Stable isotope analysis on human remains from the final Early Holocene in the southern Puna of Argentina: The case of *Peñas de las Trampas* 1.1

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In this work are presented the results of isotopic analyses made on bone remains of human individuals ($n = 6$) from the southern Puna of Argentina dated to the final Early Holocene (ca. 8230–8000 BP). They were found in structures located in Peñas de las Trampas 1.1, a rockshelter at 3582 m.a.s.l. in Antofagasta de la Sierra, in the southern Argentinian Puna. They contain multiple secondary burials. Bone fragments were recovered from at least six individuals, three in each structure. Stable isotopes of Carbon ($\delta^{13}C$) and Nitrogen ($\delta^{15}N$) analysis were aimed at defining aspects related to the palaeodiet of the six individuals within the palaeo-economic subsistence spectrum typical of hunter–gatherers. It is worth noting that these human remains are among the earliest from North-Western Argentina, where funeral practices are related with the transportation of certain anatomical parts. The palaeodietary inference considers, on the one hand, the extreme aridity of this geographical area and its impact on the isotopic ecology. And, on the other, it takes into account the fact that four of the six individuals under study were breastfed infants. The results are in agreement with the expected values of the period, which has been characterized as the beginning of the arid Altithermal.

Keywords: Stable isotopes, Early Holocene, Puna, Palaeodiet, Hunter–gatherers, Breastfeeding, Aridity

Introduction

Palaeodietary studies for the period comprising the transition between Early and Middle Holocene remain an understudied subject for the archaeology of the Puna and the whole North-Western (NW) region of Argentina. This is due to the scarcity of human remains discovered for this period. Inferences about diet are usually made based on elements found in the archaeological record. These are food remains such as, vegetables-like carob, beans and peppers-and animals, especially Camelidae and Chinchillidae.

This paper aims at introducing palaeodietary analyses in the Argentine Puna during the final phase of the Early Holocene by analysing the stable isotope ratios of Carbon and Nitrogen ($\delta^{13}C$ and $\delta^{15}N$) in the human bone of six individuals. Data interpretation will be followed by a model, based on the available information, for the reconstruction of food chains in the past. The palaeodietary inference will consider the extreme aridity of this geographical area and its impact on the isotopic ecology, as well as the fact that four of the individuals under study were unweaned.

Description of the archaeological site

The area under study is in the Department of Antofagasta de la Sierra, located in the north of the Province of Catamarca, in NW Argentina (Fig. 1). Nowadays, this region is an extreme desert (with less than 50 mm annual rainfall) located over 3400 m.a.s.l. within the region ecologically known as Salt Puna (in Southern Puna of Argentina). The burial remains studied come from an archaeological site called *Peñas de las Trampas 1.1* (PT1.1), located in a rockshelter of huge dimensions situated at 3582 m.a.s.l. Recent findings in PT1.1 have yielded two radiocarbon dates: 10190 BP (UGA-01975) and

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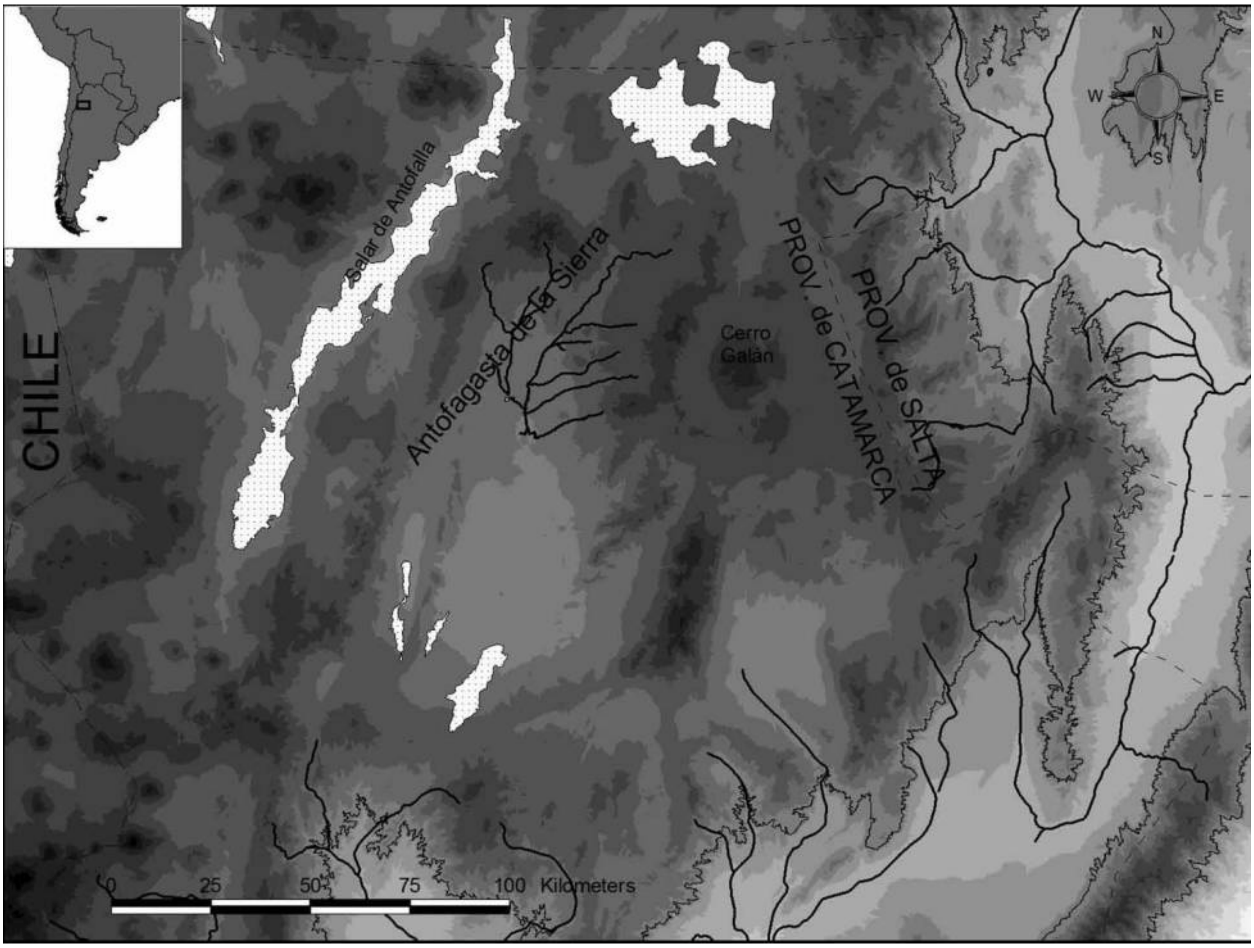

