Journal of Archaeological Science 32 (2005) 1176–1189

Journal of Archaeological SCIENCE

http://www.elsevier.com/locate/jas

Reconstructing northern Chinese Neolithic subsistence practices by isotopic analysis

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Received 24 August 2004; received in revised form 18 February 2005

Abstract

Stable isotope signatures of both human and non-human animal bone samples indicate that Neolithic farmers of the Yellow and Wei River basins in China potentially cultivated millet for two reasons: as a staple for human consumption and as fodder for domesticated animals, specifically pigs, dogs, and perhaps chicken. Bone samples were analyzed from four Neolithic sites: Jiangzhai, Shijia, Xipo, and Kangjia, spanning the time period from 7000 to 4000 years ago. A combination of very high carbon isotope ratios ($\delta^{13}C = -7.7 \pm 0.4‰$) and low nitrogen isotope ratios ($\delta^{15}N = 7.5 \pm 0.5‰$) in samples of Xipo pig and dog bone suggests that these monogastric animals consumed substantial quantities of C_4 plants, almost certainly millets. In fact, the proportion of C_4 plants in animal diets appears to have been even greater than that in human diet. Stable isotope values ($\delta^{13}C = -10.0 \pm 0.8‰$; $\delta^{15}N = 8.3 \pm 0.5‰$) of human bone collagen recovered at Jiangzhai and Shijia indicate a staple role for millets, as well as the consumption of both wild and other non-C_4 domesticated plant foods. As millet agriculture and animal husbandry apparently depended on one another, a strong mutualism between them was likely established in northern China during the Neolithic. We propose that variable redistribution of agricultural products between humans and animals, depending on the availability of wild resources and annual fluctuations in agricultural output, helped ensure the stability of Neolithic human subsistence in the Yellow and Wei River basins.

Keywords: Biological anthropology; Carbon and nitrogen stable isotopes; Neolithic China; Yangshao; Longshan; Paleodiet

Recent excavations have greatly expanded our understanding of how agriculture and animal husbandry developed in the Yellow River Basin of northern China. Specific cultural trajectories for the Chinese Neolithic can now be traced in many areas [38,40,52,53,69,84]. Heavy reliance on millet agriculture and pig tending is inferred for the Yangshao and Longshan Neolithic cultures based on stable isotope analysis of human bone [13], as well as on examination of paleobotanical and faunal remains [36,51,84,85,88–90]. These findings are also consonant with the presence of abundant agricultural tools, an advanced ceramic technology, and the ubiquity of large storage pits [15,29]. Nevertheless, analysis of human skeletal remains from three Yangshao sites, Beiliu, Jiangzhai, and Shijia, has revealed low frequencies of anemia indicators and carious lesions, as well as evidence of severe masticatory stress and rapid dental wear. Contrary to an assumption of a predominately

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0305-4403/$ - see front matter
doi:10.1016/j.jas.2005.02.015

agricultural regime during Yangshao, such indicators are consistent with a human diet provided by broad spectrum foraging and hunting [47,48].

In this paper we employ stable isotope analysis of human and animal bone samples to evaluate human diet and foddering practices, as well as to reconstruct environmental differences. Isotopic ratios for oxygen, nitrogen, and carbon as measured from bone give evidence of different aspects of diet and climate (e.g. [2,22,36,55,56,58]). Carbon isotope values in collagen ($\delta^{13}C_{col}$) and apatite carbonate ($\delta^{13}C_{ap}$) bone fractions reflect the degree of reliance on C_4 plants, such as millet, maize, and sugar cane. These ratios also vary with reliance on marine resources as well as freshwater fishing [18,54]. Nitrogen isotopic composition ($\delta^{15}N$) follows trophic levels and provides an estimate of the amount of animal protein in the diet [3,4,57]. Oxygen isotope values ($\delta^{18}O$) parallel the oxygen isotope composition of local meteoric water with a species-specific enrichment coefficient and are indicative of environmental differences. The proportion of heavy oxygen isotopes increases with aridity and is commonly used to reconstruct paleoclimates and the degree of seasonality [36,65].

Comparison between the $\delta^{13}C_{col}$ and $\delta^{13}C_{ap}$ values of organic and inorganic bone fractions ($\Delta^{13}C‰_{ap-col}$) measures the difference between the isotopic composition of protein sources and that of the overall diet [5,6]. Because apatite carbonate is derived from CO_2 produced by food metabolism and all dietary macronutrient fractions (fats, proteins and carbohydrates) are used for energy metabolism, the $\delta^{13}C$ value of the carbonate fraction reflects that of the total caloric base [5,6]. Experiments and observations show that in carnivores and other non-ruminants with monogastric digestive systems, apatite carbonate $\delta^{13}C$ is enriched by about 9–9.5‰ relative to the diet [5,20,68]. On the other hand, large ruminant herbivores have a diet–apatite enrichment of about 13.5‰ [10,14,35]. This excess enrichment in ruminants is probably due to co-production of ^{13}C-depleted methane and ^{13}C-enriched CO_2 by endosymbiotic methanogenic microbes [25,45]. Humans generate little methane [19] and show diet–apatite carbon isotope enrichment similar to that of other non-ruminant animals (9–9.5‰). When protein and non-protein dietary macronutrients have similar carbon isotope ratios, bone collagen $\delta^{13}C$ is enriched by about 5‰ relative to the entire diet [3,65,66,71] and the apatite–collagen difference value ($\Delta^{13}C‰_{ap-col}$) is approximately 4.5‰ for non-ruminants and 9‰ for ruminants [5,6]. When the $\delta^{13}C$ value of dietary protein is more negative than that of non-protein components, then $\Delta^{13}C‰_{ap-col}$ values will be larger than 4.5‰. This is expected for human diets in Neolithic China that may have included high protein animal foods such as fish and deer meat from C_3-based food webs, with low $\delta^{13}C$ values, and low protein C_4 plant foods such as millet, which have high $\delta^{13}C$ values. Conversely, if the meat of C_4-feeding animals is combined with C_3 plant foods, then the $\delta^{13}C$ of dietary protein will be less negative than that of the non-protein components and $\Delta^{13}C‰_{ap-col}$ values will be smaller than 4.5‰.

1. Subsistence strategies in northern China: zooarchaeological and paleobotanical data

Millet agriculture in northern China dates back to at least 8500–7000 BP [15,82]; initial experimentation with millet may have occurred as early as 12,000 BP [60,61]. Substantial evidence of millet cultivation, including both carbonized millet and agricultural tools, is available from Early Neolithic sites of the Peiligang, Cishan, and Dadiwan cultures (7500–6800 BP) [7,51]. Early millet agriculture probably had very low productivity and therefore contributed little to the human caloric base [41]. Numerous remains of butchered wild animals found at these sites suggest continued reliance on hunting and fishing during the Early Neolithic [7,26:897–903,89].

Very early carbonized rice and rice phytoliths, dated to 9000–8000 BP, are reported from the Jiahu site in Henan province [16,17]. With the exception of the remains from Jiahu, rice is not a common component in the Neolithic paleobotanical record of Shaanxi and Henan. Individual carbonized rice fragments or phytoliths have, nevertheless, been identified from Sanmenxia Nanjiaokou [76], Zhengzhou Dahecun [82], Luoyang Xigaoya [42], Mianchi Yangshaocun [8], and Dengzhou Baligang [32] in Henan, and from Huaxian Quanhucun in Shaanxi [76].

