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### Using evidence from hair and other soft tissues to infer the need for and receipt of health-related care provision

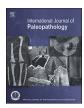
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# Using evidence from hair and other soft tissues to infer the need for and receipt of health-related care provision



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#### ABSTRACT

The Bioarchaeology of Care approach developed by Tilley is usually applied to skeletalized human remains, given the usual constraints of preservation bias that are seen with archaeological assemblages. However, other tissues, such as hair are sometimes preserved and can provide a wealth of information that can supplement the skeletal data. Archaeological hair has been analysed for drug compounds for almost thirty years. This article integrates data from hair analyses for coca metabolites, stable light isotope analysis and aDNA to expand the potential of the Bioarchaeology of Care approach using the example of a spontaneously mummified adult female from northern Chile.

#### 1. Introduction

The Bioarchaeology of Care approach developedby (Tilley, 2015a, 2015b; Tilley and Cameron, 2014; Tilley and Oxenham, 2011) has received much attention in recent years, as it provides a framework to identify and interpret incidences of care provision in pre-history. The provision of care to an individual may have improved quality of life and helped an individual live longer than if they were left untreated. This is an important consideration when investigating lifeways, as Tilley (2015b) states "Care provision is a conscious and purposive practice that involves caregiver(s) and care recipient(s), and it does not take place in a void. In any community, at any point in time, the perception of what constitutes 'health' and 'disease' and the related response to the care requirements of individual group members are shaped by a combination of cultural norms, values and belief systems; traditions; collective skills and experience; political, social and economic organisation; environmental variables; and access to resources". The interpretation of caregiving has the potential to extend our knowledge of ancient lifeways beyond just the treatment of an individual. Lorna Tilley (2015a, p. 3) notes that many diseases will never manifest in bone or preserved soft tissues; therefore, the full burden of disease in an individual and the frequency and features of caregiving response will remain unknown. Hair analysis may help address some of these issues. It is often well preserved in cases of spontaneous natural mummification in a range of different environments. It may also be found associated with skeletalized remains. Hair is largely composed of  $\alpha$ -keratin, which is largely resistant to biodeterioration except by keratinolytic

organisms (Wilson et al., 2007a,b,c; Wilson, 2008, 2017; Wilson and Tobin, 2010).

Drugs, hormones, metal ions and other compounds are incorporated into hair through a variety of different mechanisms (Henderson, 1993; Kronstrand and Scott, 2006). Biomolecular information is generally incorporated within the hair shaft as it hardens through the process of keratinisation and emerges from the follicle. This hardening of the hair shaft is responsible for capturing time-resolved information by ensuring that no further biogenic change can occur (Thompson et al., 2013; Wilson, 2005; Wilson and Tobin, 2010). Stable light isotopes offer valuable insight into palaeodietary reconstruction (Thompson et al., 2013; Wilson, 2005). Stable isotope analysis can further yield data on physiological and metabolic stress (Beaumont et al., 2015; Beaumont and Montgomery, 2016; Fuller et al., 2005, 2004; Petzke and Fuller, 2014; Petzke et al., 2010; Reitsema, 2013), as well as information on geography and migration (Ehleringer et al., 2008; Sharp et al., 2003).

The analysis of hair can thus provide a wealth of extra information to the bioarchaeologist in support of understanding the nutrition, health and well-being of individuals, as part of broader lifeways experience (see Table 1). In this article we present a case study of a spontaneously mummified adult female from Iquique, Chile who has evidence for both acute and chronic disease (Arriaza et al., 2008). We integrate chemical data from stable light isotope analysis and drug analysis within a Bioarchaeology of Care analysis. The strengths and limitations of using these data in a bioarchaeology of care analysis will be discussed.

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 Table 1

 Data that can be obtained from the analysis of archaeological hair samples.

