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Stable isotope and proteomic insights into Bronze age human dietary life history at Köhne Shahar, Northwest Iran

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ABSTRACT

Interest in subsistence strategies practiced by the Kura-Araxes communities in Southern Caucasus and the highlands of the Near East has a long history, yet direct studies of paleodiet at the scale of the individual are few. We apply serial sampling of carbon and nitrogen isotopes in 17 teeth representing 11 comingled individuals at the Kura-Araxes early Bronze Age site Köhne Shahar (KSH) in northwestern Iran. Proteomic analyses of dental enamel show seven females and four males. Isotopic results indicate an agro-pastoral diet with little or no C₄ millet. Individual isotopic biographies reveal a dietary life history that includes weaning between 1.5 and 3.7 years of age (average = 2.4 years), followed by a stable early childhood diet with little intra-individual variation through age 10 years. Isotopic shifts around 12–14 years of age suggest a change in diet that may correspond to marriage and the establishment of new household units focused more on plant foods. Gradual isotopic shifts between 14 and 20 years may represent such households developing livestock herds and increasing meat consumption. Stability in diet across this transition is consistent with village endogamy. Sex-linked differences in the age of weaning and childhood $\delta^{15}\text{N}$ values hint at differences in learning and enculturation practices. Males and about half of females were weaned earlier but had access to greater amounts of meat, suggesting they were more involved in animal husbandry and/or production of animal products (e.g., cheese, yoghurt) outside the house. By contrast, the other half of females were weaned later in childhood, but ate significantly more plant foods, suggesting they were more involved in tending gardens and producing crafts and/or plant-based foods within the house, where they had greater access to breastmilk.

1. Introduction

During the middle of the fourth millennium BCE, around the time early states and urban centers were developing in Mesopotamia (Spencer 2010), the Kura-Araxes cultural tradition developed in the South Caucasus (Batiuk et al., 2022). By the late 4th to early 3rd millennium BC, their characteristic material culture, particularly hand-made black burnished pottery, spread throughout much of Southwest Asia. Longevity that includes over a millennia of Kura-Araxes tradition, great expanse of geographical distribution spanning from the Mediterranean Ocean to the Caspian Sea, and the development of several regional cultural variants, make Kura-Araxes a critical component for understanding broader patterns in the evolution of human societies in southwest Asia (see Alizadeh et al., 2018; Batiuk et al., 2022; Palumbi

and Chataigner 2014; Rothman 2017; Sagona 2018; Samei et al., 2023).

Kura-Araxes sites are found in a wide range of locations, including fertile lowlands and seasonally inhospitable highlands. The small size of sites with ephemeral architectural remains have led many scholars to portray Kura-Araxes culture as small, egalitarian, mobile pastoralists or sedentary agriculturalists, lacking social inequality and hierarchy (Smith, 2005: 258; Kohl 2009: 250). However, Frangipane (2007) argues that the Kura-Araxes cultural communities represent horizontal egalitarianism since there is a lack of hierarchy in the size of Kura-Araxes sites and dominance of domestic functions. By contrast, Rothman (2020) argues that the sites represent vertical egalitarian societies, with leadership having influence, but not authority, and lacking social mechanisms for control (such as institutionalized violence). How subsistence practices fit into these issues is of great significance for

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understanding the spread and longevity of the Kura-Araxes culture.

Advances in stable isotope analyses over the last 20 years open new windows on studying paleodiet. A growing number of studies focus on sequential isotopic analysis of incrementally-growing tissues, especially teeth and hair, to examine life histories of people from the past (e.g., [Beaumont et al., 2015](#); [Burt, 2013](#); [Eerkens et al., 2011, 2020a](#); [Fuller et al., 2003](#); [Greenwald et al. 2016](#); [Knudson et al., 2016](#); [Sealy et al., 1995](#); [Webb et al., 2013](#); [White, 1993](#); [Wright 2013](#)). These isotopic biographies, or isobiographies, are significant in that they provide information about diet and mobility, and how those changed over months or years, at the level of the individual. By accumulating such individual-level isobiographies within a site, we can make comparisons of average behaviors across various social and biological dimensions, such as elite vs. non-elite or male vs. female.

In this study we document isobiographic dietary patterns for 11 individuals from a single comingled grave at the Bronze Age site of Köhne Shahar (KSH) in northwestern Iran. Documenting isobiographic dietary patterns at one of the major Kura-Araxes sites, we have three main goals. First, we aim to examine how diets at KSH, as measured by isotopes, compare to other sites in the broader Eurasian region, and in particular, whether any C_4 plant foods, such as millet, played a significant role in the diet of inhabitants. Second, we estimate the average age of weaning of individuals and early childhood diets, and whether there are any observable differences between males and females. Finally, we aim to examine the degree to which diets varied isotopically over the course of an individual's life, after weaning, and what such changes reveal about enculturation, marriage, and mobility.

2. Köhne Shahar Background

Köhne Shahar (KSH) is a ~15 ha (ha) settlement located 20 km to the northwest of the city of Chalدران in the Western Azerbaijan province in northwest Iran ([Fig. 1](#)). The site is found in a narrow valley between small plains and high pasturelands, providing access to both agricultural land and field grazing for domesticated animals. Geologically, the area consists of a basaltic lava field that may have been deposited by one or several volcanoes. Surrounded by two rivers, the Qizlar Chay and Beytal Chay, on two sides, the site is in a naturally defensible position with access to resources from multiple nearby ecosystems. In addition to the two rivers, several springs around the site form sources of water ([Alizadeh, 2015](#)).

KSH was first recorded by a German team in 1970s ([Kleiss and Kroll, 1979](#)), with the site re-recorded by an Iranian team during a more extensive survey in 2011. The latter survey highlighted the large physical extent of KSH and associated the site with the Kura-Araxes II and III culture-historical units based on the ceramic typology (ca. 3,300–2,800 BCE). [Alizadeh](#) suggests the site has not witnessed occupation other than Kura-Araxes Culture ([Alizadeh et al., 2015](#)). The site consists of a fortified citadel, a cemetery contemporaneous with the citadel, and an extramural residential area or “outer town”. The citadel, with an area of 2.5 to 3 ha., sits on top of a natural basaltic platform at an altitude of 1905 m a.s.l., located 20 m above the valley floor.

Excavation by the Iranian team included three field seasons (2012–2014) and nine trenches: two in the cemetery, one in the extramural area, and six in the settlement. The test trench (TT1) opened near the fortification wall inside citadel was used to diagnose the chronological sequence of this occupation ([Alizadeh et al., 2015](#)). The archaeological findings from KSH include architectural structures, as well as tools representing a wide range of subsistence and craft activities, such as mortars, pestles, grinding stones, spindle whorls, spools, loom weights, beads, and bone tools. In addition, the presence of storage jars suggests harvesting cereal grains and other agricultural activities.

The cemetery of KSH is located in the northwest of the settlement, about 300 m from the citadel. Although Kura-Araxes funerary practices vary in their associated ritual practices, grave design, and architecture (see e.g. [Poulmarc'h et al. 2014](#)), a common theme is concentration of

burials near a settlement ([Batiuk et al. 2022: 268-271](#)). This is true of KSH, where inhabitants buried the dead in a cemetery area separate from residences (see [Fig. 1](#)). During the 2013 field season, two graves in the cemetery area in trenches 5B20 and 5C7 were discovered and excavated. The grave in 5C7 did not provide significant information since it was looted in the past. However, 5B20 provided important information that comprises the materials in this study. This grave is a secondary exposure that contains numerous individuals that appear to have accumulated over time. Grave goods within the tomb were minimal, comprising only a few Kura-Araxes pottery sherds and beads, and lacked evidence of conspicuous materials. The grave itself is rectangular and stone-lined, 2.2 m long, 1.2 m wide, and 1 m deep, and includes comingled remains ([Alizadeh, 2015](#)). The walls are made of a layer of small basalt rocks laid horizontally, while the floor was covered by small pebbles. Analysis of the human skeletal remains indicates a minimum of 15 individuals, including 13 adults and two sub-adults ([Asgari 2018](#)).