Monogastric animals (pigs, dogs, and probably chicken) became established as the focus of animal husbandry in China during the Early Neolithic [51]. Pigs are likely to have been domesticated by 8000 BP [73,88,89]. Evidence of early chicken domestication is more controversial, because most of the available bird bone cannot be precisely identified. Even so, finds of chicken bones are reported from a number of Middle Neolithic sites, including Beishouling in Baoji province, Banpo in Xi'an, Jiangzhai in Lintong, and Nanzheng in Longgangsi, as well as Miaodigou in Shanxian and Baiying in Tangyin [51].

The bones of wild cattle are found sporadically at sites dating to the Neolithic; the earliest presently known came from a 10,000-year-old stratum at the Nanzhuangtou site [88]. However, domesticated cattle did not appear until the time of the Longshan tradition or later [86,90]. The herding of sheep/goats (*Ovis*/*Capra*) may have been practiced by the early Longshan period [90]. Finds of caprine bones at a number of Late Neolithic sites, including Kangjia (4500–4000 BP) [37,39], Huayuangzhuang (3800–3000 BC), and Zhukaigou I and II (4000–3580 BC) [30,88] are well attested.

They appear to be absent at Early and Middle Neolithic sites in the Yellow River basin.

From earliest Yangshao times, an abundance of improved agricultural tools indicates increasing agricultural efficiency. Large storage pits filled with millet became ubiquitous [15]. The most common species of millet – and the dominant crop – was foxtail millet (*Setaria italica*) [83,84]. Other cultivated grains included broomcorn millet (*Panicum miliaceum*), grand millet (*Andropogon sorghum*), and some rice (*Oryza sativa*) [29,76]. Leaf mustard seeds and Chinese cabbage (*Brassica* sp.) were common garden plants [84]. Pig tending achieved substantial importance during the Yangshao period and pig bones often dominate faunal assemblages from Yangshao sites [87]. However, in Lintong county (central Shaanxi), hunting remained more important and the ratio of pig bones to wild animal bones in contemporaneous sites there is lower. For instance, the frequency of pig bones in various levels at the Jiangzhai site ranged from only 14 to 41% of the total number of elements in the faunal assemblage [81].

2. Materials and methods

All the samples used in our analysis come from the Western Yangshao core area. Human bone specimens were obtained from three archaeological sites located in the Xi'an district and adjacent counties of Shaanxi province: Jiangzhai, Banpo, and Shijia (Fig. 1, Table 1) [79,81]. Samples of human bone were made available for our study by the Banpo Museum in Xi'an [26]. Pig (*Sus*) and dog (*Canis*) samples of cortical bone were collected by Ma Xiaolin from the middle Yangshao site of Xipo (6000–5500 BP) [27]. Samples of deer, along with wild and domesticated bovid bones (sheep/goat and buffalo), as well as additional pig and dog samples, were obtained from Kangjia [80], a Late Neolithic site associated with the subsequent Longshan archaeological tradition. Comparative samples of animal bones and modern millet specimens from China were kindly provided by Professor Li Liu of the University of Melbourne.

Collagen and apatite were prepared for analysis using methods described previously by Ambrose and colleagues [1,6]. The isotopic analysis of collagen was accomplished by combustion in tin foil capsules; purification of N_2 and CO_2 was performed with a Carlo-Erba elemental analyzer connected to a Finnegan MAT 252 isotope ratio mass spectrometer. Apatite carbonate was analyzed by reaction with 100% phosphoric acid at 70 °C in a Kiel automated carbonate preparation device connected to the MAT 252. Sample weights were 225–285 μg for collagen and 300–600 μg for apatite.

The number of individual elements analyzed in this study was dictated by the availability of pertinent sample materials. However, stable isotopes usually exhibit small within-sample variation. Thus, even limited samples can be very informative (e.g. [6,36,57]). An ad hoc power analysis was performed to estimate the sample sizes necessary for cross-site comparisons (Table 2), assuming that the standard deviation varied within the ranges presented in other stable isotope studies. The effect size of interest was set to 1, 1.5, and 2‰. Power analysis showed that even the small sample sizes available to us would be sufficient to detect a 1‰ difference at the commonly accepted power of 80% [64].

3. Results

3.1. Preservation

The effects of diagenesis on collagen isotopic composition were evaluated in three ways: from the amount of collagen in bone; from the amount of C and N in collagen; and from atomic C:N ratios. An acceptable range for C:N ratios is 2.9–3.6 [1,21]. The human bone samples in our study showed a very narrow range of variation in C:N ratios, from 3.29 to 3.38, indicating that collagen was well preserved (Table 3). The C:N ratios of the faunal samples varied from 3.16 to 3.29, also within the acceptable range. In our human bone samples, collagen concentration varied from 13% in the Banpo specimen to 2.4% in a Shijia specimen, with a mean concentration of $6.0 \pm 2.5\%$. Collagen concentrations in our samples were much lower than in fresh bone, which averages 20% [6], but are typical for prehistoric materials. The collagen concentration in the animal bone samples was higher, averaging $12.1 \pm 3.5\%$ (Table 3). The weight % C and % N in human collagen averaged $40 \pm 4.4\%$ and $13.2 \pm 1.6\%$, respectively. These values are very close to values for fresh collagen [1,3]. Thus, while our samples had clearly lost some organic matter, their C:N ratios and high C and N concentrations demonstrate that the organic matter recovered retained the composition of collagen. Therefore, the stable isotope values we measured should approximate those values at the time of death.

Apatite carbonate, the mineral portion of bone, can be subject to contamination and post-depositional alterations of isotopic composition, even when the collagen fraction of the bone is well preserved [74,78]. If carbon isotope ratios in apatite reflect dietary composition, then apatite ratios should correlate significantly with those of collagen [35]. We observed a strong positive correlation between collagen carbon and apatite carbon isotope ratios in the Neolithic bone samples (Fig. 2). That correlation was slightly higher in the animal bone (0.92; $p < 0.001$, $N = 11$) than in human bone (0.86; $p < 0.001$, $N = 15$), but was highly significant in both. These strong correlations between collagen and apatite carbon isotope ratios suggest that there was adequate apatite preservation in the samples. Moreover, the differences between ruminants and non-ruminants in $\delta^{13}C_{ap}$ and $\delta^{18}O$ correspond to those expected from the differences in their physiology, suggesting that these isotopic values were not completely reset by diagenesis. Since internal tests were consistent, we conclude that the samples were adequately preserved to permit dietary reconstruction.

3.2. Stable isotope values in human bone samples

The isotopic signature of the single Banpo human bone specimen differed greatly from those recognized for all other Yangshao samples analyzed (Table 4). Its collagen carbon, apatite carbon, and oxygen isotopic ratios fell far outside the range of variation of those elements in the pooled Jiangzhai/Shijia sample. It has the highest collagen concentration among the human samples, so diagenesis can be discounted. It is possible that this Banpo female lived in a considerably different ecological setting than the corresponding Jiangzhai and Shijia individuals, or subsisted on a substantially different and predominantly C_3 plant based diet. High oxygen isotope ratios suggest a warmer and/or drier climate. The nature of Banpo subsistence cannot be more precisely understood until additional bone specimens from this site are analyzed.