Information	Type of analysis	Key References
Hygiene, grooming practices	Examination of parasites (e.g. headlice, pubic lice)	Araujo et al., 2000; Arriaza et al., 2012, 2013; Ashby, 2016; Fletcher, 1994; Raoult et al., 2008; Rick et al., 2002
Diet, trophic level, breastfeeding and weaning	Stable light isotope analysis $^2{\rm H}/^1{\rm H},~^{13}{\rm C}/^{12}{\rm C},$ $^{15}{\rm N}/^{14}{\rm N},~^{18}{\rm O}/^{16}{\rm O},$ and $^{34}{\rm S}/^{32}{\rm S}$	Britton et al., 2016, 2013; Brown and Alexander, 2016; de Luca et al., 2012; Jørkov and Gröcke, 2017; Knudson et al., 2015; Macko et al., 1999; Mora et al., 2017; Poulson et al., 2013; Roy et al., 2005; Williams and Anne Katzenberg, 2012; Wilson et al., 2007b)
Migration, altitude	Stable light isotope analysis - <sup>2</sup> H/ <sup>1</sup> H, <sup>13</sup> C/ <sup>12</sup> C, <sup>15</sup> N/ <sup>14</sup> N, <sup>18</sup> O/ <sup>16</sup> O, and <sup>34</sup> S/ <sup>32</sup> S	Cerling et al., 2006; Dupras and Schwarcz, 2001; Ehleringer et al., 2008; King et al., 2018; Sharp et al., 2003; White et al., 2009
Physiological stress (e.g. injury, infection, starvation)	Stable light isotope analysis - <sup>13</sup> C/ <sup>12</sup> C, <sup>15</sup> N/ <sup>14</sup> N; Analysis of hair for cortisol; Radiogenic isotopes	Beaumont et al., 2015, 2013; Beaumont and Montgomery, 2016; D'Ortenzio et al., 2015; Frei et al., 2017, 2015, Fuller et al., 2005, 2004; Petzke and Fuller, 2014; Petzke et al., 2010; Reitsema, 2013
Genetics – individual traits, maternal lineage	aDNA analysis – genomic and mitochondrial DNA	Bengtsson et al., 2012; Gilbert et al., 2008, 2007, 2004; Keller et al., 2012; Larsson and Tjälve, 1979; Moller et al., 1992; Orlando et al., 2015; Rasmussen et al., 2010
Exposure to pollutants	Wide variety of methods used to detect heavy metals e.g. arsenic, mercury compounds, lead	Arriaza et al., 2010; Byrne et al., 2010; Egeland et al., 2009, 1999; Fresnais et al., 2015; Kakoulli et al., 2014

#### 2. Hair analysis and coca chewing

Coca chewing is a uniquely Andean practice that is an important part of Quechua and Aymara identity to the present day (Allen, 2002, 1981). Coca (*Erythroxylum* sp.) is regarded as sacred and as a result is an important part of ritual offerings and sacrifices (Rätsch, 2005, p. 249; Wiedemann, 1979). Coca is also used as a medicinal treatment to treat digestive ailments, altitude sickness and mouth ulcers, since cocaine, which is present in coca leaves, is an effective topical anaesthetic (Biondich and Joslin, 2016; Weil, 1981, 1978). Coca is also used to suppress hunger and fatigue, and produces a sense of wellbeing and contentment (Bolton, 1976; Hanna, 1974; Plowman, 1986). Coca also has a complex social, ritual and political role in modern Andean communities, which is likely to be a continuation of a very long tradition (Allen, 2002, 1981; Naranjo, 1981; Weil, 1995).

Archaeological evidence suggests that coca chewing has extensive antiquity in South America. Coca leaves and calcite (an alkaline substance) have been found in domestic contexts dating to 8000 cal BP in the Nanchoc Valley in Northern Peru (Dillehay et al., 2010). Coca leaves have also been found in early coastal contexts, for example Engel (1963) found leaves he thought were coca along with deposits of burnt lime. Indirect evidence of coca chewing comes from figures with a characteristic bulge in the cheek (Donnan, 2007; Vidart, 1991) as well as the discovery of gourd-like containers used for storing lime (Lunniss, 2017; Stolberg, 2011). Coca leaves are also common grave offerings on the Peru/Chile coast from around 400 CE (Aufderheide, 2003).

Dental analysis of human remains from coastal Peru has indicated that habitual coca chewing was related to periodontitis and antemortem tooth loss (Indriati and Buikstra, 2001; Langsjoen, 1996; Pezo-Lanfranco et al., 2017; Turner, 1993).

The pathological changes associated with coca chewing consist of cervical root caries with root exposure on the buccal surfaces of posterior dentition (particularly 2nd and 3rd molars). Indriati and Buikstra

(2001) found that 87.5% of individuals with buccal root caries also had positive hair tests for BZE, a biomarker for coca use, suggesting a link between coca chewing and ante-mortem tooth loss.

The aetiology of the caries related to coca chewing is poorly understood. Indriati and Buikstra (2001) hypothesise that the anaesthetic qualities of cocaine metabolites released during the "chewing" of coca supresses the activity of the salivary glands around the coca quid. The reduction of the salivary flow results in a dry mouth, which is correlated with a higher frequency of carious lesions in a modern clinical context (Indriati and Buikstra, 2001; Murphy and Boza, 2012). Turner (1993) suggests that the mixture of coca leaves and lime caused damage to the alveolar bone.