For this study, 17 teeth representing 11 individuals were selected for stable isotope analyses. Because most of the teeth are disassociated from pelves or other sexually diagnostic elements, we also analyzed enamel from each tooth for proteomics to estimate sex for each individual (see [Parker et al., 2019](#)). Finally, collagen from four teeth was submitted for radiocarbon dating.

3. Stable isotope methods

Human collagen is preferentially synthesized from ingested protein ([Ambrose and Norr, 1993](#); [Fernandes et al., 2012](#); [Kellner and Schoeninger, 2007](#); [Tieszen and Fagre, 1993](#)). Carbon isotopes ($^{13}C/^{12}C$, expressed as $\delta^{13}C$ relative to an international standard) in bone or tooth collagen provide an estimate of the source of dietary protein for that person while those tissues were forming. In most terrestrial ecosystems carbon isotopes are used to trace the importance of C_3 vs. C_4 plants in the diet. Most economically-important plants around the world, including wheat and barley, are C_3 plants that discriminate against the heavier ^{13}C during photosynthesis, resulting in $\delta^{13}C$ values between -30‰ and -22‰ ([Ehleringer et al., 1993](#); [Farquhar et al., 1989](#)). By contrast, C_4 photosynthesis produces tissues with $\delta^{13}C$ values typically between -16‰ and -10‰ . While there are less C_4 photosynthesizers globally, several important crop plants such as millet and sorghum fall in this category ([Cerling et al., 1998](#); [Ehleringer et al., 1991](#); [Tippie and Pagani, 2007](#)). Millet, in particular, is known to have spread across western Asia and eastern Europe during the Bronze Age, and could have played a role in KSH diet ([Martin et al. 2021](#); [Pospieszny et al., 2021](#); [Trifonov et al., 2017](#)). Freshwater aquatic foods can fall between these extremes, but as noted below should be distinctive in their nitrogen isotopic composition (due to increases in trophic level). Trophic-level effects (discussed below) on $\delta^{13}C$ are relatively small, on the order of 0.5–1.0‰ with each level ([Fuller et al., 2006](#)). Thus, in this study, we use $\delta^{13}C$ mainly to estimate the importance of wheat and barley, or C_3 plants, vs. sorghum and millets, or C_4 plants, in the diet.

Nitrogen isotopes ($^{15}N/^{14}N$, expressed as $\delta^{15}N$ relative to an international standard) reflect the general trophic level of consumed foods ([Minagawa and Wada, 1984](#); [Schoeninger, 1985](#)). In organisms, nitrogen fractionates during the synthesis of biological tissues, favoring the retention of the heavier ^{15}N (and excretion of the lighter ^{14}N). As a result, $\delta^{15}N$ increases by about 2–4‰ with each trophic level in an ecosystem with a similar basal source of nitrogen. In most terrestrial systems, there are essentially three trophic levels, plants, herbivores, and carnivores. By contrast, in aquatic environments there are more trophic levels, resulting in greater enrichment of ^{15}N at the top of the food chain (e.g., large fish). Some complicating issues include that basal $\delta^{15}N$ values are often elevated in arid regions ([Schwarcz et al., 1999](#)). As well, fertilizing agricultural fields with nitrogen-rich materials (e.g., fish meal, guano) can shift the basal $\delta^{15}N$ signature in an ecosystem ([Bogaard et al., 2007](#); [Fraser et al., 2011](#)), particularly in cereals. Thus,

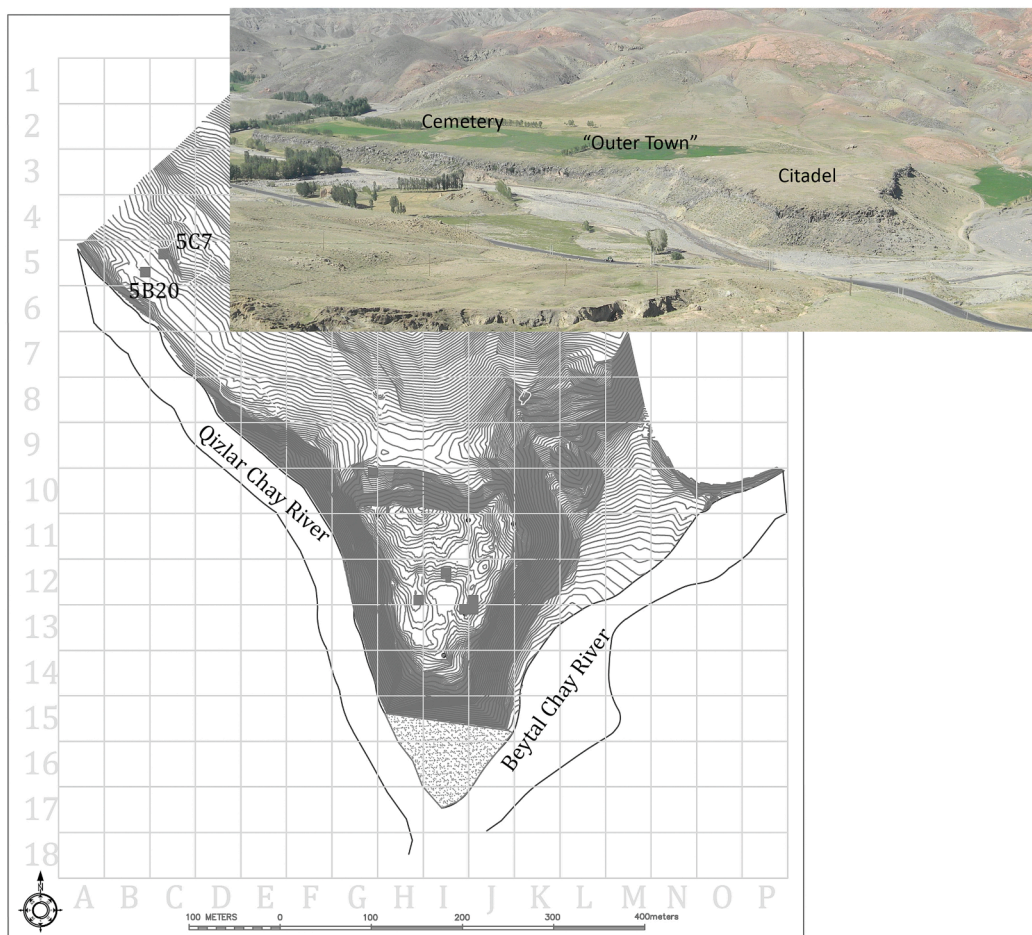


Fig. 1. Köhne Shahar location and map.

vegetarians can display significantly higher $\delta^{15}\text{N}$ values than we would otherwise expect if they are consuming agricultural products from fields with large inputs of fertilizer enriched in ^{15}N (Poulson et al., 2013; Szpak et al., 2012).