Figure 1 Map of the area.

10030 BP (LP-1788) (Martínez *et al.* 2010a, 2010b). These dates were obtained from coal samples from the core of two combustion structures associated with a few lithic artefacts and burnt bone remains of modern fauna (unit 3E). Dates provide evidence of the oldest cultural component of PT1.1, and although they are spatially very close to one of the burial structures (see Fig. 2), they are chronologically and stratigraphically distinct, since the events related to the funerary practices only began ca. 8440 BP.

Two funerary structures, named Funerary Structures 1 and 2 (FS1 and FS2, see Fig. 2), were found within the rockshelter of PT1.1. They are both very similar and consist of suboval-shaped diggings carefully lined with grass (*Grammineae*) sheaves at the sides and bottoms, with no lid. FS1 has been partially described in a previous article (Martínez *et al.* 2007) and is 97 × 65 × 46 cm, while FS2, discovered later, is 115 × 80 × 42 cm (length, width and depth, respectively). The grass sheaves from each structure have been radiocarbon dated and allow us to determine that FS1 was built at 8440 BP (UGA-9073) and FS2, at 8210 BP (UGA 9097) (Martínez 2012).

Human bones were found inside both structures and they come from at least three individuals in each structure (total minimum number of individuals = 6; Calisaya *et al.* in preparation). The recovered bone remains are very well preserved, but the skeletons are not complete. In fact, most of the bones were missing, which has been interpreted as a result of cultural practices, for some bones showed anthropic postmortem marks. Likewise, the presence of carnivore traces or those of other attritional agents is almost null.

Regarding the age of the individuals, the presence of three sub-adults has been determined at FS1 (all of them under 10 years of age), and one adult (ca. 20 years old) and two sub-adults (under 2 years of age) at FS2. It has been impossible to determine the sex of the individuals due to their young age and, in the case of the adult found at FS2, to the fact that the diagnostic bones were missing (Calisaya *et al.* in preparation).

As regards the specific chronological situation of the human remains, the bone collagen of all individuals from both structures was accelerator mass spectrometer (AMS) dated, in order to compare them to the dates on grass from the structures and to evaluate the potential synchronicity of their deaths. The obtained dates are detailed in Table 1.