Table 1
Location and chronological placement of archaeological sites that provided samples for this study

Site	County	Years ago
Banpo	Xi'an city, Shaanxi	6800–6300
Jiangzhai	Lintong, Shaanxi	6900–6000
Shijia	Weinan, Shaanxi	6300–6000
Xipo	Lingbao, Henan	6000–5500
Kangjia	Lintong, Shaanxi	4500–4000

Table 4 shows that stable isotope values overlapped substantially between the Jiangzhai and Shijia human bone samples. The mean collagen carbon isotope ratios of those samples from the Jiangzhai and Shijia sites were practically the same (−10.0‰), suggesting a heavy reliance on C_4 plants, presumably millet. There was some between-site difference in the $\delta^{15}N$ and $\Delta^{13}C_{ap-col}$ values (8.78 ± 0.4‰ and 5.80 ± 0.7‰ for Jiangzhai vs. 8.10 ± 0.5‰ and 6.19 ± 0.7‰ for Shijia in $\delta^{15}N$ and $\Delta^{13}C_{ap-col}$, respectively). These values generally indicate a mixed diet drawn from terrestrial food sources, including a heavy reliance on C_4 plants and some consumption of animal products.

No difference was observed in $\delta^{18}O_{pdb}$ values, which averaged −9.5‰ in both samples. The standard deviation of $\delta^{18}O_{pdb}$ values for the Shijia samples is

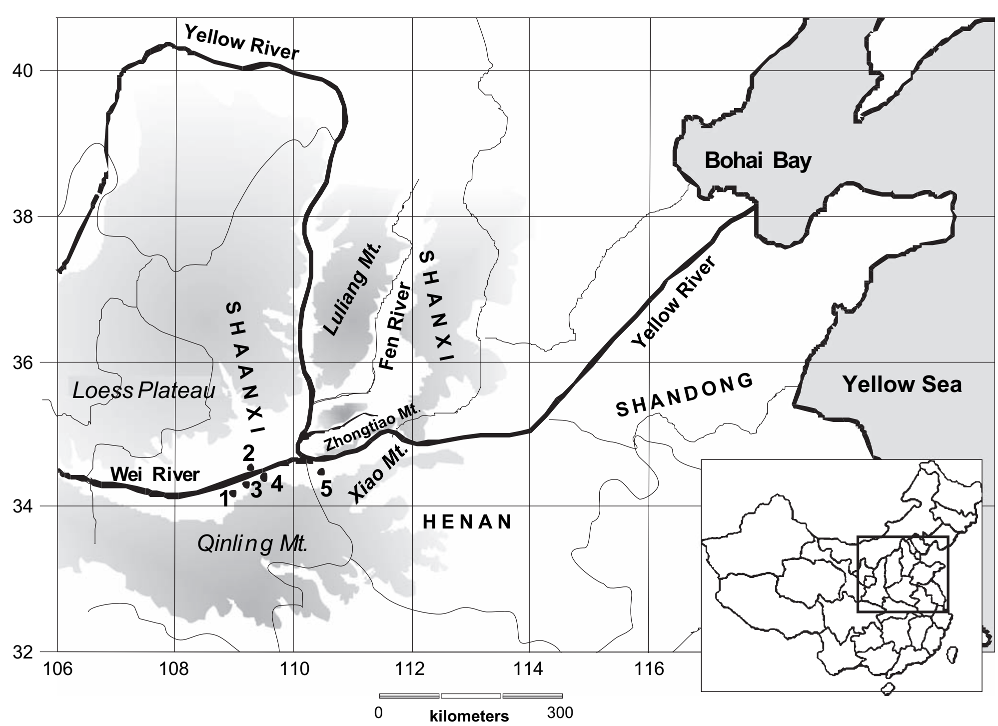

Fig. 1. Location of archaeological sites along the Wei and Yellow rivers: 1 – Banpo (34°20′N 109°10′E), 2 – Kangjia (34°38′N, 109°20′E), 3 – Jiangzhai (33°N, 109°E), 4 – Shijia (34°30′N, 109°30′E), 5 – Xipo (34°30′N, 110°40′E).

Table 2
Sample sizes necessary to detect mean differences of 1, 1.5, and 2‰ by stable isotope analysis for the range of standard deviations from 0.4 to 1.0[a]

Effect size				
Anticipated SD	0.4	0.6	0.8	1.0
1	4	7	11	17
1.5	2	4	6	8
2	2	2	4	6

[a] Sample sizes are estimated for a 0.05 significance level and 80% power based on a parametric formula from Sokal and Rolf [64].

remarkably low (0.06‰). One obvious possibility is that oxygen isotope values of the Shijia bone samples were entirely reset by the process of apatite-phase diagenesis. Oxygen isotope alterations tend to proceed faster than those for carbon [74]. Such low variation in $\delta^{18}O_{pdb}$ for the Shijia specimens might also be a consequence of the unusual burial practice evident at this cemetery [24]. At Shijia, bodies apparently were accumulated over an extended period of time and then interred in large common graves. Individuals from the same grave probably were people who died within a short time period and might have been kinsmen. Since all analyzed specimens came from two adjacent multiple burials, they likely represent contemporaries who experienced a very similar environmental regimen and thus show little variation in oxygen isotopic values.

Two-way analysis of variance was used to address the differences between results from the Jiangzhai and Shijia sites, as well as to evaluate possible sex-based differences in stable isotope values (Table 5). A significant difference was observed between the Jiangzhai and Shijia samples for $\delta^{15}N$ ($F = 7.61$, $p = 0.02$, 1 and 9 df). Lower $\delta^{15}N$ and somewhat larger $\Delta^{13}C_{ap-col}$ values for the Shijia sample suggest a somewhat smaller proportion of meat products in the human diet at Shijia. No significant difference was found between males and females; while site by sex interaction for the carbon apatite–collagen difference ($F = 4.45$, $p = 0.06$) approached, but did not reach a significant level.

Table 3
Collagen concentration, carbon and nitrogen concentrations, apatite yields, and carbon concentrations in human and other mammal bones from Neolithic sites in the Yellow River Valley of China