Breastfeeding involves a shift in trophic level relative to the mother. Breastmilk is synthesized by a lactating female and includes protein that is one trophic level above her diet, resulting in an increase in $\delta^{15}\text{N}$ for the breastfeeding child (Fuller et al., 2006). As a child is weaned and transitions to solid foods, $\delta^{15}\text{N}$ should decrease by 2–4 ‰ regardless of the basal source of nitrogen. In this study, we use $\delta^{15}\text{N}$ in early-growing teeth, especially permanent first molars, to trace weaning patterns and the trophic level of dietary protein in children. With weaning, $\delta^{13}\text{C}$ often follows a similar, though less marked decrease (approximately 0.5–1 ‰ decrease during weaning), and can be used as a second marker of the weaning process. We estimate the age of weaning in permanent first molars by examining the age-controlled isotopic curve, and finding the point where $\delta^{15}\text{N}$ (and typically also $\delta^{13}\text{C}$) reaches a local minimum in the first set of serial samples. Because weaning was complete by this age (i.e., $\delta^{15}\text{N}$ no longer decreases after this age), we average the age estimate of this point and the one before it (where weaning was still ongoing), to estimate the approximate median age of weaning.

We note that other physiological factors, such as starvation and catabolism of muscle, can also have effects on $\delta^{15}\text{N}$ (Beaumont and Montgomery 2016; Fuller et al., 2006; Neuberger et al., 2013). However, as our isotopic samples average diet over periods of 0.5 to 2.0 years, and starvation events are typically much shorter than this, we believe such effects are unlikely to have a major impact on our interpretations.

We focus on the analysis of dentinal collagen. For each tooth, we photographed and then cut the molar in half longitudinally to isolate a complete crown-to-root section. During tooth growth, dentin accumulates in an accretionary fashion from the Dentin-Enamel Junction (DEJ) to the Apical Root Tip (ART) over time. For a permanent first molar, this process begins at about age 0 years at the DEJ, passes the Cementum-Enamel Junction (CEJ) around age 2.8 years, and continues to the ART until the tooth is finished growing at around age 9.5 years (Hillson, 1996). For a permanent third molar, formation begins at the DEJ between 7 and 10 years of age, the CEJ forms between 12 and 16 years, and the ART between ages 18 to 25 years. We use these landmarks and ages to calibrate ontogenetic time within a tooth, and to assign serial samples to median ages.

Layers of dentin accumulate in planes approximately parallel to the occlusal surface within the tooth crown. Each layer takes several weeks to form. Within the root, these parallel layers start to angle, and in three dimensions appear as cones of dentin with truncated tops. This latter point is significant as our tooth sectioning methods does not facilitate cutting cones of material. Instead, we cut horizontal sections that cross growth planes within the root, resulting in time-averaging (Eerkens et al., 2011). We estimate, that non-adjacent serial samples in our study include only minor temporal overlap (i.e., are nearly independent in the ontogenetic time they represent). Note further that teeth with significant occlusal wear (below the DEJ) will be missing the earlier growth. In several cases of first molars in this study, occlusal wear removed two or more years of growth, likely obliterating much of the weaning signal. Likewise, some teeth were missing part of the ART, either due to damage or because the individual died before the tooth finished growing, resulting in loss of the latest-forming dentin within the tooth.

We removed all enamel and cementum from the exterior using a rotary hand-held drill and reamed out the pulp chamber to remove secondary dentin. The tooth half was then demineralized in a solution of 0.5M hydrochloric acid (HCl) in a refrigerator set at 5°C. HCl was changed every other day until the sample was completely demineralized (generally 1–2 weeks). Following demineralization, any secondary dentin that was not reamed out during drilling was also removed (secondary dentin typically separates from the primary dentin after demineralization). The tooth was then rinsed with dH_2O , and sliced into parallel serial sections, each 1 mm thick, beginning at the apical root tip

and working up towards the crown. Following slicing, each serial sample was placed in a separate vial and immersed in 125 M NaOH (sodium hydroxide) for 24 h to remove humic acids. The sample was rinsed with dH_2O to remove any residual NaOH and placed in slightly acidic pH3 water in an oven set to 70 °C to solubilize collagen. Solubilized collagen was then freeze-dried to remove all remaining water, isolating the collagen fraction.

Approximately 1 mg of collagen was weighed out from each serial section for stable isotope analysis. In some cases, there was not enough collagen from a serial sample, and adjacent section(s) had to be combined to achieve a total of 1 mg. Carbon ($^{13}\text{C}/^{12}\text{C}$) and nitrogen ($^{15}\text{N}/^{14}\text{N}$) for each serial sample was measured by continuous-flow mass spectrometry (PDZ Europa ANCA-GSL elemental analyzer interfaced to a PDZ Europa 20–20 isotope ratio mass spectrometer) at the Stable Isotope Facility, University of California Davis. Carbon isotope ratios, $\delta^{13}\text{C}$, are reported expressed in permil notation (parts per thousand) relative to the PeeDee Belemnite standard (arbitrarily set at 0 ‰), while N isotope ratios, $\delta^{15}\text{N}$, are expressed against N_2 in modern atmospheric air (also arbitrarily set to 0 ‰). Instrument precision is less than 0.2 ‰ for both isotope values. Analyses also produce an atomic C/N ratio, reflecting sample quality. Previous studies suggest that ancient collagen samples with C/N values between 2.9 and 3.6 produce accurate isotopic values (DeNiro, 1985).

4. Proteomic methods

Due to the fragmentary nature of the burials, we were only able to estimate sex for one individual using traditional osteological markers (Individual 161, a male). To augment sex estimation, we used proteomic methods on a single tooth from each individual (including the one identified as male). For one individual (Individual 256), we analyzed two different teeth to verify consistency of the method.

Amelogenin genes are located on both the X and Y sex chromosomes in humans and play a major role in the biosynthesis of enamel (Kwak et al. 2016; Mazumder et al. 2014; Prajapati et al. 2016). The different chromosomes, however, code for distinctive isoforms of amelogenin proteins, AMELX and AMELY (Garvin et al. 2012; Madel et al. 2016), that can be preserved in enamel over archaeological time scales (Parker et al. 2019; Stewart et al. 2017). Fragments of these proteins can be detected using shotgun liquid chromatography mass spectrometry and assigned to the AMELX or AMELY isoform.

We follow methods established in Parker and colleagues, (2019; see also Buonasera et al., 2020). In short, powdered enamel samples (19–20 mg) were demineralized, treated with trypsin (1 μL of 0.5 $\mu\text{g}/\mu\text{L}$, Thermo Pierce Inc.), incubated at room temperature for approximately 20 h, and passed over solid phase extraction to remove organic contaminants. Digested peptides were then desalted and concentrated, and applied to liquid chromatography-tandem mass spectrometry (LC-MS/MS) on a Thermo Scientific Exploris Orbitrap Mass spectrometer in conjunction with a Proxeon Easy-nLC II HPLC (Thermo Scientific) and Proxeon nanospray source.