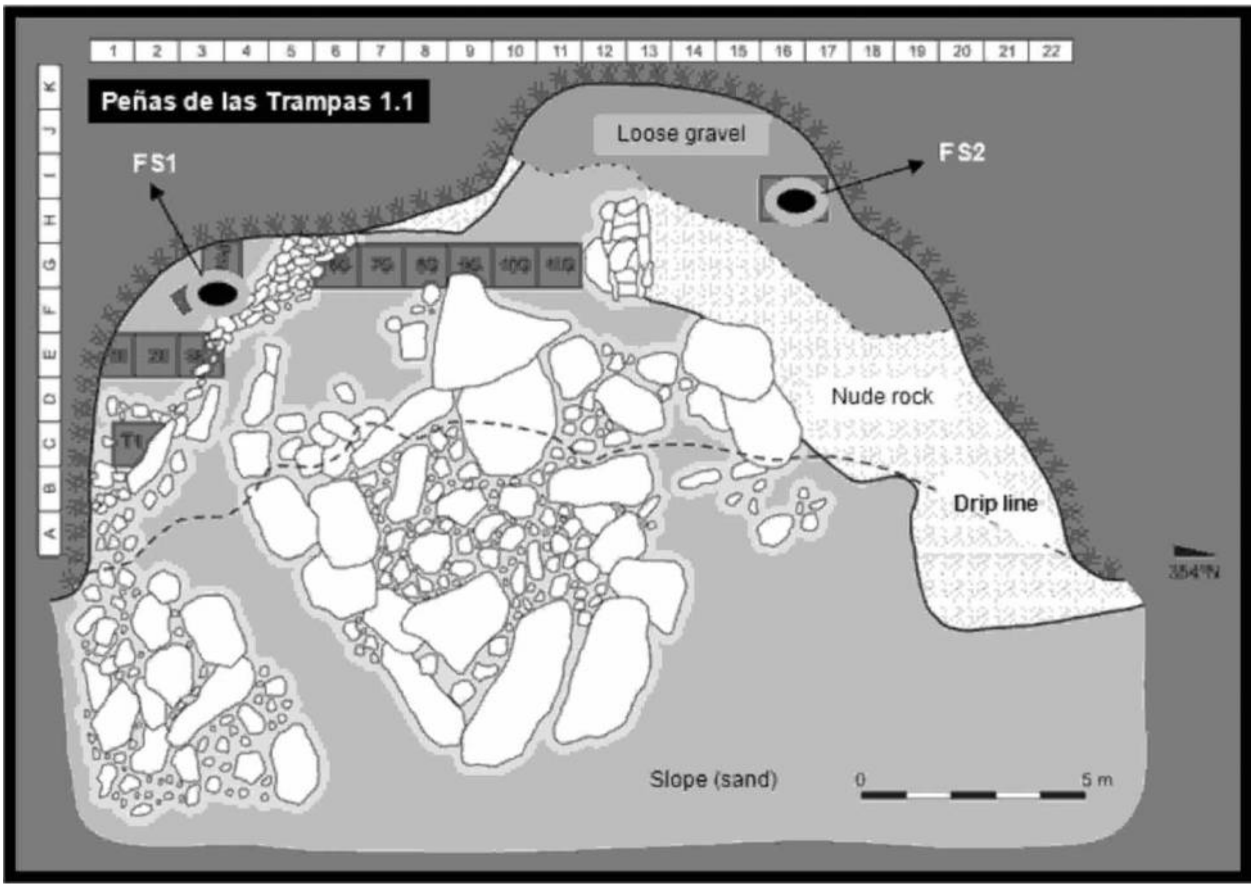

Figure 2 Plan sketch of *Peñas de las Trampas 1.1* rockshelter.

It is noticeable that most of the dates of the individuals from both structures are grouped within the short span of 8260–8110 BP, even considering the dates error ranges. Statistically speaking, they are synchronic, except for the date of 8000 BP of individual 3 from FS2, which is statistically dissociated from the

Table 1 Dates of all the individuals from FS1 and FS2 in PT1.1. Calibration curve ShCal04, (McCormac *et al.* 2004); software used, OxCal v4.1.5 (Bronk Ramsey 2009)

Sample ID	Lab.	Material	RCYBP	δ^{13}C	Calibrated dates, cal BC (68.2%)	Calibrated dates, cal BC (95.4%)
PT1.1 FS1. Ind.1	UGAMS 4994	Human bone	8230 ± 30	−17.29	7282–7279 (1.0%) 7250–7231 (6.8%) 7186–7077 (60.4%)	7309–7069 (95.4%)
PT1.1 FS1. Ind.2	UGAMS 4995	Human bone	8140 ± 30	−16.14	7135–7103 (15.2%) 7084–7040 (53.0%)	7180–7022 (85.6%) 6969–6944 (1.9%) 6938–6913 (2.4%) 6882–6835 (5.5%)
PT1.1 FS1. Ind.3	UGAMS 4996	Human bone	8150 ± 30	−14.5	7136–7102 (21.3%) 7085–7046 (46.9%)	7184–7026 (91.2%) 6962–6949 (0.6%) 6934–6917 (1.0%) 6880–6842 (2.6%)
PT1.1 FS2. Ind.1	UGAMS 4997	Human bone	8170 ± 30	−16.15	7141–7096 (34.7%) 7089–7055 (33.5%)	7292–7274 (1.3%) 7251–7230 (1.5%) 7186–7040 (92.6%)
PT1.1 FS2. Ind.2	UGAMS 4998	Human bone	8210 ± 30	−19,26	7177–7075 (68.2%)	7305–7218 (17.2%) 7196–7059 (78.2%)
PT1.1 FS2. Ind.3	UGAMS 4999	Human bone	8000 ± 30	−18.35	7024–6966 (18.1%) 6948–6935 (3.8%) 6916–6881 (11.7%) 6840–6750 (29.9%) 6722–6706 (4.7%)	7033–6693 (95.4%)
PT1.1 EF1	UGA 9073	Grass	8440 ± 40	−22.58	7527–7451 (52.8%) 7401–7370 (15.4%)	7548–7338 (95.4%)
PT1.1 EF2	LP 1782	Grass	7790 ± 90	−25	6644–6474 (68.2%)	6907–6888 (0.6%) 6828–6413 (94.8%)

rest (C. Greco, pers. comm. 2010). The 8440 BP (UGA-9073) dating on grass that corresponds to the building of FS1 is not fully consistent with the datings obtained on the individuals deposited there, as there would be a delay between the construction of the structure or, strictly speaking, the cutting of the grasses, and the death of the individuals. For FS2, the datings are also consistent, although here the death of the human individuals is quite simultaneous with the building of the funerary structure. In any case, the potential variations of radiocarbon dating methods (AMS versus standard dating, bone collagen versus plant remains) should be considered.

Even if the death of most of the individuals took place quasi-synchronically, the human remains of individual 3 would have been placed later than the others in FS2. This suggests a certain continuity (local tradition?) regarding the use of both burial structures and the burial practices for more than 500 years if we consider the earliest date that FS1 was built and the latest human date in FS2 (8480–7970 years BP including SD). This kind of funeral practice may likely be related to an ancestor cult linked to an early sense of social circumscription or territoriality (Aschero 2007). In the Peñas de las Trampas 1.1 case, the specific factors that caused the death of these individuals, mainly of the newborns, within that brief period are still to be determined.

Isotopic methodology

Stable isotope analysis, especially that of Carbon ($^{13}C/^{12}C$) and Nitrogen ($^{15}N/^{14}N$),[1] has been established as independent and reliable evidence to test hypotheses for palaeodietary issues. This is due to the fact that isotopic signals from human tissues derive from assimilated food and they remain constant after the individual's death (Ambrose 1993). $\delta^{13}C$ analysis allows to distinguish different sources of food, for carbon becomes part of the food chain after being assimilated by vegetables in different ways (Ambrose 1993). There is a differential routing of carbon atoms to collagen and hydroxyapatite. In collagen ($\delta^{13}C_{co}$) it derives mainly from dietary proteins, and in the case of hydroxyapatite ($\delta^{13}C_{ap}$), it is produced also from carbohydrates and fats (Krueger and Sullivan 1984). Therefore, there is an offset related to the fact that animal protein amino acids always contribute to collagen production while only some of the plant amino acids take part. In this way, the ratio of both fractions ($\Delta^{13}C_{apatite\text{-}collagen}$‰) can be used to discuss how protein and non-protein sources differ regarding their isotopic values. Thus, we refer here to dietary protein, where inferences are carried out based on the values of collagen and we do the same on hidroxyapatite samples with respect to the overall diet.