The biomarkers of coca use include cocaine and benzoylecgonine (BZE). These compounds are incorporated into hair as it grows. Hair tests for cocaine have been routine in forensic cases for many years since commercially available radioimmunoassay (RIA) kits were first used to demonstrate the presence of cocaine and opiates in the hair of drug users (Valente et al., 1981). These commercial kits were used to first detect BZE in hair from archaeological sites in Peru and Chile (Cartmell et al., 1991a). The results of drug analyses of ancient hair from mummies from Peru, Chile and Argentina (Arriaza et al., 2008; Cartmell et al., 1994, 1991a; Cartmell et al., 1991b; Rivera et al., 2005; Springfield et al., 1993; Wilson et al., 2007b) has confirmed the widespread use coca in Andean communities over millennia in multiple regions and amongst different populations (Table 2).

#### 3. Stable light isotope analysis and dietary reconstruction

The use of stable light isotope analysis of archaeological hair is an established method of reconstructing past diets. In recent years increased attention has also been paid to the effect of physiological stress (e.g. pregnancy), starvation and malnutrition, since the metabolic balance of an individual can impact isotope ratios in tissues that are

**Table 2** A summary of findings – coca metabolites in mummy hair.

Culture	Approx. Time Period	Method of Analysis	Positive Tests	Reference
Chinchorro	3000 - 2000 BCE	RIA; GC-MS	0/23 (0%)	(Cartmell et al., 1994, 1991a,b; Cartmell et al., 1991a,b; Springfield et al., 1993)
Quiani	1500 - 1250 BCE	RIA; GC-MS	0/3 (0%)	
Alto Ramirez	350 - 250 BCE	RIA; GC-MS	1/3 (33%)	
Cabuza	400 - 1000 CE	RIA; GC-MS	10/16 (63%)	
Maitas Chiribaya	1000 -1250 CE	RIA; GC-MS	54/97 (56%)	
San Miguel	1050 - 1300 CE	RIA; GC-MS	2/8 (25%)	
"Inca"	1400 - 1500 CE	RIA; GC-MS	9/13 (69%)	
Alto Ramirez	1000 - 900 BCE	RIA; GC-MS	2/11 (18%)	(Rivera et al., 2005)
Cabuza	c. 1000 - 1300 CE	RIA	3/7 (43%)	(Arriaza et al., 2008)
Inca (ritual context)	1430 - 1520 CE	LC-MS/MS	3/3 (100%)	(Wilson et al., 2007b; 2013)
Alto Ramirez Cabuza	1000 - 900 BCE c. 1000 - 1300 CE	RIA; GC-MS RIA	2/11 (18%) 3/7 (43%)	(Arriaza et al., 2008)



**Fig. 1.** IQU-95 T3, an adult female, aged 40–50 at death. Recovered as part of a salvage project from a site near Iquique, Chile in 1995. Image courtesy of the late Professor Arthur Aufderheide.

forming during periods of metabolic stress (Baković et al., 2017; Beaumont et al., 2015, 2013; Beaumont and Montgomery, 2016; D'Ortenzio et al., 2015; Eerkens et al., 2017; Fuller et al., 2005; Mekota et al., 2009; Neuberger et al., 2013; Petzke and Fuller, 2014; Petzke et al., 2010). When a body experiences nutritional or physiological stress the body's response is to go into a catabolic state in which the body breaks down muscle protein (cachexia) for the body to use elsewhere to create proteins (D'Ortenzio et al., 2015). The ensuing result is increasingly negative  $\delta^{15}$ N values. The end result of nitrogen catabolism is the oxidation of amino acids to urea and uric acid, both of which are depleted in  $^{15}$ N with respect to the substrate amines (D'Ortenzio et al., 2015). As a result, the lighter  $^{14}$ N is preferentially excreted, with the remaining tissues becoming enriched in the heavier isotope, resulting in increased  $\delta^{15}$ N values (Katzenberg and Lovell, 1999).

#### 4. Case study

In 1995, the late Professor Art Aufderheide was involved in a rescue excavation of many spontaneously mummified individuals from sites near to Iquique, a town on the Chilean coast. These individuals were analysed by various methods and published in 2008 (Arriaza et al., 2008). These individuals exhibited a range of pathological changes that were visually apparent and confirmed by aDNA analysis. Here we discuss one of these individuals, known as IQU-95 T3, as a case study, showing how stable isotopic and biochemical data derived from hair can be used to augment palaeopathology data in a bioarchaeology of care analysis.