Mass spectrometry datasets (RAW format) were processed using PEAKS™ (version X) peptide matching software (Bioinformatics Solutions Inc., Waterloo, ON). The FASTA formatted UNIPROT *Homo sapiens* reference protein database (<https://www.uniprot.org/proteomes/UP000005640>) was modified to include additional FASTA protein entries of peptide sequences from all splice variants associated with AMELX_HUMAN (Q99217-1, -2, -3) and AMELY_HUMAN (Q99218-1, -2) proteins gene products (Salido et al. 1992; Simmer 1995). The reference database was further modified to incorporate a decoy database and was validated in PEAKS™ software (Zhang et al. 2012). Peptide-matching spectral assignment was conducted using default conditions with the following exceptions: error tolerance, precursor mass, 15 ppm, Fragment ion, 0.5 Dalton (molecular mass), cleavage with trypsin with up to two missed cleavages, and up to two non-specific cleavages. The algorithm assumed all cysteines were carbamidomethylated, and the peptide

was partially modified by deamidation (NQ), oxidation (MHW), pyroglutamate conversion from glutamate and glutamine, and methionine dioxidation. All peptide assignments were filtered by a one-percent false discovery rate. Each peptide was quantified by summing the intensity of each signal for the peptide-specific primary precursor mass over charge ratio (m/z). Signals from all peptides specific to either AMELY_HUMAN or AMELX_HUMAN were combined into a single metric (combined intensities [CI] per milligram enamel) for each protein. Analyses were conducted using R, version 3.4.4 (R Core Team 2018). Firth's (1993) bias-reduced logistic regression was implemented using the R package brglm, version 0.6.1 (Kosmidis 2017). We estimated probability of female sex for samples with no AMELY_HUMAN signal using a linear logistic regression (see Buonasera et al., 2020), where $PrF < 0.5$ is considered indeterminate.

5. Samples

Seventeen teeth representing 11 individuals were included in this study. Only one individual could be assigned sex based on osteological indicators (Buikstra and Ubelaker 1994). Based on the state of tooth development and eruption (e.g., incomplete roots, degree of occlusal wear; Hillson 1996), five individuals represent the remains of adults, four appear to have died as older adolescents or young adults, and two appear to be children under the age of 11 years (Table 1).

In addition to the human teeth, two small pieces of alveolar bone adhering to human teeth, and 21 faunal bones from the site (ten bovinds, nine caprids, one deer, and one lagomorph), were also analyzed for C and N isotopes. Table 1 presents the human sample dataset, and analyses performed by individual.

6. Results

Proteomic analyses indicate that of the 11 individuals analyzed, four were male and seven were female. We were able to verify the osteological estimate of Individual 161 as male. Further, the two teeth from Individual 256 both revealed the presence of AMELY_HUMAN, consistent in the indication that this individual was male. Table 2 shows the results from the proteomic analyses.

AMS dates for four collagen samples produced nearly identical age ranges, all overlapping in their two-sigma ranges (Table 3). The dates fall within the early part of the third millennium BC, a period that Kushnareva (1997) and Munchaev (1975) refer to as the Early Bronze Age for the region.

All the stable isotope data can be found in the supplementary data associated with this article. Overall, collagen was well-preserved in KSH

Table 1 Human samples included in this study from KSH.

Indiv. #	Age	Osteo. Sex	Elements Analyzed	# Prot. Samples	# Isotopic Samples	AMS Date
161	Adult	M	LLM1	1	12	
163	Adult	Ind	LLM1, LLM3, Bone	1	25	1
165	Adult	Ind	URM1, LRM3, Bone	1	21	1
167	Adult	Ind	URM1	1	12	1
168	~16	Ind	URM2	1	10	
248	Adult	Ind	LLM1, LRM3	1	19	
255	16-18	Ind	URM1, URM3	1	17	1
256	15-16	Ind	UM1, URM3	2	2	
258A	4-6	Ind	LLdM2, LLdM2	1	15	
258B	9-11	Ind	LRP3	1	6	
258C	10-15	Ind	LRM1	1	11	

Notes: Indiv. = Individual; Osteo. Sex = Osteological sex estimate; Prot. = Proteomic.

Table 2 Results from proteomic analyses of enamel.

Individual #	Age	Tooth Analyzed	Sample Size	Probability Female	Sex Estimate
161	Adult	LLM1	20.1	0	M
163	Adult	LLM3	20.1	0	M
165	Adult	LLM3	19.6	96.1	F
167	Adult	URM1	20.7	95.8	F
168	~16	URM2	20.4	98.9	F
248	Adult	LRM3	20.3	0	M
255	15-18	URM1	20.1	96.3	F
256	14-16	UM1	20.1	0	M
		URM3	20.7	0	M
258A	4-6	LLdM2	20.3	97.2	F
258B	9-11	LRP3	22.5	93.8	F
258C	10-15	LRM1	22.1	96.7	F

Table 3 AMS dates and calibrated age ranges.

Individual #	Lab Code	AMS date BP	Calibrated 2-sigma Age Range BC (median date)
163	D-AMS 033,457	4159 ± 32	2630-2880 (2754)
165	D-AMS 033,458	4169 ± 31	2630-2880 (2764)
167	D-AMS 033,459	4196 ± 39	2640-2900 (2779)
255	D-AMS 033,460	4130 ± 32	2580-2870 (2724)

samples. C/N ratios vary between 3.0 and 3.4 for all human samples (averaging 3.2), within the range recommended by DeNiro (1985) for well-preserved samples. Likewise, %C varied between 23 % and 46 % (average = 36 %), while %N varied between 8 % and 17 % (average = 13 %). These values too, are in keeping with recommendations by Ambrose (1990, 1993) and Van Klinken (1999; see also Sealy et al., 2014).

Stable isotope data reveal a relatively uniform dietary profile for individuals from KSH, with low inter-individual variation. Tooth serial samples vary in $\delta^{13}C$ between -19.1 ‰ and -16.9 ‰ (with an average of -18.7 ‰), while $\delta^{15}N$ varies between 10.0 ‰ and 14.1 ‰ (with an average of 11.2 ‰). The most notable outliers within the dataset are from early-growing serial samples that correspond to periods of time that precede weaning.

From the serial samples, we were able to construct partial dietary life histories for most of the individuals. Fig. 2 shows an example of an isobiography for individual 255, a female. This person died after the first molar had completed growing, after the crown of the third molar had fully formed, but before the root of the third molar completed. Based on

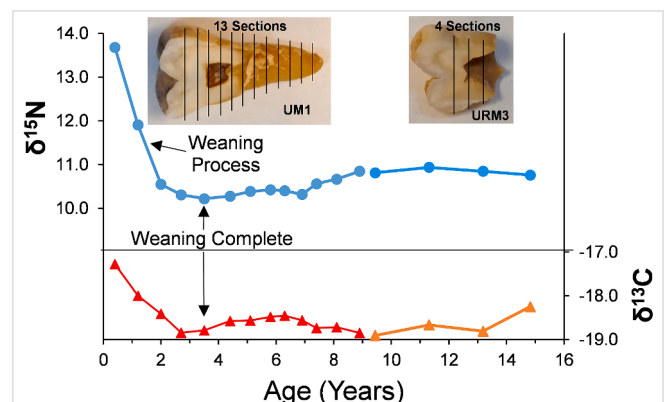


Fig. 2. Serial $\delta^{13}C$ and $\delta^{15}N$ samples from UM1 and URM3 of Individual 255.

the stage of growth, this individual likely died around age 15–18 years. Cross sections of first and third molar are shown above the isotopic curves. Superimposed over the cross-sections, we show the serial sampling strategy with vertical black lines showing where the tooth was cut. As can be seen, the fourth serial section of the upper M1 straddles the CEJ, placing its median age at about 2.7 years. The blue lines in the upper part of the figure show the dietary curve for $\delta^{15}\text{N}$, plotted using the left vertical axis. As shown, $\delta^{15}\text{N}$ starts at a high of around 13.8 ‰ in the crown of the first molar, and then drops quickly across the next two serial samples to around 10.6 ‰, representing what is expected of a full trophic level drop in dietary protein, as would be expected of weaning. For this female, the amount of breastmilk decreased rapidly between ages 0 and 2 years. Between ages 2 and 3.5 years $\delta^{15}\text{N}$ continued to decrease but at a much slower rate. The minimum $\delta^{15}\text{N}$ is associated with the serial section that has a median age of 3.5 years. Weaning must have taken place prior to the formation of this section. Across these same samples, $\delta^{13}\text{C}$ also decreases, and reaches a minimum in the section with a median age of 2.7 years. Based on the combined $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ curves, we estimate this individual was weaned sometime between 2.7 and 3.5 years of age, or a median age of 3.1 years. After being weaned, individual 255 had a steady diet, with little subsequent evidence for major isotopic shifts between ages 3.5 and 15 years when she died.