Site	ID#	Species	Collagen			C:N	Apatite carbonate	
			Col wt.%	C wt.%	N wt.%		Apa wt. %	C wt.%
Banpo	04	human	13.53	43.28	14.83	3.36	65.08	0.88
Jiangzhai I	03	human	6.68	39.94	13.46	3.35	67.07	1.15
Jiangzhai II	02	human	4.38	36.83	12.47	3.29	65.57	1.36
Jiangzhai I	01	human	6.27	41.8	13.68	3.34	64.84	1.17
Jiangzhai II	08	human	6.25	44.29	15.02	3.34	62.84	1.03
Jiangzhai II	06	human	4.58	40.85	13.67	3.36	58.73	1.12
Shijia	12	human	7.25	42.48	14.34	3.35	60.77	0.92
Shijia	05	human	8.00	42.21	11.59	3.33	61.98	1.17
Shijia	09	human	5.03	35.95	12.42	3.35	67.58	1.10
Shijia	10	human	2.4	26.29	8.85	3.32	43.66	1.72
Shijia	11	human	4.63	40.45	13.53	3.36	63.77	1.35
Shijia	07	human	5.9	43.91	15.06	3.36	64.01	1.06
Shijia	13	human	5.73	41.48	14.18	3.34	66.46	1.12
Shijia	15	human	5.23	40.00	13.57	3.38	64.52	1.05
Shijia	16	human	3.92	39.79?	12.38	3.34	59.01	1.16
Human mean ± SD			5.99 ± 2.51	39.97 ± 4.44	13.27 ± 1.59	3.34 ± 0.02	62.39 ± 5.83	1.16 ± 0.20
Xipo	26	pig	10.87	42.18	15.23	3.23	80.49	1.09
Xipo	27	pig	10.13	30.82	11.09	3.24	74.36	0.88
Xipo	28	dog	14.84	17.14	6.10	3.28	79.17	1.43
Kangjia	19	pig	16.65	20.93	7.55	3.23	71.74	0.90
Kangjia	21	pig	13.85	36.83	13.43	3.20	84.09	0.90
Kangjia	22	pig	13.22	40.78	14.90	3.21	67.50	1.07
Kangjia	20	dom. dog	13.64	39.52	14.35	3.21	62.50	1.05
Kangjia	23	dom. dog	5.61	18.83	6.91	3.18	68.57	1.00
Kangjia	24	*Bubalus*	12.43	39.15	14.45	3.16	69.05	1.09
Kangjia	25	sheep	17.94	40.26	14.63	3.21	68.18	0.99
Kangjia	17	deer	12.77	40.99	14.53	3.29	69.05	1.07
Kangjia	18	bovine	11.07	41.87	14.68	3.33	71.05	0.88
Animal mean ± SD			12.90 ± 3.32	33.40 ± 9.78	12.11 ± 3.56	3.22 ± 0.04	72.25 ± 6.55	1.04 ± 0.15

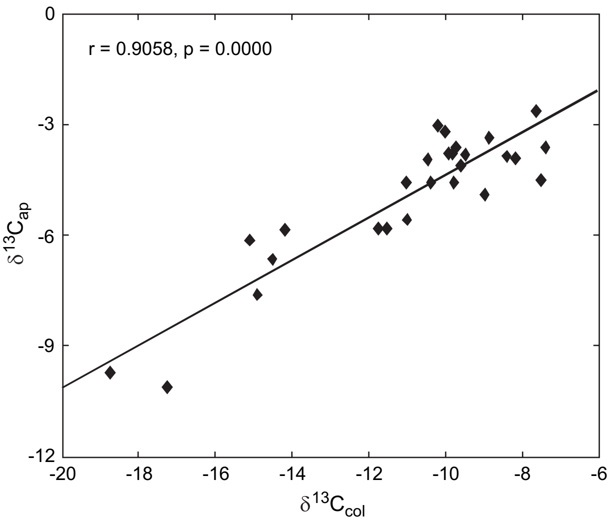

Fig. 2. Correlation between $\delta^{13}C_{col}$ and $\delta^{13}C_{ap}$ in human and animal bones. Statistical significance of the correlation suggests that variation in $\delta^{13}C_{ap}$ is related to differences in diet and physiology rather than to digenetic processes.

3.3. Stable isotope values for animal bone and plant samples

Samples of animal bone from the Xipo and Kangjia archaeological sites and modern millet were analyzed to help establish the expected isotopic composition of food sources utilized by humans during the Chinese Neolithic. Millet, a typical C_4 plant, has high $\delta^{13}C$ values; the mean value of modern Chinese millet samples was −11.7‰. The ^{13}C content was the same in seeds, leaves, and stalks (Table 6). These estimates are very close to those reported for modern millets and sorghum from the Nile River valley (mean $\delta^{13}C = -11.7‰$) [59,77]. The $\delta^{15}N$ values derived from modern Chinese samples were higher for seeds (mean $\delta^{15}N = 2.85 \pm 0.9‰$) than for leaves ($\delta^{15}N = 1.82‰$) and stalks ($\delta^{15}N = -0.04‰$).

Carbon isotope values of preindustrial millets almost certainly were higher than those of the modern samples available for our analysis due to changes in the isotopic composition of atmospheric CO_2 [44,67,68,70]. A sample of 1500-year-old sorghum analyzed by Schwarcz and White [59] yielded a $\delta^{13}C$ value of −9.7‰, which is considerably higher than that of modern millets. Following Schwarcz and White [59] and Tieszen and Fagre, who found a similar difference between modern and archaeological maize [68], we have added a 1.5‰ correction coefficient to all plant $\delta^{13}C$ values derived from the prehistoric samples to adjust for the observed shift over time in the isotopic composition of atmospheric CO_2 (see Table 6).

The isotopic differences between ruminant and non-ruminant animal bone samples from the Neolithic sites of northern China (Table 7, Fig. 3) conform to the patterns predicted by their digestive physiologies [3,25]. A relatively large carbon apatite–collagen difference for herbivores (mean $\Delta^{13}C_{ap-col} = 8.37 \pm 0.9‰$) is due to the ruminant digestion process [14,25,35,10,45]. A low average $\delta^{15}N$ value of 7.14‰ for the tested samples is consistent with the low trophic level of herbivorous cervids and bovids. High $\delta^{18}O_{pdb}$ values for these taxa (mean $= -5.00 \pm 1.3‰$) generally are found because ^{18}O-enriched water from leaves is an important moisture source for ruminants [36].

Table 4
Isotopic composition of human bone collagen and apatite carbonate from Yangshao sites

Site	ID#	Sex, age	Collagen		Apatite carbonate		$\Delta^{13}C‰_{ap-col}$	Repeat difference[a]		
			$\delta^{15}N‰$	$\delta^{13}C‰$	$\delta^{13}C‰$	$\delta^{18}O‰_{pdb}$		$\delta^{13}C‰_{col}$	$\delta^{15}N‰$	$\delta^{13}C‰_{ap}$
Banpo	04	F, 40–50	9.05	−15.00	−7.60	−7.84	7.40	0.16	0.19	0.14
Jiangzhai	03	F, 35–45	8.89	−9.93	−3.78	−9.64	6.15	0.34	0.23	0.04
Jiangzhai	02	M, adult	9.18	−11.21	−5.57	−8.90	5.64	0.53	0.41	
Jiangzhai	01	M, 45–55	9.12	−8.59	−3.85	−9.65	4.74	0.38	0.34	
Jiangzhai	08	F, adult	8.37	−9.60	−4.11	−9.98	5.71	0.04	0.43	0.54
Jiangzhai	06	F, adult	8.34	−10.68	−3.94	−9.18	6.74	0.12	0.42	0.07
Jiangzhai mean ± SD			8.78 ± 0.40	−10.00 ± 1.01	−4.25 ± 0.75	−9.47 ± 0.43	5.80 ± 0.74	0.28	0.37	0.22
Shijia	12	M, adult	7.31	−9.05	−3.62	−9.37	5.43	0.29	1.36	
Shijia	05	?, 6–8	7.73	−10.03	−3.77	−9.44	6.26	0.41	0.34	
Shijia	09	M, 60+	7.98	−10.38	−3.03	−9.46	7.35	0.59	0.35	
Shijia	10	M, adult	8.83	−10.51	−4.58	−9.43	5.93	0.84	0.37	
Shijia	11	F?, 12–15	7.91	−9.20	−3.35	−9.53	5.86	0.02	0.66	0.03
Shijia	07	F?, adult	8.21	−11.19	−4.59	−9.59	6.60	0.19	0.32	
Shijia	13	M, 60+	8.08	−10.21	−3.18	−9.42	7.03	0.11	0.40	
Shijia	15	F, 25–35	8.55	−10.15	−4.60	−9.43	5.55	0.04	0.76	
Shijia	16	M, 25–35	8.29	−9.50	−3.82	−9.37	5.68	0.14	0.40	
Shijia mean ± SD			8.10 ± 0.50	−10.02 ± 0.65	−3.84 ± 0.63	−9.45 ± 0.06	6.19 ± 0.67	0.29	0.55	

[a] Repeat difference are based on two analyses of the same specimen.