As regards $\delta^{15}N$ values can be used by archaeologists and ecologists to estimate the trophic level of the organism, since consumers present a typical enrichment of about 3–4‰ in ^{15}N relative to their diets (De Niro and Epstein 1981). As carbohydrates and fats do not have nitrogen, the nitrogen isotope ratio in tissue is mainly a function of the source of dietary protein plus the effect of trophic enrichment (Ambrose *et al.* 1997). However, nitrogen isotope ratios can be altered by factors other than trophic fractionation. A negative correlation between herbivore ^{15}N and annual rainfall has been recorded in different continents (Sealy *et al.* 1987). Some authors have suggested such correlation is accounted for by the presence of a higher content of nitrate and ammonia in salty soils, which are characteristic of arid environments (Pate 1994; Amundson *et al.* 2003). In this sense, it has been proposed recently that in the dry Argentine Puna the variation in isotopic values $\delta^{15}N$ in herbivores for the duration of the Holocene is owed to changes in the preponderance of inorganic over organic soils (Samec *et al.* 2014).

Also, the effect of trophic fractionation between sub-adults and their mothers must be taken into account (Fogel *et al.* 1989; Fuller *et al.* 2006). Based on previous work, it is imperative to take into account this variable to avoid making skewed inferences about the diet of the population under study. For example, before inferring that the carbon enrichment in sub-adult individuals is due to possible consumption of crassulacean acid metabolism (CAM) or C_4 resources, we need to rule out the possibility of this enrichment being the product of a trophic level effect due to breastfeeding. However, in this particular case we do not have enough adult reference values to compare.

Palaeodietary background and dietary expectations

So far, the dietary expectations for human populations in the Argentine Puna during the Early Holocene, based on the archaeological record, consist of a protein-focused diet based on the hunting of wild animals (namely camelids – *Vicugna vicugna* and *Lama guanicoe*-, chinchillids, and cervids), coupled with the gathering of local vegetables (such as the tuberous root commonly called soldaque – *Hipsocharis* sp., carob – *Ceratonia siliqua*, cacti – Opuntia sp., peppers – Capsicum sp.), plus some

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[1] Stable isotope ratios are expressed using the delta notation (δ‰). The difference between carbon isotopic values of organic and inorganic bone fractions is expressed with the Δ symbol (according to Ambrose *et al.* 1997). δ represents the difference between the isotopic ratio of the sample under study and that of a standard which, in the case of carbon, is the V-Pee Dee Belemnite carbonate, and in the case of nitrogen, the atmospheric (AIR) N_2. As this difference is very small, the relationship is expressed in parts per mil (‰) according to the following equations: $\delta^{13}C$ (‰) $= [(^{13}C/^{12}C_{sample} - (^{13}C/^{12}C)_{standard/}(^{13}C/^{12}C)_{standard}]*1000$; $\delta^{15}N$ (‰) $= [(^{15}N/^{14}N)_{sample} - (^{15}N/^{14}N)_{standard/}(^{15}N/^{14}N)_{standard}]*1000$.

wild Leguminosae and Cucurbitaceae from temperate regions obtained as a result of the great mobility of the Puna inhabitants (Yacobaccio 1994; Martínez *et al.* 2010a, 2010b).

However, palaeodietary studies using an isotope-based methodology are still scarce in the region. As regards those made in NW Argentina for strict hunter–gatherers, there is only one study for the period 10200–8420 BP (Olivera and Yacobaccio 2002). The study focused on one adult individual one adult individual found at the Huachichocana site (Province of Jujuy, Argentine Dry Puna) that showed $\delta^{13}C_{col} = -15.73‰$ and $\delta^{15}N_{col} = 10.47‰$ (Olivera and Yacobaccio 2002). It presents a remarkable value for the hydroxyapatite fraction ($\delta^{13}C_{ap} = -3.64‰$), considering this is a hunting and gathering population. This apatite value, within the range usually found among societies that grow and consume maize, has been explained by the authors as the result of the inclusion of CAM plants (*Opuntia* sp. and *Tilandsia* sp.), which are available in the Andean record.

With regard to the isotopic register of individuals associated with hunting-based-economies, this remains the case. Table 2 summarises the available information for the Argentine Puna during the Holocene Period. Unfortunately, neither the C/N relationship nor the methodology employed for hydroxyapatite extraction are available for these individuals.

With regard to the isotopic register of the associated fauna, we possess a series of values that correspond to the current area under study. They belong to the archaeological site Salamanca Cave, in Antofagasta de la Sierra (3560 m.a.s.l.) although the more systematic development is that available for Dry Puna. It consists of a series of measurements on the Hornillos 2 site, at 4200 m.a.s.l., with occupations dated equally in the Early and Mid-Holocene, from the years 9710 to 6130 BP. The results of this study demonstrate little significant difference between the values $\delta^{13}C$ of Camelidae regarding both periods (being -17.9 ± 0.9 for the early period and $-17.2 \pm 1.7‰$ for the middle), but instead a major difference in the case of values $\delta^{15}N$ ($+6.5 \pm 1.9$ and $+8.4 \pm 1.6$). This was explained, as already mentioned, based on the changes in the soil composition within the frame of a marked increase in aridity. Also for this sector of the Puna we have at our disposal a value for the Early Holocene, corresponding to a Camelidae sample ($\delta^{13}C$ -17.6), discovered at 3800 m.a.s.l. and with a chronology of between 9050 and 9200 BP (Fernández *et al.* 1991).