Fig. 3 shows the isobiographic data for Individual 163, a male. The permanent first molar is slightly worn, removing some of the earliest-forming dentin. However, the remaining crown sections show a decrease in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ consistent with weaning. In this case, we estimate the individual was weaned between 1.6 and 2.2 years of age, or a median of 1.9 years. After weaning, $\delta^{13}\text{C}$ remained stable through age 20 years, while $\delta^{15}\text{N}$ shows some fluctuations, including a slight increase around age 6 years, and a notable dip around age 13–14 years. The cause of these later fluctuations in $\delta^{15}\text{N}$ are not clear given this information alone, but are likely related to dietary changes associated with availability of different foods (e.g., increases in meat) that could be associated with changes in social status or residential shifts (e.g., post-marital residence shifts that led to increased or decreased intake of animal-based protein).

By contrast, Fig. 4 shows serial isotopic samples from an upper right second molar (URM2) from individual 168, a female. Because the earliest dentinal tissues began forming around age 2.5–3 years, we expect that most or all of the weaning process was likely to have happened before this tooth started forming. Indeed, Fig. 4 shows very little change in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ across the 10 serial samples, indicating that this individual had an isotopically stable diet during childhood and early teenage years.

Using the serial samples, Table 4 provides our age of weaning estimates, the isotopic composition of foods before weaning (i.e., pre-weaning, which is generally the earliest-forming $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ maxima), isotopic estimates at the termination of weaning (i.e., weaning

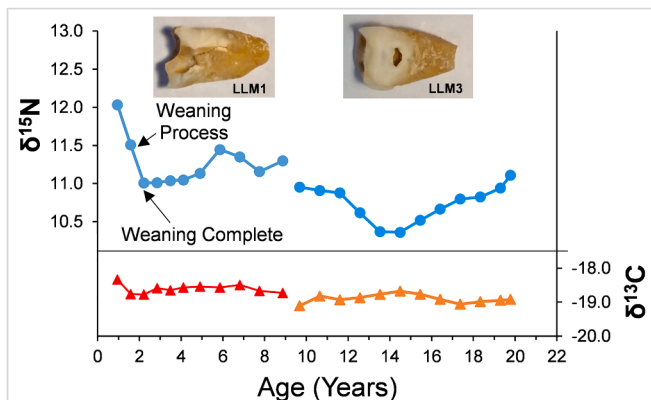


Fig. 3. Isobiography for Individual #163.

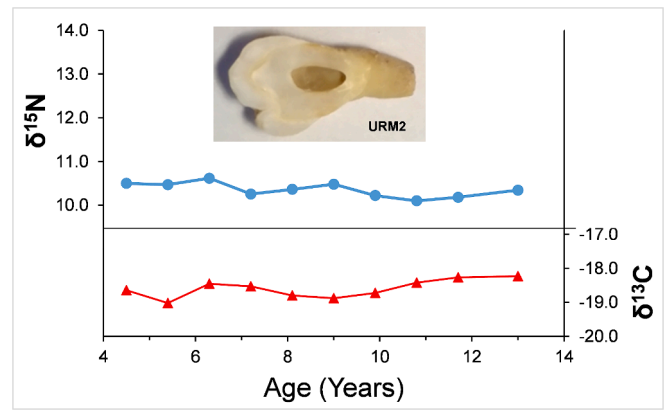


Fig. 4. URM2 from Individual 168, showing very little change in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ across the serial samples. Note incomplete apical root tip.

food), along with isotopic estimates for each individual at different age windows: 5–7 years, 7–9 years, 9–15 years, and 15–21 years. We were able to generate weaning age estimates for eight KSH individuals. These estimates range between 1.9 and 3.7 years, with an average of 2.4 years (and standard deviation of 0.8 years). We note that of the eight, the three males tend to have younger ages of weaning (2.2, 1.9, and 1.9 years; average = 2.0 years) than the five females (1.5, 1.9, 3.0, 3.1, and 3.7 years; average = 2.6 years).

Fig. 5 shows all serial samples with age-estimates for $\delta^{13}\text{C}$ (bottom) and $\delta^{15}\text{N}$ (top). Males values are represented by triangles and females by circles. Although there is variation within sexes, the figure shows that male and female isotopic values largely fluctuate in concert. This suggests that sex did not play a significant role in the isotopic composition of diet. Each half of the graph also shows a polynomial regression through all the data points, which represents something of “average” dietary life history for individuals from KSH for nitrogen (top) and carbon (bottom).

Table 5 presents the stable isotope results for KSH faunal remains. Again, C/N, %C and %N values are consistent with well-preserved bone collagen.

Fig. 6 plots the post-weaning $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values for humans, coded by sex, as well as the values for faunal remains. As shown, humans are systematically 2–6 ‰ higher in $\delta^{15}\text{N}$ than bovids, caprids, and deer, with no overlap between humans and these fauna. Humans are also 0.5 ‰ higher in $\delta^{13}\text{C}$, on average, though there is greater overlap between the human and faunal $\delta^{13}\text{C}$. Interestingly, the single lagomorph sample is elevated in $\delta^{15}\text{N}$, similar to the humans, though higher than all other samples in $\delta^{13}\text{C}$. As in Fig. 5, the graph also shows that males and females largely overlap for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$.

7. Discussion

Although subsistence practices of Kura-Araxes communities has been a major question for decades, knowledge is still limited (Sagona 2018: 273–279). Recent studies (see Herrscher et al., 2021; Samei et al. 2023) shed some light, but detailed studies of faunal and palaeobotanical remains are minimal. Instead, a range of proxy evidence such as the small size of most sites, the ephemeral nature of architecture, and the presence of sites in both fertile lowlands and seasonally-inhospitable highlands, have been used to portray Kura-Araxes as small and egalitarian communities of mobile pastoralists or semi-sedentary agriculturalists (Alizadeh et al. 2018).

To contextualize the KSH isotopic data, Fig. 7 plots $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ data from KSH against a sample of other Mesolithic and Bronze Age populations in Eurasia. Of course, we cannot know all of the baseline isotopic values in these vastly different environments. However, the goal here is simply to capture a wide range of different subsistence strategies

Table 4

Age of weaning estimates, and average dietary isotopic values at different life history stages.