Table 5
Two-way analysis of variance of stable isotope ratios for Jiangzhai and Shijia human bone specimens[a]

Factors	Sex		Site		Site/sex interaction	
	F	p	F	p	F	p
$\delta^{15}N‰$	0.06	0.34	**7.61**	**0.02**	2.25	0.16
$\delta^{13}C‰_{col}$	0.15	0.70	0.01	0.99	0.00	0.94
$\delta^{13}C‰_{ap}$[b]	0.62	0.46	2.01	0.19	3.71	0.08
$\delta^{18}O‰_{pdb}$	1.97	0.19	0.02	0.87	0.50	0.49
$\Delta^{13}C‰_{ap-col}$[b]	0.63	0.44	1.69	0.22	4.45	0.06

[a] F values significant at $p < 0.05$; 1, 9 df are boldfaced.
[b] Variables that are analyzed with $\delta^{13}C‰_{col}$ as the covariate.

The collagen carbon isotope values obtained from ruminant samples ranged from −18.76‰ to −14.20‰ (average −16.33 ± 2.1‰), suggesting a predominantly C_3 plant based diet for these animals, although those values are not sufficiently negative to indicate an exclusively C_3 diet. The $\delta^{13}C_{col}$ values of prehistoric herbivores grazing on unforested and predominantly C_3 pastures typically average −21 to −19‰ [3,31]. Higher values were observed for herbivores that presumably had some admixture of C_4 plants in their feed, such as bovid and cervid bone from Çatalhöyük ($\delta^{13}C_{col}$ ranges from −19.1 to −18.1‰) [55]. Thus, our results show that neither wild cervids nor bovids in Neolithic China had a diet completely free of C_4 plants.

Two bovid specimens tested (ID# 18 and ID# 24) exhibited especially high $\delta^{13}C_{col}$ values (−15.11‰ and −14.20‰) (Fig. 3A). Both specimens were initially identified as the bones of domesticated water buffalo [37], but were later re-examined and reclassified on the basis of dental morphology as representing a wild species of *Bubalus* (Li Liu, personal communication). The high $\delta^{13}C_{col}$ values found for these animals suggest that there was a substantial proportion of C_4 grasses in the pastures favored by them. Perhaps they were attracted to the vegetation of disturbed habitats around human settlements, where they would have had access to wild *Setaria viridis*, a common road-side weed, or to fields of domesticated millet.

Pig and dog bone samples from the Yangshao site of Xipo and the Longshan site of Kangjia displayed very high $\delta^{13}C_{col}$ values (−7.7 ± 0.4 and −10.8 ± 2.7‰, respectively), similar to those found for Yangshao humans and typical for animals with a heavy reliance on C_4 plants. Results for monogastric animals (pigs and dogs) from Xipo (Yangshao) and Kangjia (Longshan) differed systematically (Table 7). Xipo pigs and dogs had a less negative $\delta^{13}C_{col}$, lower $\delta^{15}N$, a lower difference between apatite and collagen carbon, and more negative $\delta^{18}O_{pdb}$ values than those from Kangjia. Lower $\delta^{13}C_{col}$ and higher $\delta^{15}N$ demonstrate that Kangjia pigs and dogs utilized a smaller proportion of C_4 foods and subsisted at a higher trophic level than those from Xipo.

A one-way analysis of variance was used to test the significance of these Xipo–Kangjia differences (Table 8). Significant differences were observed between Xipo pigs and dogs as compared to conspecifics from Kangjia for nitrogen and apatite carbon stable isotope ratios, as well as for $\Delta^{13}C_{ap-col}$. Collagen carbon means were not significantly different, perhaps because of greater variation in $\delta^{13}C_{col}$ for the Kangjia samples (values ranging from −7.5 to −14.5‰).

In addition, very large differences in $\delta^{18}O_{pdb}$ ($F = 38.86$, $p = 0.0007$, 1 and 6 df) were observed between the samples of Xipo and Kangjia monogastric animals. Oxygen isotope ratios tend to parallel the isotopic composition of local meteoric water. Water becomes enriched with heavy oxygen in arid environments due to the preferential departure of ^{16}O with evaporation, evapotranspiration, and passive water losses through the nose and mouth [36,65]. Thus, results obtained for the monogastric animal samples from Kangjia suggest a water or temperature regime different from that at Xipo (Fig. 3B,C, see also Fig. 5).

Less negative $\delta^{18}O$ values for the Kangjia animal bone probably reflect relatively greater aridity in the local ecosystem. Global environmental changes associated with the end of the Holocene Climatic Optimum might be responsible for the large differences in oxygen isotope values between Yangshao and Longshan bone samples (see Fig. 5). The Kangjia site is chronologically later than the Xipo, Jiangzhai, and Shijia sites. Increased

Table 6
Isotopic composition of modern millet (*Setaria italica*)

Setaria italica	ID#	$\delta^{15}N‰$	$\delta^{13}C‰$	$\delta^{13}C‰$ corrected[a]
Seeds	30	2.41	−11.98	−10.48
Seeds	31	1.83	−11.72	−10.22
Seed	32	3.87	−11.55	−10.05
	32 dupl	3.29	−11.89	−10.39
Mean seeds ± SD		2.61 ± 0.89	−11.81 ± 0.15	−10.31
Stalks	33	−0.04	−11.78	−10.28
Leaves	34	1.82	−11.70	−10.2

[a] $\delta^{13}C‰$ values corrected for the atmospheric changes in carbon composition according to Marino and McElroy [44] and Tieszen and Fagre [68].