As concerns vegetable resources, there is no direct measurement available of the sample in this chronology. However, values of the resources have been obtained thanks to the radiocarbon dating and through the measurement of current resources in Puna. The research of the area under study has focused above all on the resources in agricultural production (Killian Galván and Salminci 2014). The corresponding information for Antofagasta de la Sierra is synthesised in Table 3.

It is important to emphasise that there has been a significant correlation registered in the Puna between altitude and values $\delta^{13}C$ in Camelidae (Fernández and Panarello 1999–2001) owing to the changes in the proportions of flora C_3 and C_4 along the latitudinal gradient. A similar tendency has been noted in $\delta^{15}N$ due to the greater presence of humidity at higher altitudes (Samec *et al.* 2014). In this sense, it shows that the middle values of the samples in both Punas, dry and salty, differ most likely since Hornillos 2 is found at a higher elevation than Salamanca Cave. Therefore, we consider that the trends found in the Dry Puna are adequate to deal with the record under discussion here.

Materials and analytical methods

The isotopic analyses were carried out on collagen and hidroxyapatite samples. Preparation and analysis were conducted at the Center for Applied Isotope Studies University of Georgia, USA. The bone sample for analysis is first thoroughly washed, using an ultrasonic bath, and wire brushed to remove external material. To remove diagenetic carbon compounds, the bone is soaked overnight in 1 N acetic acid. The samples are then washed free of acetic acid by repeated decantation.

For analysis of carbon isotopes in bioapatite, approximately 100–500 mg of the cleaned bone powder is transferred to a vacuum flask and evacuated to remove air. The bone is then reacted under vacuum with about 2 ml degassed 1 N HCl. The reaction is usually completed within 20 minutes or even less and can be monitored to determine whether or not collagen pseudo-morphs have sunk to the bottom. If the bone is suspected of being highly altered, this reaction should be performed at freezing point – 0°C – in the ice bath to improve collagen recovery.

The released CO_2 is purified by cryogenic separation and collected in sealing tubes for AMS and stable isotope analyses. Bioapatite content in modern bone is about 1%, and about 0.8% in modern enamel. However, in archaeological samples that have been treated with acetic acid, this value should be in the range of 0.4–0.7%; therefore, 100 mg of bone should yield about 0.4–0.7 mg of carbon. If yields are much higher, it is probable that diagenetic carbonates were not completely removed (Cherkinsky 2009; Zazzo and Saliege 2011).

For collagen extraction the crushed bone was treated with 1 N HCl at 4°C for 24 hours. The

Table 2 Values $\delta^{13}C$ and $\delta^{15}N$ in humans for the Early, Middle and Late Holocene of NW Argentina

Provenience	Archaeological site	Estimated age	$\delta^{13}C_{co}$	$\delta^{13}C_{ap}$	$\delta^{15}N$	Chronology BP
Dry Puna	Huachichocana	Adult	−15.73	−3.64	10.47	ca. 10,200-8420
Dry Puna	Inca Cueva 4	Adult	−16.90	~	14.17	ca. 5300
Dry Puna	Salar de Pastos Grandes*	25–35	−16.80	~	~	3738 ± 46
Antofagasta de la Sierra	Quebrada Seca 3	Unborn	−16.56	~	13.44	4410 ± 50
Antofagasta de la Sierra	Punta de la Peña 11	4 months	−14.90	~	14.36	3210 ± 50

Olivera and Yacobaccio (2002); *López and Miranda (2007–2008).

residue was filtered, rinsed with deionised water and under slightly acid condition ($pH = 3$), and heated at 80°C for 6 hours to dissolve collagen and leave humic substances in the precipitate. The collagen solution was then filtered to isolate pure collagen and dried out. The dried collagen was combusted at 575°C in evacuated/sealed Pyrex ampoule in the present CuO. The carbon dioxide and nitrogen were cryogenically separated for analysis. The resulting carbon dioxide was cryogenically purified from the other reaction products and catalytically converted to graphite using the method described by Vogel *et al.* (1984). $^{14}C/^{13}C$ ratios were measured using the CAIS 0.5 MeV accelerator mass spectrometer. The sample ratios were compared to the ratio measured from the Oxalic Acid I (NBS SRM 4990). The sample $^{13}C/^{12}C$ ratios were measured separately using a stable isotope ratio mass spectrometer and expressed as $\delta^{13}C$ with respect to Pee Dee Belemnite with an error $<0.1‰$. The sample $^{15}N/^{14}N$ ratios were measured separately using a stable isotope ratio mass spectrometer and expressed as $\delta^{15}N$ with respect to atmospheric air nitrogen, with an error $<0.1‰$.

Results

The obtained $\delta^{13}C_{co}$, $\delta^{15}N$ and $\delta^{13}C_{ap}$ values, shown in Table 2, indicate that all of the samples are satisfactory for palaeodietary inference. In the case of collagen, the C/N ratio is used to determine if the isotopic signature is the primary one (in which case, $C/N = 2.9–3.6$, after De Niro 1985).