Indiv.#	Sex	Age of Wean	Pre-wean		Wean Food		5-7 Years		7-9 Years		9-12 Years		13-21 Years	
			$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$
161	M	2.2	-18.5	12.9	-18.8	10.5	-18.8	10.8	n/a	n/a	n/a	n/a	n/a	n/a
163	M	1.9	-18.3	12.0	-18.8	11.0	-18.5	11.3	-18.7	11.2	-18.9	10.8	-18.9	10.7
165	F	3.0	-18.1	12.8	-18.4	11.4	-18.3	11.7	-18.4	11.6	-19.0	11.1	-18.8	10.7
167	F	3.7	-16.9	14.1	-18.9	10.0	-18.7	10.2	-18.8	10.4	n/a	n/a	n/a	n/a
168	F	n/a	n/a	n/a	n/a	n/a	-18.7	10.5	-18.7	10.4	-18.5	10.2	-18.2	10.3
248	M	1.9	-18.8	13.5	-18.8	11.3	-18.6	11.5	-18.8	11.8	-19.0	10.6	-18.8	10.7
255	F	3.1	-17.3	13.7	-18.8	10.3	-18.5	10.4	-18.7	10.6	-18.8	10.9	-18.5	10.8
256	M	n/a	n/a	n/a	n/a	n/a	-18.4	11.8	n/a	n/a	-18.7	10.6	n/a	n/a
258A	F	1.9	-18.5	13.8	-18.3	11.3	n/a	n/a	n/a	n/a	n/a	n/a	n/a	n/a
258B	F	n/a	n/a	n/a	n/a	n/a	-18.7	11.6	-18.7	11.7	n/a	n/a	n/a	n/a
258C	F	1.5	-18.7	13.9	-18.8	11.3	-18.7	11.3	-18.6	11.2	n/a	n/a	n/a	n/a
Avg.	2.4	-18.1	13.3	-18.7	10.9	-18.6	11.1	-18.7	11.1	-18.8	10.7	-18.7	10.7	
Male	2.0	-18.5	12.8	-18.8	10.9	-18.6	11.4	-18.7	11.5	-18.9	10.7	-18.9	10.7	
Fem.	2.6	-17.9	13.7	-18.7	10.9	-18.6	11.0	-18.6	11.0	-18.7	10.7	-18.5	10.6	

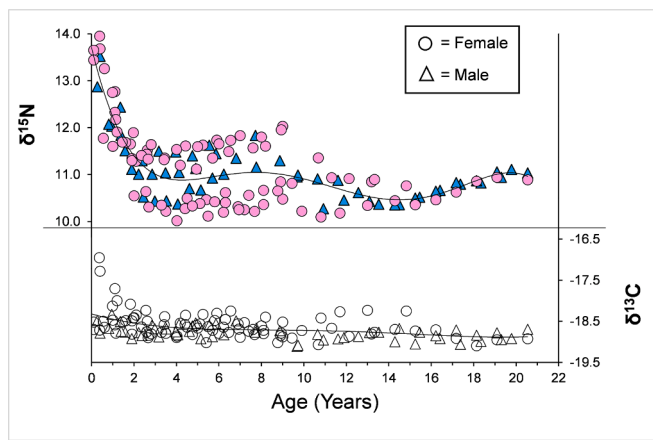


Fig. 5. All KSH serial sample data with age-specific estimates, $\delta^{13}\text{C}$ (bottom) and $\delta^{15}\text{N}$ (top), circles represent females and triangles males.

Table 5

$\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ for faunal remains at KSH.

Family	Catalog #	$\delta^{13}\text{C}$	$\delta^{15}\text{N}$	C/N	%C	%N
Caprid	1403	-18.8	8.3	3.2	40 %	14 %
Caprid	14 1380	-19.4	7.5	3.3	43 %	15 %
Caprid	1084 bC100	-18.9	6.6	3.4	39 %	14 %
Caprid	1131	-19.5	5.8	3.4	50 %	17 %
Caprid	1144 bC10	-18.5	7.0	3.4	55 %	19 %
Caprid	1221	-19.7	7.8	3.2	36 %	13 %
Caprid	1221 bG6	-19.3	7.4	3.4	46 %	16 %
Caprid	1222	-19.4	7.4	3.1	39 %	15 %
Caprid	1222 bC12	-19.2	6.8	3.3	44 %	15 %
Bovid	1125	-19.8	7.6	3.1	33 %	12 %
Bovid	1209	-19.2	8.3	3.2	39 %	14 %
Bovid	1210	-19.6	6.8	3.2	40 %	15 %
Bovid	1221 bG6	-19.7	7.5	3.2	36 %	13 %
Bovid	1209 hB16	-19.1	8.3	3.2	38 %	14 %
Bovid	1210 hB17	-19.5	6.7	3.2	38 %	14 %
Bovid	1403 bC34	-18.8	8.1	3.3	40 %	14 %
Bovid	1215 h1319	-20.0	6.2	3.5	24 %	8 %
Bovid	14 1329	-19.0	8.1	3.4	62 %	21 %
Bovid	12 1123	-19.1	8.2	3.4	20 %	7 %
Cervid	114 1281	-19.0	5.3	3.5	63 %	21 %
Lagomorph	14 1269	-17.9	10.5	3.4	52 %	18 %

from marine foragers, to freshwater foragers, to C_3 and C_4 agriculturalists, and see how KSH fits into such a broad Eurasian context. The sites emphasize particular foods within the diet, though of course, these societies are likely to have mixed a range of foods. Mesolithic hunter-gatherer groups include coastal foragers with a heavy marine diet

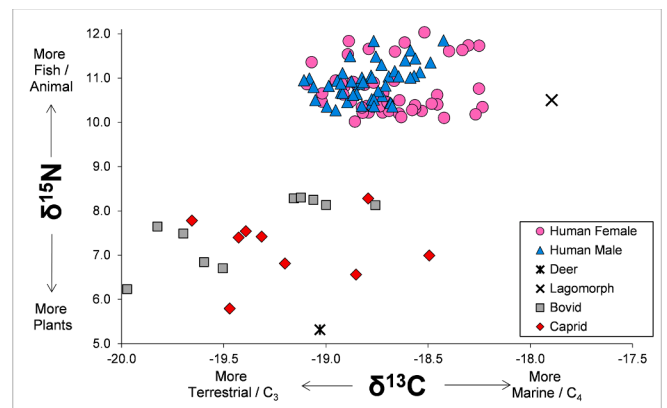


Fig. 6. Comparison of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ for fauna and humans (post-weaning values only) at KSH.

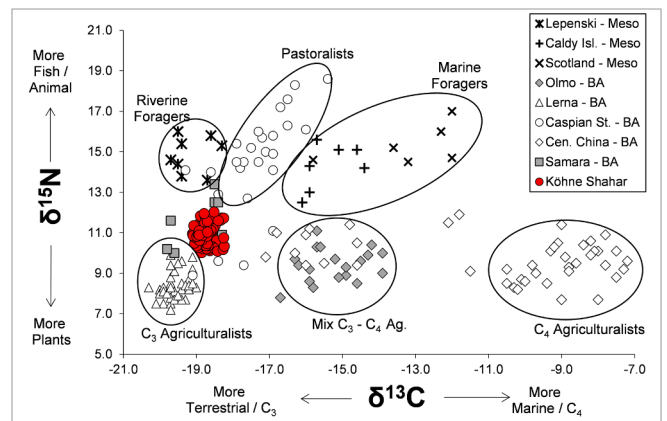


Fig. 7. Comparison of post-weaning Köhne Shahr stable isotope values against other Eurasian populations representing different subsistence strategies.

from the British Isles (Schulting and Richards 2002a, 2002b) and individuals buried at Lepenski Vir along the Danube River with a diet focused on riverine foods (Nehlich et al., 2010). Bronze Age populations include a coastal settlement at Lerna, Greece focused especially on C_3 plants (Triantaphyllou et al., 2008), an inland group from Olmo di Nogara in northern Italy farming a mix of C_3 plants and C_4 millet (Tafari et al., 2009), a population from central China reliant primarily on C_4 millet (Cheung et al., 2017), a population from the Caspian Steppes that relied heavily on domesticated animals (Shishlina et al., 2012), and

Bronze Age people from the site of Tepe Hissar in Iran (Afshar et al. 2019). In terms of proximity, Tepe Hissar is closest to KSH, though it lies closer to the Caspian Sea, and it also overlaps temporally with KSH.