Table 7
Organic and inorganic isotopic composition of faunal remains from the Yangshao site of Xipo and the Longshan site of Kangjia

Site	KP#	Species	Collagen		Apatite carbonate		$\Delta^{13}C‰_{ap-col}$
			$\delta^{15}N‰$	$\delta^{13}C‰$	$\delta^{13}C‰$	$\delta^{18}O‰_{pdb}$	
Xipo	26	pig	7.49	−7.40	−3.61	−10.20	3.79
Xipo	27	pig	7.96	−7.65	−2.61	−9.46	5.04
Xipo	28	dog	6.91	−8.18	−3.89	−8.94	4.29
Xipo mean ± SD			7.45 ± 0.53	−7.74 ± 0.40	−3.37 ± 0.67	−9.53 ± 0.63	4.37 ± 0.63
Kangjia	19	pig	7.77	−11.53	−5.80	−7.06	5.73
Kangjia	21	pig	9.57	−11.76	−5.82	−7.71	5.94
Kangjia	22	pig	8.71	−7.53	−4.51	−6.35	3.02
Kangjia	20	dog	9.48	−8.97	−4.89	−6.80	4.08
Kangjia	23	dog	9.84	−14.53	−6.66	−7.21	7.87
Kangjia monogastric mean ± SD			9.07 ± 0.84	−10.86 ± 2.71	−5.54 ± 0.85	−7.02 ± 0.51	5.33 ± 1.86
Kangjia	24	*Bubalus*[a]	6.56	−14.20	−5.87	−5.69	8.34
Kangjia	25	dom. sheep	6.61	−18.76	−9.71	−3.98	9.05
Kangjia	17	deer	7.96	−17.25	−10.14	−3.87	7.11
Kangjia	18	*Bubalus*[a]	7.42	−15.11	−6.13	−6.47	8.98
Kangjia ruminants mean ± SD			7.14 ± 0.68	−16.33 ± 2.06	−7.96 ± 2.28	−5.00 ± 1.29	8.37 ± 0.90

[a] The bovine specimens were originally identified as domestic water buffalo [37]. However, Liu now attributes them to a wild species of the genus *Bubalus* (Liu, personal communication).

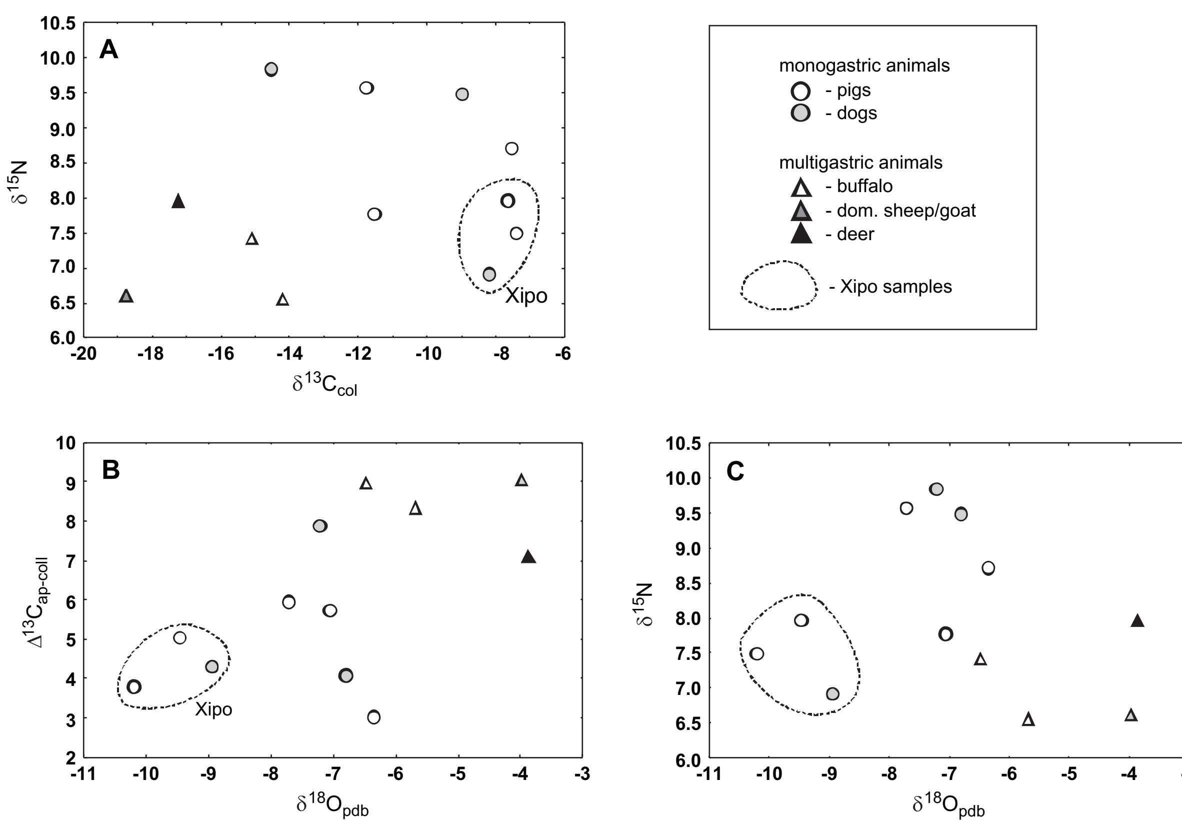

Fig. 3. Stable isotope composition of animal bones from the Neolithic sites of Xipo and Kangjia: (A) bone collagen carbon vs. nitrogen; (B) bone apatite oxygen vs. the difference between apatite carbon and collagen carbon; (C) bone apatite oxygen vs. collagen nitrogen.

Table 8
One-way ANOVA comparison of stable isotope values between Xipo (Yangshao) and Kangjia (Longshan) monogastric animals[a]

	Xipo vs. Kangjia	
	F	*p*
$\delta^{15}N‰$	**8.73**	**0.025**
$\delta^{13}C‰_{col}$	4.96	0.103
$\delta^{13}C‰_{ap}$[b]	**9.98**	**0.025**
$\delta^{18}O‰_{pdb}$	**38.86**	**<0.001**
$\Delta^{13}C‰_{ap-col}$[b]	**9.94**	**0.025**

[a] *F* values significant at $p < 0.05$, 1, 6 df are boldfaced.
[b] Marks variables that are analyzed with $\delta^{13}C‰_{col}$ as covariate.

aridity during that later time frame is indicated by a number of paleoclimatic studies [11,62,63].

4. Discussion

4.1. Food webs during the Neolithic in northern China

Several interesting observations can be made when the isotopic composition of the human and animal bone is compared. The data summarized in Fig. 4 show that humans buried at the Jiangzhai and Shijia sites and monogastric animals from Xipo and Kangjia shared a millet-rich diet. For pigs and dogs, we estimate that millet comprised close to 90% of their diet at Xipo and 65–85% at Kangjia. For Jiangzhai and Shijia, $\delta^{13}C_{col}$ values derived from the analysis of human bone allow us to estimate that millets comprised 75–85% of the human diet, a proportion that falls between the estimates derived for Xipo and Kangjia monogastric animals, respectively. These estimates are based on the corrected carbon isotope values for millets (Table 6) and wild herbivores (Table 7), assuming 5‰ positive fractionation between diet and collagen [3,6,35,56,66,70,71]. An additional correction of 1–2‰ for carnivore vs. prey collagen [12] is appropriate for meat-dominated diets.

Domesticated pigs and dogs commonly scavenge human refuse for food. Thus, their stable isotope profiles tend to imitate those of their owners [28]. However, small walled features recognized during the excavations at Xipo are interpreted as possible sties [43], indicating that pigs might have been kept in confinement there and foddered. These animals could have been raised for sacrifice in rituals and/or consumption at feasts and thus received special treatment, including a diet more rich in millet than that of their owners. On the other hand, it seems unlikely that dogs were being raised for food at that same time. Canids are represented almost exclusively by mature individuals and dog remains constitute only 1.3% of all animal bone recovered at Xipo, suggesting they were few in number.