By grouping the samples by burial structure (Fig. 3A), the segregation of carbon isotopic values is not clear, since the value obtained for the adult individual from FS2 overlaps with those from FS1. However, segregation is clear in the case of nitrogen. $\delta^{13}C_{co}$ values (‰) present a mean value of $-16.9 \pm 1.7‰$. In at least three cases it has a protein diet controlled by a photosynthetic pattern C_3. However, three of the individuals appear to posses a higher contribution of resources under the pattern C_4. Accordingly, the mean $\delta^{13}C_{ap}$ (‰) value is $-13.9 \pm 1.3‰$, and as we will see later, it is expected in diets with a significant vegetable component under the photosynthetic pattern C_3. In the case of $\delta^{15}N$, values show less variation as compared to carbon values in collagen, with a mean of $+13.8 \pm 0.9‰$, suggesting a trophic chain probably enriched from its base. This could possibly be the case if we consider that the characteristic aridity of the Puna environment could have been more intense during the chronological period under study (Tchilinguirian 2009, 2011). As Samec *et al.* (2014) show, this scenario has its correlate in herbivores, given that since the Mid-Holocene, the values $\delta^{15}N$ are found to be more enriched than in

Table 3 Isotopic values of available nutrient resources in Antofagasta de la Sierra (in Mondini *et al.* 2010)

Archaeological sites	Species	$\delta^{13}C$	$\delta^{15}N$	C/N	Chronology BP
Flora					
Punta de la Peña 4	*Geoffroea decorticans*	−25.7	n/a	n/a	1460 ± 40
Corral Alto		−26.4	n/a	n/a	720 ± 40
Punta de la Peña 9		−23.9	n/a	n/a	1480 ± 40
Alero sin cabeza	*Prosopis* sp.	−26.3	n/a	n/a	1745 ± 40
Fauna					
Salamanca Cave	Big Camelidae	−16.2	9.1	3.3	ca. 3500
Salamanca Cave	Camelidae	−14.3	9.4	3.3	6250 ± 70
Salamanca Cave	Camelidae	−16.4	8.7	3.4	~
Salamanca Cave	Camelidae	−16.3	7.9	3.3	7500 ± 60
Salamanca Cave	Small Camelidae	−16.4	9.4	3.4	7550 ± 60
Salamanca Cave	*V. vicugna*	−18.8	7.8	3.4	7540 ± 50
Salamanca Cave	*Lama guanicoe*	−15.3	8.9	3.3	7540 ± 50
Salamanca Cave	Camelidae	−17.6	9.7	3.3	7620 ± 60
Salamanca Cave	Small Camelidae	−9.9	10.9	3.3	7990 ± 60
Salamanca Cave	Camelidae	−15.4	9.5	3.3	8100 ± 50
Salamanca Cave	Small Camelidae	−17.3	9.2	3.5	8100 ± 50
Mean	−15.8	9.1			
SD	2.3	0.8			

Table 4 Obtained $\delta^{13}C_{co}$, $\delta^{15}N$ and $\delta^{13}C_{ap}$ values, and age estimation of the individuals from Peñas de las Trampas 1.1

Sample ID	$\delta^{13}C_{co}$	$\delta^{13}C_{ap}$	$\Delta^{13}C$	$\delta^{15}N$	C%	N%	C/N	Estimated age
PT1.1 EF1. Ind.1	−17.29	−12.34	4.95	14.1	37.61	12.1	3.6	10 years ± 12 months
PT1.1 EF1. Ind.2	−16.14	−14.62	1.52	14.8	35.06	12.47	3.3	2 years ± 6 months
PT1.1 EF1. Ind.3	−14.5	−13.71	0.79	15.0	38.87	13.4	3.4	1 year ± 4 months
PT1.1 EF2. Ind.1	−16.15	−12.62	3.53	13.5	45.96	17.17	3.1	~20 years
PT1.1 EF2. Ind.2	−19.26	−16.02	3.24	12.5	35.41	11.47	3.6	9 months ± 3 months
PT1.1 EF2. Ind.3	−18.35	−14.17	4.18	13.2	33.5	12.09	3.2	8 months ± 3 months

prior and later periods. The values $\delta^{13}C_{ap}$ (Fig. 3B) overlap even more than in the collagen, although one of the individuals possesses a much lesser signal than the rest, that is to say with a higher presence of vegetables C_3 in its diet. With respect to the difference $\Delta^{13}C_{ap\text{-}co}$ (Fig. 3C), a value greater than 4.4‰ corresponds to a depleted $\delta^{13}C$ value in proteins relative to that of the whole diet (Ambrose *et al.* 1997). Those lactating individuals who possess more enriched $\delta^{15}N$ values are those that have a protein and total diet with similar isotopic signals.

Concerning the rest of the isotopic values of humans in the Puna, the two infant individuals (one corresponding to the beginning of Late Holocene), as well as the adult one, are found in the same distribution as the humans in the Peñas de las Trampas 1.1 (or PT1.1). The one individual that differs from the group is that of Early Holocene, owing to a low $\delta^{15}N$ value.

Based on the little available information for the reconstruction of the isotopic ecology, we can begin an initial paleodietary inference. We follow here the proposal put forth by Newsome *et al.* (2004), who estimated the contribution of vegetable and meat resources. This approach requires a series of corrections. In the first place, the 4‰ fractionation should be applied between bone-muscle in values $\delta^{13}C_{co}$ of meat resources (there is no difference between tissues in the $\delta^{15}N$ values). In this way we can correct the values of carmelidae. On the other hand, for the estimated diet of humans, we consider the fractionation for collagen proposed for Drucker and Bocherens (0.8–1.3‰ in $\delta^{13}C$ values and 3–5‰ in $\delta^{15}N$ values, 2004), but we still assume a fractionation of 5‰ in carbon in order to include the plants in the same plot. Also we just consider a fractionation factor of 4‰ for $\delta^{15}N$ values. In Table 3 we described the values of vegetables from Antofagasta de la Sierra. Furthermore, we add the current vegetable values (corrected for *Suess effect*), measured in Puna: a sample of *Hipsochoris* sp. (C_3) and two sampes of *Opuntial* sp. (CAM).