Values for post-weaning serial samples from KSH are plotted in Fig. 7 as red circles. As shown, the KSH samples fall in a restricted area between C₃ Bronze Age agriculturalists on the Greek Coast (Lerna; open triangles), riverine foragers from Lepenski Vir (stars), and Bronze Age pastoralists from the Caspian Steppes (open circles). This implies that the post-weaning KSH diet included a mix of C₃ plant products (e.g., wheat and barley) and animal foods, with perhaps small amounts of fish. The nearby Tepe Hissar III individuals (open squares) are slightly lower in $\delta^{13}\text{C}$ indicating that freshwater fish was more important in the diet than at KSH.

Based on the $\delta^{13}\text{C}$ values, there is little evidence for incorporation of C₄ foods, such as millet, or marine foods at KSH. The lack of millet is in line with recent research showing that broomcorn millet did not reach Southwest Asia and Iran until about 2050 BCE (Huang et al., 2023; Martin et al., 2021), postdating the individuals from KSH. In short, the KSH isotopic data is consistent with an agro-pastoral lifeway with little to no C₄ plant consumption (see Piro 2009; Longford et al. 2009).

Age at weaning estimates for KSH individuals average 2.4 years (n = 8). Using identical techniques, this figure is earlier than that recorded in many archaeologically-older Californian hunter-gatherer populations (Eerkens and Bartelink 2013) and agriculturalists on American Samoa (Eerkens et al., 2019). However, it is similar to that recorded among agricultural populations along the Nile Valley (Eerkens et al., 2018) and some intensive Late Period hunter-gatherers in California (Eerkens et al., 2020b; Eerkens et al., 2022). The KSH age of weaning is also later than that recorded among 1800s Europeans (Henderson et al., 2014; Väre et al., 2022; Waters-Rist et al., 2022).

Age-controlled serial samples, as shown in Fig. 5, suggest isotopic overlap in male and female diet. However, there is greater variation in female diets, especially in $\delta^{15}\text{N}$, and especially between the years following weaning (3–4 years of age) and early adolescence (age 13–15 years). The plot suggests a set of females with generally higher $\delta^{15}\text{N}$ (individuals 165, 258B, and 258C), and a set of females with $\delta^{15}\text{N}$ about 1.0–1.5‰ lower (individuals 167, 168, and 255). After age 13 years, the sample size is small (n = 5 individuals), but male and female values are identical.

By contrast, all of the males plot within a narrower isotopic range during childhood. Between the ages of 5 and 9 years (see Table 4) these male values fall more within the range of the higher $\delta^{15}\text{N}$ female group, leading to average $\delta^{15}\text{N}$ values about 0.4–0.5‰ higher than for all females of the same age range. Combined with the slightly earlier average age of weaning for males (2.0 years) relative to females (2.6 years), this could indicate a difference in enculturation practices (and diet) for males and females. For example, if males were largely responsible for grazing flocks of caprids and/or bovinds away from the village, and began to learn these skills during childhood years, they could have been out in the field for extended periods of time. During such work, these males could have been consuming slightly higher amounts of milk and/or animals encountered during such work (e.g., eggs from bird nests, wild hares). This may account for the higher average male $\delta^{15}\text{N}$ during these years. If boys started learning these activities at an early age, between the second and third year of life, they may have been spending more time away from their mothers than girls. This could lead to earlier weaning for boys than girls.

While some girls may have been engaged in similar work away from the house (i.e., those with higher $\delta^{15}\text{N}$), others may have been associated more with domestic and/or agricultural activities with greater access to plant-derived foods (e.g., bread, fruit), leading to the set of females with lower $\delta^{15}\text{N}$ values. Assuming domestic skills were learned from female kin within the house, such training may have afforded greater and extended access to breastmilk, either directly from their mother or from another lactating female relative. This could explain the later estimated ages of weaning for many of the girls. Indeed, the two girls with early

age-at-weaning estimates (less than 2.0 years) have higher $\delta^{15}\text{N}$ in their weaning foods as if they had been consuming more meat or eggs, while girls weaned later (after age 3.0 years) have lower $\delta^{15}\text{N}$ values for weaning foods as if they had been consuming more plant-based foods.

On average, KSH fauna do not overlap with, and are 2–6‰ lower in $\delta^{15}\text{N}$ than humans, and are overlapping, but slightly lower on average, in $\delta^{13}\text{C}$. Because nitrogen in wild plants is mainly sourced through fixation from the atmosphere by soil and root bacteria, and because $\delta^{15}\text{N}$ in air is similar around the globe, $\delta^{15}\text{N}$ in wild plants tends to be limited in range, both within tissues from the same plant (Evans, 2001), and between plants in the same ecosystem (DeNiro and Epstein 1981; et al., 1999). Typically plant $\delta^{15}\text{N}$ varies between –3‰ and 3‰ in natural ecosystems (Dawson et al., 2002). Tissues in animals consuming these wild plants will be one trophic level higher, or 2–4‰ higher than the baseline plants (Perkins et al., 2014; Schoeninger and DeNiro 1984). By contrast, plants growing in fertilized fields draw some of their nitrogen from fertilizers, such as animal dung, which is higher in $\delta^{15}\text{N}$ (Szpak et al., 2012). Animals feeding on plants from fertilized fields will also be higher in $\delta^{15}\text{N}$.

Unfortunately, we do not know the baseline $\delta^{15}\text{N}$ in plants surrounding KSH. If we take the single wild cervid bone from KSH ($\delta^{15}\text{N}=5.3$ ‰) as representative of baseline values for herbivores foraging on wild browse, the systematically higher $\delta^{15}\text{N}$ values in KSH caprids and bovinds, between 5.8‰ and 8.3‰, suggest domesticated animals were consuming a mixture of wild browse and crops grown in fertilized fields. Further, the low $\delta^{13}\text{C}$ values indicate primarily, or exclusively, C₃ browse for these animals. The greater variation in faunal $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ also suggests domesticated animals were eating plants from a broader range of environments than humans.

That livestock were consuming at least some wild browse, suggests these animals were regularly taken for grazing, a task that necessitates human oversight. As we suggest above, this may have been a gender-biased activity undertaken mostly by younger males, leading to slight differences in the diets and weaning ages between males and females. Interestingly, the single lagomorph analyzed shows $\delta^{15}\text{N}$ values similar to those recorded in KSH humans (at 10.5‰). This suggests the rabbit was eating foods similar in isotopic composition to those humans ate, most likely including plants grown in fertilized fields. This could indicate it was a domesticated rabbit eating table scraps, or alternatively, that it was “wild” but living on the edge of the human-created landscape consuming plant remains thrown out with the garbage or plants from agricultural fields. If the latter, its presence in the KSH midden may indicate some degree of pest management was practiced at the site.

While the weaning process imparted significant changes in $\delta^{15}\text{N}$ early in life, there is evidence for additional dietary change in later years of humans. There seems to be a second dietary shift around age 10–12 years. While there is little to no change in $\delta^{13}\text{C}$, around this age, $\delta^{15}\text{N}$ isotopes become less variable between individuals of the same sex, become equal for males and females, and are on average about 0.5‰ lower than before this age. Then, between ages 14 and 20, $\delta^{15}\text{N}$ values slowly rise again for both males and females.

This teenage pattern, from ages 14–20 years, could indicate a distinctive dietary regime for teenage individuals. For example, if recently married individuals were establishing new households, and sharing foods within this economic unit, this could explain the isotopic convergence of male and female diets. As well, if such recently married individuals did not own a significant number of domesticated animals (especially animals of slaughterable age), they may have consumed greater quantities of plant products, leading to a decrease in $\delta^{15}\text{N}$ values for both males and females. As these individuals aged and started to build up their own flocks, and started to slaughter more of this flock, diets may have increased in the quantity of meats, leading to slowly increasing $\delta^{15}\text{N}$ values across several years as seen in Fig. 5.