The differences found between animal and human $\delta^{13}C$ values might be related to an overall lower reliance on millet agriculture in Shaanxi province, where population density was lower than in Henan, and where hunting apparently continued to be an important subsistence strategy throughout the Yangshao period. To some extent, human carbon might have been enriched with heavy stable isotopes, not only by direct consumption of millet, but also by the consumption of pigs, dogs, and/or chicken reared on millet and eggs laid by C_4 plant-fed chicken; the actual proportion of millet in the human diet could have been rather modest. Thus, the severe dental wear and masticatory stress, low incidence of caries, relatively tall stature, and low frequency of

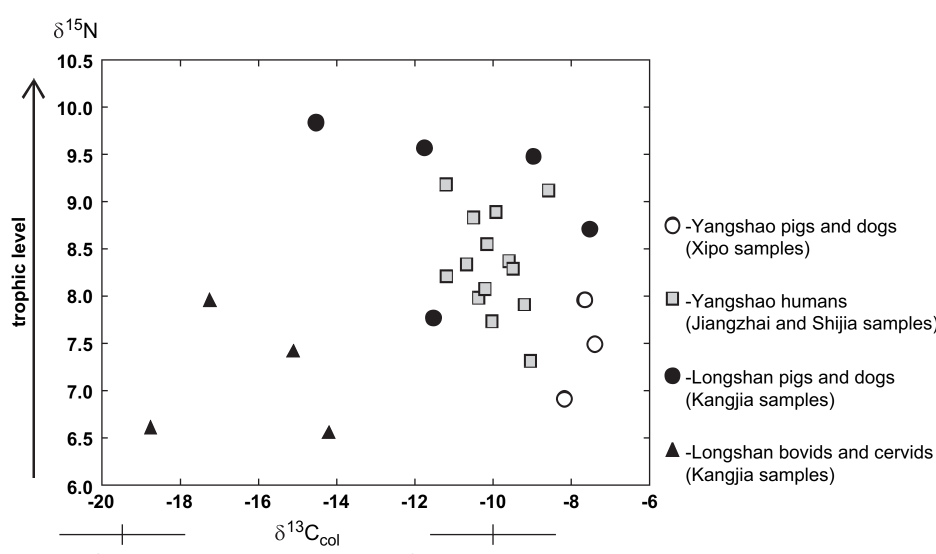

Fig. 4. Stable isotope composition of human and animal bone collagen samples obtained from Neolithic sites in the Wei and Yellow River valleys.

indicators of physiological deficiencies and stress previously reported [48] for humans buried at Shijia and Jiangzhai may be attributed to their reliance on a diverse subsistence regime that included plants and animals of high dietary value, both wild and domesticated.

Variation in the isotopic composition of the mineral bone fraction is presented in Fig. 5. As is typical for farmers subsisting at a low trophic level, Yangshao human bone samples generally exhibited a large difference in carbon isotope values ($\Delta^{13}C_{ap-col}$) between apatite and collagen. Mean $\Delta^{13}C_{ap-col}$ values of 5.8 ± 0.7 and $6.2 \pm 0.7‰$, respectively, were observed for the Jiangzhai and Shijia samples, consistent with a diet in which protein had a lower $\delta^{13}C$ value than that of the total diet.

The trophic level of Xipo monogastric animals, as estimated by $\delta^{15}N$, was lower than for contemporaneous humans and comparable to the bovids and cervids, suggesting a largely vegetarian diet for the Xipo pigs and dogs. The small difference between the $\delta^{15}N$ values derived for monogastric and multigastric animals is easily accounted for by variation in the isotopic composition of various parts of a millet plant. Assuming that ruminants ate mainly the leaves and stalks, while pigs and dogs were fed cereals, the $\delta^{15}N$ difference between Xipo monogastric animals and ruminants corresponds to the $\delta^{15}N$ difference between seeds, stalks and leaves. Measured $\delta^{15}N$ values for seeds and leaves average 2.6‰ and 1.8‰, respectively (Table 6). Thus, we expect an approximately 0.8‰ difference between $\delta^{15}N$ values for seed and leaf eaters. An observed 0.31‰ difference between Xipo monogastric animals and herbivores suggests that some stalks and leaves may have been added to pig fodder otherwise consisting mainly of cereals. Together, the very high $\delta^{13}C_{col}$ values, typical for a C_4 plant diet, and very low $\delta^{15}N$ values, typical for herbivores, suggest that millet, rather than household refuse, was the principal component of animal fodder at Xipo.

The diets of Kangjia and Xipo monogastric animals differed significantly. Some of these differences might have been a consequence of variable soil composition. However, the correspondence between carbon and nitrogen isotopic values implies that dietary shifts could at least in part be responsible for these differences. Kangjia pigs and dogs had lower $\delta^{13}C_{col}$ and thus ate less millet. They also had higher $\delta^{15}N$ and thus had a higher trophic level than the Xipo conspecifics. With the exception of one pig specimen (ID# 22), $\delta^{15}N$ values of Kangjia pigs and dogs are higher than those of Yangshao domesticated animals and humans (Fig. 4). The relatively high trophic level of Kangjia monogastric animals could have been due to increased reliance on household refuse and/or foraging on wild C_3 plant and animal resources outside the settlements.

In addition, the Kangjia site was densely populated and rodents were probably abundant there. Consequently, the hunting of rodents and other pests could have increased the trophic level of dogs. The very small difference between apatite carbonate and collagen $\delta^{13}C$ values in samples #20 and 22 ($\Delta^{13}C_{ap-col} = 4.08$ and 3.02‰, respectively) suggests that carbon isotope composition of the dietary protein source was heavier than that of the overall diet. Rodents feeding on millet could have supplied ^{13}C enriched protein, while household refuse with a lighter carbon isotope composition would still have been the main source of calories for Kangjia dogs and pigs.

Heavy carbon could have also come from the feces of millet-eating farmers. Terracotta images of pigsties combined with toilets have been recovered from Eastern Han (25–220 AD) tombs in Henan [46]. If this practice was already common during the Late Neolithic, the high trophic level of Kangjia pigs might be accounted for by

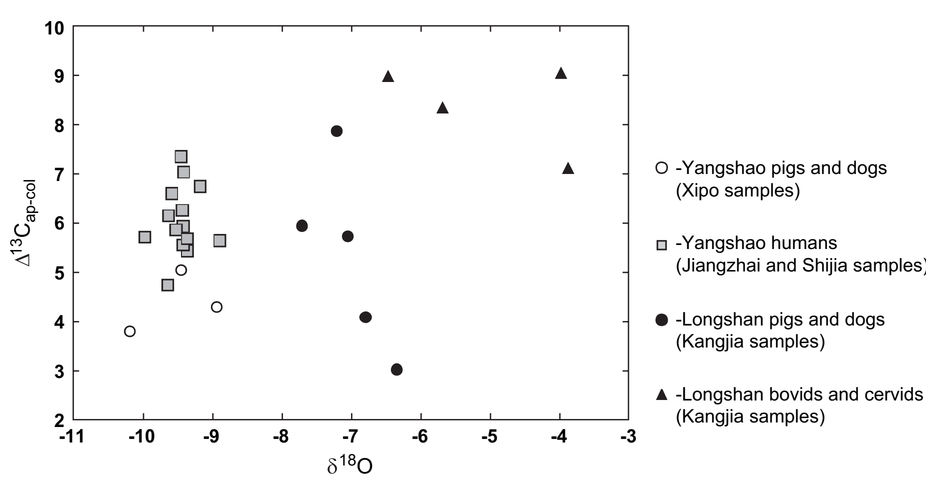

Fig. 5. Stable isotope composition of human and animal bone apatite samples obtained from Neolithic sites in the Wei and Yellow River valleys.

trophic level enrichment from the recycling of organic matter within the site.