If we consider the age groups, Carbon and Nitrogen isotope values are not as expected. Individuals at the breastfeeding age present both richer and lesser values than adult and young individuals. As seen in Fig. 4, the estimated diet of the individual associated with the Early Holocene is consistent with the values of Camelidae of this period. The rest of the individuals are either associated with the values of Salamanca Cave or the values of Hornillos 2 of the Mid-Holocene.

As concerns the group belonging to the Peñas de las Trampas 1.1, if we consider that above 4000 m.a.s.l. the diets of Camelidae are found to be dominated by C_3 values, we can argue that the lesser values, as much in $\delta^{13}C_{co}$, as in $\delta^{15}N$, could come from higher altitudes, than the rest. Above all, if we consider the aforementioned, the isotopic signal of those breastfeeding would be higher than that could have been offered by the mother.

By this methodological route, the vegetable deposits appear to be non-existent. However, although it proves problematic determining how important were the vegetable contribution in diets based on the measurements of collagen, we can observe its importance

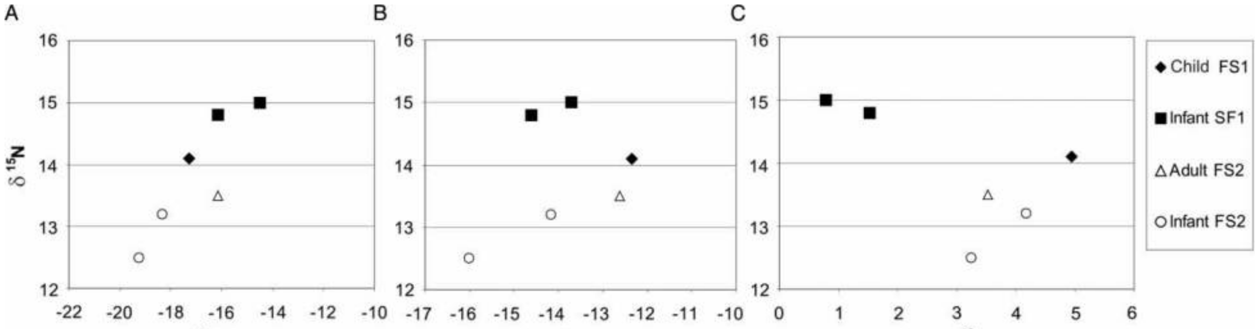

Figure 3 (A) $\delta^{13}C_{co}$ vs. $\delta^{15}N$ values; (B) $\delta^{13}C_{ap}$ vs. $\delta^{15}N$ values; (C) $\Delta^{13}C_{ap\text{-}co}$ vs. $\delta^{15}N$ values; discriminated by funerary structure and age range.

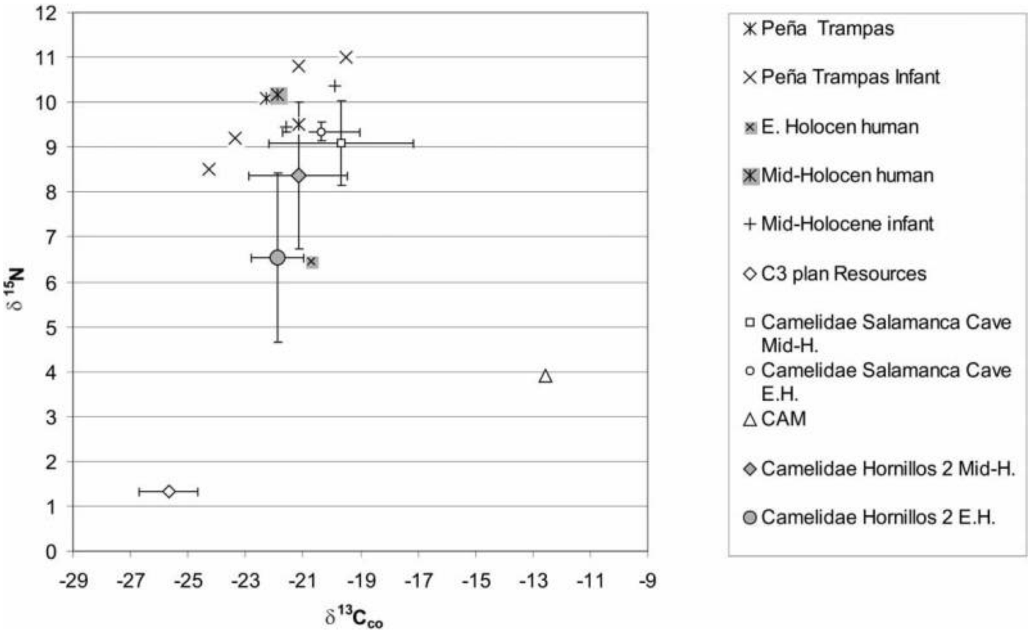

Figure 4 Estimated human diets and resources from Early and Mid-Holocene.

based on the values in hidroxyapatite in the individuals of Peñas de las Trampas 1.1. Based on the values, we can infer that there existed a greater presence of C_3 resources, given the values of the estimated diets.

Considering the fractionation of +9.4‰, the inferred diets are within the range of δ^{13}C −25.02‰ and −21.3‰, that is to say, they are in agreement the chañar (*Geoffroea decorticans*), carob, and soldaque values.

It is imperative to perform a sampling of these present resources in Antofagasta de la Sierra, in order to know the values δ^{15}N, which would allow us to improve the model presented here.

