 50:559–65. <https://doi.org/10.1016/j.jas.2014.06.021>
- MacKinnon AT. 2015. Dietary reconstruction of medieval and early modern spanish populations using stable isotopes of carbon and nitrogen. [pdf] Chico State. Available at: <https://scholarworks.calstate.edu/downloads/1z40kt39g> [Accessed 6 January 2023].
- Malinowski A, Strzałko J. 1989. Antropologia. Warszawa – Poznań. PWN.
- Minagawa M, Wada E. 1984. Stepwise enrichment of 15N along food chains: further evidence and the relation between 15N and animal age. Geochim Cosmochim Acta 48:1135–40. [https://doi.org/10.1016/0016-7037\(84\)90204-7](https://doi.org/10.1016/0016-7037(84)90204-7)
- Mnich B, Mueller-Bieniek A, Nowak M, Wilczyński J, Pospuła S, Szostek K. 2020. Terrestrial diet in prehistoric human groups from Southern Poland based on human, faunal and botanical stable isotope evidence. J Archaeol Sci Rep 32:102382. <https://doi.org/10.1016/j.jasrep.2020.102382>
- Pate FD. 1994. Bone chemistry and paleodiet. J Archaeol Method Theory vol. 1, no. 2:161-209. Available at: <http://www.jstor.org/stable/20177309> [Accessed 6 January 2023].
- Pate FD. 1997. Bone collagen diagenesis at Roonka Flat, South Australia: Implications for isotopic analysis. Archaeol Ocean 32(2):170–5.
- Pate FD. 1998. Stable carbon and nitrogen isotope evidence for prehistoric hunter-gatherer diet in the lower Murray River basin, South Australia. Archaeol Ocean 33(2):92–9. <https://doi.org/10.1002/j.1834-4453.1998.tb00409.x>
- Pate FD, Henneberg RJ, Henneberg M. 2016. Stable carbon and nitrogen isotope evidence for dietary variability at Ancient Pompei, Italy. Mediterr Archaeol Archaeom 16(1):127–33. <https://doi.org/10.5281/zenodo.35526>
- Pearson JA, Bogaard A, Charles M, Hillson SW, Larsen CS, Russell N, Twiss K. 2015. Stable carbon and nitrogen isotope analysis at Neolithic Çatalhöyük: evidence for human and animal diet and their relationship to households. J Archaeol Sci 57:69–79. <https://doi.org/10.1177/1469605315582983>
- Piličiauskas G, Jankauskas R, Piličiauskienė G, Craig OE, Charlton S, Dupras T. 2017. The transition from foraging to farming (7000–500 cal BC) in the SE Baltic: a re-evaluation of chronological and palaeodietary evidence from human remains. J Archaeol Sci Rep 14:530–42. <https://doi.org/10.1016/j.jasrep.2017.06.004>

- Polet C, Katzenberg MA. 2003. Reconstruction of the diet in a mediaeval monastic community from the coast of Belgium. *J Archaeol Sci* 30(5):525–33. [https://doi.org/10.1016/S0305-4403\(02\)00183-8](https://doi.org/10.1016/S0305-4403(02)00183-8)
- Pospieszny Ł, Makarowicz P, Lewis J, Górski J, Taras H, Włodarczak P. et al. 2021. Isotopic evidence of millet consumption in the Middle Bronze Age of East-Central Europe. *J Archaeol Sci* 126:105292. <https://doi.org/10.1016/j.jas.2020.105292>
- Reitsema LJ, Crews DE, Polcyn M. 2010. Preliminary evidence for medieval Polish diet from carbon and nitrogen stable isotopes. *J Archaeol Sci* 37(7):1413–23. <https://doi.org/10.1016/j.jas.2010.01.001>
- Reitsema LJ. 2013. Beyond diet reconstruction: stable isotope applications to human physiology, health, and nutrition. *Am J Hum Biol* 25(4):445–56. <https://doi.org/10.1002/ajhb.22398>
- Reitsema LJ, Kozłowski T, Crews DE, Katzenberg MA, Chudziak W. 2017. Resilience and local dietary adaptation in rural Poland, 1000–1400 CE. *J Anthrop Archaeol* 45:38–52. <https://doi.org/10.1016/j.jaa.2016.11.001>
- Reitsema LJ, Kozłowski T, Makowiecki D. 2013. Human-environment interactions in medieval Poland: a perspective from the analysis of fauna stable isotope ratios. *J Archaeol Sci* 40:3636–46. <https://doi.org/10.1016/j.jas.2013.04.015>
- Richards MP, Trinkaus E. 2009. Isotopic evidence for the diets of European Neanderthals and early modern humans. *Proc Natl Acad Sci* 106(38):16034–9. <https://doi.org/10.1073/pnas.0903821106>
- Robbins CT, Felicetti LA, Sponheimer M. 2005. The effect of dietary protein quality on nitrogen isotope discrimination in mammals and birds. *Oecologia* 144(4):534–40. <https://doi.org/10.1007/s00442-005-0021-8>
- Robson HK, Andersen SH, Clarke L, Craig OE, Gron KJ, Jones AKG, et al. 2016. Carbon and nitrogen stable isotope values in freshwater, brackish and marine fish bone collagen from Mesolithic and Neolithic sites in central and northern Europe. *Environment Archaeol* 21(2):105–18. <https://doi.org/10.1179/1749631415y>
- Schoeninger MJ, DeNiro MJ. 1984. Nitrogen and carbon isotopic composition of bone collagen from marine and terrestrial animals. *Geochim Cosmochim Acta* 48:625–39. [https://doi.org/10.1016/0016-7037\(84\)90091-7](https://doi.org/10.1016/0016-7037(84)90091-7)
- Schoeninger MJ. 2014. Stable isotope analyses and the evolution of human diets. *Annu Rev Anthropol* 43(1):413–30. <https://doi.org/10.1146/annurev-anthro-102313-025935>
- Schulting R. 2018. Dietary shifts at the mesolithic–neolithic transition in Europe: An overview of the stable isotope data. In: JA Lee-Thorp, MA Katzenberg, editors. *The Oxford Handbook of the Archaeology of Diet*. Oxford: Oxford University Press.