4.2. Broader implications

Stable isotope analysis of human and animal bone from Neolithic China presents compelling evidence that millet was used in two ways: as a human dietary source and as fodder for monogastric animals. Consonant with our results, stable isotope analysis of pig bones from the Longshan site of Liangchengzhen in southeastern Shandong suggested a high proportion of millet in the feed of those domesticated animals [34]. Household refuse is usually assumed to be the primary source of nutrients for pigs [9]. It may seem counterintuitive that Neolithic farmers would dedicate a large proportion of their agricultural production to animal feed, especially at a densely populated site such as Xipo, since this would considerably reduce the available human caloric base. At the same time, one might wonder whether a Neolithic family growing millet would be able to generate enough refuse to support even a single pig, whose caloric requirements, pound for pound, are at least as high as those of a human.

A subsistence strategy dedicating a high proportion of agricultural production to the feeding of pigs is known ethnographically for the Tsembaga, a Maring-speaking group in upland New Guinea [49]. The Tsembaga could support only 0.24 pigs per person using food waste alone, yet actually tended about 0.83 pigs per person, by relegating a considerable quantity of their agricultural production to pig fodder. Pigs consumed 40.7% of all tubers harvested by the Tsembaga; only 15% of that was peel wastage not normally eaten by humans. The Tsembaga cultivated about as much land per pig (0.15 acres/pig) as they did per person [49:61].

A consequence of this practice for the Tsembaga is that it reduces the carrying capacity of their land and appears to restrict population growth [49:62], while also buffering against crop failures by redistributing agricultural products among humans and domesticated animals. Over 90% of calories consumed by pigs are lost through metabolic processes, so the large number of pigs supported results in a human population peaking below the theoretical carrying capacity of their territory.

The ethnographic and ethnohistoric records document a number of cases in which pigs were reared primarily for use in rituals, competitive feasts, and/or exchange, while also supplying proteins, fat, and calories for general dietary wellbeing [23,49,72,75]. A similar situation may have pertained for Yangshao. The probable symbolic importance of pigs during the Chinese Neolithic is indicated by a clay pig statue found at a Peiligang site [73] and by the association of pig bones with particularly wealthy burials of the Dawenkou and Longshan cultures in Shandong province [33].

A dynamic balance between two aspects of subsistence, pig husbandry and millet agriculture, would result in a very stable subsistence system. A large number of millet-fed pigs could be supported in years with plentiful harvests. In bad years, attended by crop failures or the depletion of wild resources, the human caloric base could be sustained through the sacrifice of pigs and dogs and re-directing millet for human consumption, thereby avoiding the trophic cost of producing pork.

If a tight mutualism was established between millet agriculture and animal husbandry in China during the Neolithic, we would expect these two aspects of human subsistence to develop in parallel. Indeed, several correspondences between pig and millet domestication can be found in the archaeological record of the Yellow and Wei River valleys. Millet agriculture in this area dates back at least to 8500–7000 BP [15,84]; the earliest experimentation with millet may have occurred as long ago as 12,000–10,000 BP [60,61]. Similarly, the earliest pig bones recovered in an archaeological context in northern China date back to 10,000 BP [88]. While these earliest pig bones likely belonged to wild animals, pigs and dogs became the focus of animal husbandry in China during the Early Neolithic, by about 8000 years ago [51,73,88,89].

Intensification of millet production during the Yangshao period coincided with increasing proportions of pig bones in faunal assemblages. At the Yangshao site of Xipo, pig remains comprised 84% of all recovered animal bone [42]. In the Yangshao strata of Bancun (6000–4500 BP), the frequency of pig bones was 80%, or 35% more than in the earlier Peiligang strata at the site. Sites in central Shaanxi were exceptional, given that hunting continued to be important throughout Yangshao times there, probably due to lower population density. For the earliest level at Jiangzhai (6860–6320 BP), pig bones comprised 41% of the faunal remains, but this proportion exhibited a consistent decline through the succeeding levels [81]. At the Zijing site in southern Shaanxi, the frequency of pig bones increased around middle Yangshao, from 13% for Zijing II (6900–5900 BP) to 31% for Zijing III (5480–4820 BP). Increased reliance on pig tending during the Yangshao period can be attributed to increased agricultural output due to improving technology and an environmental trend towards higher temperatures and humidity between 7000 and 5000 BP [50,62,63].

5. Conclusions

The complexity of millet-based food webs in the Yellow and Wei River basins during the Neolithic is revealed by stable isotope analysis of human and animal bone samples. The high $\delta^{13}C$ values for human bone that were observed might have resulted from direct consumption of millet and/or from eating millet-fed domesticated animals, including pigs, dogs, and chicken, as well as from eating the eggs of millet-fed chicken. High $\delta^{13}C$ values for pigs and dogs could have resulted from high proportions of millet in their feed, as in the case of Xipo animals, and/or from access to household refuse, human feces, and millet pests, i.e. mice and rats. Somewhat elevated $\delta^{13}C$ values observed for wild and domesticated bovids could be attributed to their grazing on unprotected millet fields, the presence of some wild C_4 plants around human habitats, i.e. *S. viridis*, and/or the addition of millet hay to the feed of domesticated herbivores. In other words, at different stages of the Neolithic Period: farmers ate millet; farmers ate pigs and dogs that were fed millet; dogs hunted mice feeding on millet; pigs ate feces containing semi-digested millet; and humans hunted wild herbivores that were attracted to millet fields. Notwithstanding the multiplicity of interpretations available, it is clear that during the Chinese Neolithic, millet was an important source of food, not only for humans, but also for their domesticated animals. Thus, the Neolithic farmers of what are now Shaanxi and Henan provinces relied heavily on millet in two ways: as a staple for direct human consumption and to support their domesticated animals.

Acknowledgements

We would like to acknowledge the support of grants from the Sigma Xi Foundation to E. Pechenkina, as well as the University of Missouri-Columbia Research Council and Research Board to R. Benfer. Support for mass spectrometry instrumentation was provided to S. Ambrose by National Science Foundation grant SBR 98-71480.

We express our gratitude to the staff at the Banpo Museum, especially the archaeologist Wang Zhijun, who were most helpful and professional in making our research time there productive. Professor Baozu Yu of the National Foreign Language University in Xi'an also assisted us in many ways, especially by providing us with competent translators who made our work possible. We greatly appreciate the help and comments of Li Liu of LaTrobe University. We thank Paula Tomczak, Marie Balasse, and Sallie Greenberg for assistance in sample preparation and analysis, as well as Arthur Rostoker and Warren DeBoer for valuable comments on earlier versions of this manuscript.

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