Discussion

As expressed before, one of the aims of this study is to categorise the relative importance of the food items consumed by the individuals under study, particularly the importance of vegetables having a photosynthetic pathway different form the most abundant C_3-type in the area. Considering the values of $\delta^{13}C_{\text{apatite-collagen}}$ we can state that the protein components of at least four individuals are higher than the rest of the diet, a fact that is in agreement with a diet including tubers as source of carbohydrates. Regarding the consumption of *Amaranthus* sp., this pseudocereal, which follows a C_4 photosynthetic pathway and has high-protein content, was found in the archaeological record of the area but later in time. For example, a wild species of *Amaranthus* sp. has been identified in Peñas de la Cruz 1.1 for the 7900–7200 BP period (Arreguez *et al.* 2013). Thus, we can say that pseudocereals would not be relevant dietary components in the early individuals under study.

The collagen records consistently show an offset towards proteins at the expense of energy, which would be explained by the consumption of herbivores with a C_4 diet. In agreement with this proposition, the isotopic data for vicuñas in the Puna region indicate that, nowadays, these animals prefer to eat Gramineae (Yacobaccio *et al.*, 2009; Samec 2011) and it is likely that in the past these animals included C_4 Gramineae such as *Sporobolus rigens* grass. In any case, the need to generate values adjusted to an adequate regional isotopic ecology needs to be emphasised, since environmental conditions in the Early Holocene do not have modern counterparts. As mentioned, so far we only possess values generated by Mondini *et al.* (2010) for camelids from Cueva Salamaca.[2] As can be seen in Fig. 4, the protein diet could be explained by the consumption of these herbivores with a C_4 diet that show extreme values both for nitrogen ($\delta^{15}N = 10.9$‰) and carbon ($\delta^{13}C = -9.9$‰).

Regarding breastfeeding and weaning, this sample, in spite of its small size, allows us to account for the different types of diets that can be present during childhood. In the case of those individuals who died within their first year of age, they present depleted stable isotopes values, as opposed to those who outlived their first year. Unfortunately, the individuals' skulls are not available so as to make a more accurate age estimation. And given the nature of the sample, we cannot discuss how different the values of the sub-

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[2] Further archaeological and modern samples from Antofagasta de la Sierra are currently under analysis by this team and will soon be available.

adults were under this trophic effect from those corresponding to their mothers. For the time being, the more depleted $\delta^{13}C_{co}$ and $\delta^{15}N$ values in both infants under 1 year of age can be attributed most probably to their mothers' consumption of tubers (with a C_3 photosynthetic pathway) and food containing proteins with more depleted isotopic values than the rest of individuals.

As pointed out by Tchilinguirian (2009), an increase in aridity started in the area in the period under study. This setting explains not only the $\delta^{15}N$ values in our human individuals, but also those in the camelids studied by Mondini *et al.* (2010), which would have consumed pastures enriched in nitrogen isotope. These pastures could have undergone an adaptive process as water became scarce. In the same way, there could have been an increased consumption of C_4 *Graminea* as the habitually consumed resources (C_3) became scarcer. However, recent studies made by Samec *et al.* (2014) do not fully support this theory because although they hold that the C_3–C_4 rate could have undergone modifications, they point out that the samplings made so far do not indicate any statistically relevant difference.

Lastly, the fact should be pointed out that in light of zooarchaeological research developed in the area (e.g., Mondini and Elkin 2014), the obtained isotopic values in humans could have been the result of a diet rich in animal protein. However, they were not based exclusively on large herbivores and included some rodents and even locusts. In fact, taking into account values obtained in present-day studies in the Argentine Puna (Fernández and Panarello 1999–2001), the *Ctenomys* rodent presents an enriched value for carbon probably because of its feeding preferences in the Puna pastures. The carbon enriched values presented in this work would be consistent with the consumption of smaller fauna, especially Chinchillidae rodents, known to have been relatively more important – although always in a small proportion – in the Early Holocene than in later periods. In the Mid-Holocene, camelid exploitation became even more significant than before in Antofagasta de la Sierra, a trend also observed in other Puna localities (Mondini and Elkin 2014). In any case, this food item could have been less significant for some segments of the hunting–gathering population, for instance, infants and women. It is then crucial to study the isotopic signals of Chinchillidae specimens, both present and archaeological.

Conclusion

The aim of this work has been to discuss aspects of the palaeodiets of individuals living in the Antofagasta de la Sierra area in the Argentinian Puna during the final Early Holocene. Generally speaking, the values obtained are in agreement with the expected ones for the period, which has been characterised as the onset of an extensive arid period. We have also taken into account the fact that four out of the six individuals under study were infants.

In the first place, we could determine that the protein component of the diet was, at least for these individuals, isotopically more positive than the rest of the diet, indicating that these individuals preferred tuber consumption to that of *Opuntia* (with CAM pattern). Besides, this observation is consistent with the consumption of herbivores whose diet had a substantial amount of C_4 pastures. The reasons for this herbivores preference for such pastures will be the subject of further research.

In the second place, and bearing in mind that our interest is to understand the economic aspects of these hunting societies based on their diets, we have noticed that at least four individuals could have still been breastfeeding and close to weaning. This could explain the isotope values, especially those of nitrogen. However, the similarly high values of $\delta^{15}N$ in the juvenile and adult individuals lead us to think that aridity undoubtedly had a strong impact at the base of the food chain. This is especially so in the case of the 10-year-old child, in whom we expected depleted values due to the growing stage it was undergoing (for a discussion on how rapid growth rates in youth and adolescence affect nitrogen isotope ratios, see Water-Rist and Katzenberg 2010). The weight of the impact of water availability in our results is further reinforced by the presence of coeval fauna which also bear nitrogen-enriched values when compared to their present-day counterparts. That is to say, despite being individuals located chronologically at the point of transition, the values could still be associated with the changes attributed to the Mid-Holocene.

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