The pattern also suggests a more endogamous post-marital residence pattern. If individuals had frequently emigrated from their natal village upon marriage (i.e., exogamy), we would expect to see greater shifts in

diet as they began to reside in new domiciles in new villages with presumably more divergent dietary practices. Instead, the KSH tooth data show relatively stable isotopic values throughout late childhood and teenage years, and similarity between individuals. While $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ are less ideal at tracking mobility, this hypothesis could be tested in future studies using other isotopic systems such as $\delta^{34}\text{S}$ and $^{87}\text{Sr}/^{86}\text{Sr}$.

8. Conclusions

Overall, isotopic findings are consistent with an agro-pastoral diet at Köhne Shahar, as expected. The AMS dates on collagen from four individuals indicate the people in the sample date to the early part of the Bronze Age, roughly 2900–2600 cal BC. Overall, $\delta^{13}\text{C}$ values are low, indicating that C_4 foods were either not present or of minor importance. This contrasts with later Bronze Age periods in west Asia where some C_4 plants, particularly millet, were important in local diets (Huang et al., 2023). Excepting one lagomorph, $\delta^{13}\text{C}$ values are also low in the sampled fauna, showing that C_4 foods were also insignificant in animal browse or fodder. As shown in Fig. 7, the dietary pattern at KSH is distinctive when compared to other populations across Eurasia known to have focused on either agriculture or pastoralism, or consumption of C_4 plants.

Beyond these basic findings, the serial stable isotope samples from 17 teeth representing 11 individuals, as well as isotopic data on 20 associated fauna, help reveal important details that shed new light on Bronze Age lifeways and the organization of society in northern Iran. In particular, we observe difference in sex and especially age, that inform on life history experiences of individuals buried at the site. We summarize several findings below.

First, inter-individual variation in stable isotope values is low within the KSH human population, indicating similar dietary practices among individuals. This result is consistent with findings by Herrscher and colleagues (2021), who document low isotopic variation across a small sample of humans from several Kura-Araxes sites in the south Caucasus. Relative to the humans, isotopic variation within the 10 bovids and 8 caprids from KSH is much higher. The ranges of bovid and caprid $\delta^{13}\text{C}$ are 35 % and 41 % higher, respectively, than humans, while $\delta^{15}\text{N}$ is 24 % and 4 % higher. In short, animals seem to have had greater dietary variation than humans. This finding is consistent with previous stable isotope analyses of caprid and bovid enamel showing animals were accessing different water sources (Samei et al., 2023), and hence plants, across a wide range of environments. Likewise, Herrscher and colleagues (2021) document considerable isotopic variation in animal bones across several Kura-Araxes sites.

Second, an average age of weaning of 2.4 years indicates significant investment on the part of mothers in their children. Breastfeeding is energetically expensive and also decreases female fertility by lowering the chances of pregnancy in natural-fertility populations (Ellison 1995). Assuming the average inter-birth interval exceeded the average age of weaning, this suggests relatively low rates of population growth in this early Bronze Age population.

Third, we observe differences in the average age at weaning for boys and girls, with boys weaned on average about one half year earlier than girls (2.0 vs. 2.6 years). We hypothesize that early enculturation into gender-biased behaviors may explain this pattern. Young boys may have started training or helping with animal husbanding activities in their third year of life, presumably with other male relatives, taking them away from their mothers for extended periods of time. These absences may have encouraged earlier weaning for boys. By contrast, later weaning for three of five young girls implies they stayed closer to their mothers, perhaps while learning important life skills from female relatives, fostering an extended periods of breastfeeding relative to boys. The remaining two girls may have been learning to work outside the house away from their mothers, leading to earlier weaning. Archaeological findings from KSH support heavy investment in craft production, specialties that may have been learned early in life by inhabitants at the

site. Thus, Alizadeh and colleagues (2018) find evidence for several workshops at KSH, including locations involved in metallurgy, ceramics, stone bead, and bone tool production (Samei and Alizadeh 2020).

Fourth, we observed a bimodal distribution in $\delta^{15}\text{N}$ for females between the ages of 3 and 10 years of age. This indicates two different activity patterns for females after weaning. We hypothesize that the group of females with lower $\delta^{15}\text{N}$ were more bound to the domestic sphere (as for the later-weaned girls above), perhaps tending gardens and learning skills within the house. During these activities, girls likely consumed a greater proportion of plant foods. By contrast, a second group of females has higher $\delta^{15}\text{N}$. These girls may have been performing more tasks outside the household, perhaps engaged in animal husbandry (grazing and/or milking livestock), or perhaps learning to make cheese, yoghurt, or other animal-derived food products. These activities may have allowed them to consume a higher percentage of animal-derived foods as part of such training. All boys between the ages of three and ten years group with the latter high- $\delta^{15}\text{N}$ females, suggesting similar activities. These patterns point to important difference in the division of labor and enculturation practices.

Fifth, intra-individual variation in isotopic values between 3–10 years is low, indicating that once a child started on a particular learning trajectory as above, they stayed within that behavioral and dietary pattern. Our serial samples represent approximately 0.5–2 year intervals (depending on the tooth). If there had been marked changes in diet across the seasons, for example heavy consumption of fish during spring runs or greater-than-normal consumption of plant foods during fall harvest, we would expect to see greater intra-tooth variation. Instead, the stable values suggest either a more monotonous diet throughout the year and/or significant storage of foods to even out seasonal variations in resource availability.

Sixth, a slight decrease in $\delta^{15}\text{N}$ around age 12–14 years, followed by a slow increase between 14 and 20 years, for both males and females, may correlate with marriage, the establishment of new households, and gradual development of these economic units. Within new households access to animal meat may have been limited initially, especially if it took a number of years to build up herds of caprids and/or bovids. Perhaps by late teenage years, married couples (or groups if polygamy was practiced) may have built up enough capital to begin supporting children themselves, and could start enculturating the next generation in similar gender-biased tasks.

Finally, relative stability in dietary isotopes within an individual, and similarities between females and males during teenage years, implies relatively low rates of residential mobility. If people had been moving between villages, especially villages set in different ecological niches, for example as part of post-marital residence shifts, we would expect to see greater changes across serial samples in the isotopic composition of diet. In many small-scale societies around the world, such long-distance marriage is necessary to avoid inbreeding. The findings here imply that KSH had a large-enough population base that such long-distance marriages were not necessary. This finding, too, has significant support in previous studies at KSH (Alizadeh et al., 2015, 2018; Samei et al., 2019), as well as Kura-Araxes Bronze Age sites more generally (Frangipane 2015; Longford and Sagona 2021).

CRediT authorship contribution statement

Jelmer W. Eerkens: Writing – review & editing, Writing – original draft, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization. **Sepideh Asgari:** Writing – review & editing, Writing – original draft, Resources, Investigation, Formal analysis, Conceptualization. **Karim Alizadeh:** Writing – review & editing, Writing – original draft, Resources, Investigation, Conceptualization. **Diana Malarchik:** Writing – review & editing, Writing – original draft, Investigation, Formal analysis. **Samantha Cramer:** Investigation, Funding acquisition, Formal analysis. **Glendon Parker:** Writing – review & editing, Writing –

original draft, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper.

Data availability

Data are available in the [supplementary file](#).

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Appendix A. Supplementary data

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