- Schwarcz HP, Schoeninger MJ. 1991. Stable isotope analyses in human nutritional ecology. *Am J Phys Anthropol* 34(S13):283–321. <https://doi.org/10.1002/ajpa.1330340613>
- Schwarcz HP, Schoeninger MJ. 2012. Stable isotopes of carbon and nitrogen as tracers for paleo-diet reconstruction. In: M Basakaran, editor. *Handbook of environmental isotope geochemistry*. Berlin, Heidelberg: Springer. 725–42.
- Sealy JC, van der Merwe NJ, Lee-Thorp JA, Lanham JL. 1987. Nitrogen isotopic ecology in southern Africa: implications for environmental and dietary tracing. *Geochim Cosmochim Acta* 51:2707–17. [https://doi.org/10.1016/0016-7037\(87\)90151-7](https://doi.org/10.1016/0016-7037(87)90151-7)
- Smith BN, Epstein S. 1971. Two categories of $^{13}\text{C}/^{12}\text{C}$ ratios for higher plants. *Plant Physiol* 47(3):380–4. <https://doi.org/10.1104/pp.47.3.380>

- Sponheimer M, Robinson T, Ayliffe L, Passey B, Roeder B, Shipley L. et al. 2003. An experimental study of carbon-isotope fractionation between diet, hair, and feces of mammalian herbivores. *Can J Zool* 81(5):871–6. <https://doi.org/10.1139/z03-066>
- Tomczyk J, Szostek K, Lisowska-Gaczorek A, Jelec P, Trzeciecki M, Zalewska M, Olczak-Kowalczyk D. 2021. Dental caries and breastfeeding in early childhood in the late Medieval and Modern populations from Radom, Poland. *International J Osteoarchaeol* 31(6):1169–79. <https://doi.org/10.1002/oa.3028>
- Tomczyk J, Szostek K, Lisowska-Gaczorek A, Mnich B, Zalewska M, Trzeciecki M, Olczak-Kowalczyk D. 2020a. Dental caries and isotope studies in the population of Radom (Poland) between the 11th and 19th centuries. *Int J Osteoarchaeol* 30(6):778–88. <https://doi.org/10.1002/oa.2908>
- Tomczyk J, Regulski P, Lisowska-Gaczorek A, Szostek K. 2020b. Dental caries and stable isotopes analyses in the reconstruction of diet in Mesolithic (6815–5900 BC) individuals from Northeastern Poland. *J Archaeol Sci: Rep* 29:102141. <https://doi.org/10.1016/j.jasrep.2019.102141>
- van der Merwe NJ, Vogel JC. 1978. ¹³C content of human collagen as a measure of prehistoric diet in woodland North America. *Nature* 276(5690):815–6. <https://doi.org/10.1038/276815a0>
- van Klinken GJ. 1999. Bone Collagen Quality Indicators for Palaeodietary and Radiocarbon Measurements. *J Archaeol Sci.* 26:687–95. <https://doi.org/10.1006/jasc.1998.0385>
- Vařeka P. 2020. Archaeology of the St. Nicolas Church in demolished village of Libkovice (Liquitz) – excavations in 1995–1996 (North Bohemian Brown Coal Mining Area). A Contribution to the research of medieval village churches in Bohemia. *Archaeol Hist* 45(1):185–202. <https://doi.org/10.5817/AH2020-1-8>