#### 5. Methods

#### 5.1. Drug analysis of hair

A 50 mg sample of hair was taken from a braid of hair belonging to IQU-95 T3, an adult female recovered from a rescue excavation that took place in 1995 near Iquique in northern Chile. The hair was manually brushed to remove external debris, then cut into small pieces. The sample was incubated with 1 ml 0.1 M Hydrochloric acid for 20 h at 45 °C (Valente et al., 1981). The samples were then filtered using a 3 ml Luer-lok syringe with a Minispike Acrodisc syringe filter (13 mm diameter; with GHP membrane; pore size 0.2  $\mu m$ , VWR International, Lutterworth, UK) that had previously been conditioned with LC–MS grade acetonitrile. The samples were blown down under nitrogen at 45 °C and re-constituted with 200  $\mu l$  LC–MS grade acetonitrile.

Chromatographic separation was achieved using a Waters 2695 separations module (Waters, Manchester, UK) interfaced with a Micromass Quattro Ultima triple quadrupole mass spectrometer (Waters, Manchester, UK), equipped with MassLynx 4.0 software. Ionisation was achieved using electrospray ionisation in positive ion mode (ES+).

The separation module was equipped with an Allure<sup>TM</sup> PFP Propyl column (Thames Restek, Buckinghamshire, UK). Column dimensions:  $30 \times 2.1$  mm, (Particle size:  $5 \mu m$ , pore size  $60 \, \mathring{A}$ ) with a  $10 \times 2.1$  mm Allure<sup>TM</sup> PFP Propyl guard cartridge (Thames Restek, Buckinghamshire, UK) fitted into a Trident<sup>TM</sup> 1 cm holder with in-line filters (Thames Restek, Buckinghamshire, UK).

Ion transitions were determined for several psychoactive compounds. For IQU-T3 there was only a signal for BZE (Ion transition 290 > 168; retention time  $0.79 \, \text{min}$ ).

#### 5.2. Isotope analysis of hair

Scalp hair samples were prepared for isotopic analysis using standard protocols. Samples were soaked overnight in 1:2 (vol/vol) chloroform/methanol, sonicated and rinsed three times in deionized water before lyophilization. Each shaft was oriented and cut into 15-mm serial segments working from the proximal end. Samples and internal standards were weighed into tin capsules for carbon and nitrogen isotope analyses, conducted using a ThermoFinnigan Delta Plus XP mass spectrometer and elemental analyser (EA/IRMS, elemental analyser/isotopic ratio mass spectrometer).

Data are reported in the conventional delta notation relative to internationally recognized standards: Pee Dee Belemnite ( $^{13}C_{V\text{-PDB}}$ ) for carbon; air ( $\delta^{-15}N_{AIR}$ ) for nitrogen (Wilson et al., 2007a,b,c; Thompson et al., 2013).

#### 6. The individual

IQU-95 T3 (see Fig. 1) was a naturally mummified adult buried in a flexed position, wrapped in fine layers of tightly woven textiles and wearing thick-soled sandals (Aufderheide, 1995). The sex of the individual was determined to be female on physical examination of the surviving external genitalia and morphology of the Os coxae. Age at death was estimated to be 40–50 years, based on age related change in the pubic symphysis, dental wear and the presence of osteophytes on the vertebrae. There was widespread preservation of soft tissues, including some viscera (both lungs, heart, liver, small and large intestine, bladder and brain), as well as skin and scalp hair. Aufderheide noted that the bones of this individual were unusually lightweight, suggestive of osteopenia (Arriaza et al., 2008).

There was extensive post-mortem damage to the facial area, with the nasal bones and maxilla completely absent. The mandible is present. All six molars of the mandible have been lost during life. No dental caries was present in the mandibular dentition. Aufderheide noted that there were no signs of porotic hyperostosis, cribra orbitalia or any other obvious markers of physiological stress. The lungs were still identifiable, despite other organs having completely decomposed. It was noted that there were pleural adhesions on the lungs, possibly from a bout of pneumonia that the individual had completely recovered from. Ancient DNA analysis ruled out the presence of tuberculosis in this individual (Arriaza et al., 2008).

Ancient DNA analysis of a tissue sample revealed that this individual would have lived with Chagas Disease (Arriaza et al., 2008). Chagas Disease is an insect vector borne parasitic disease caused by *Trypanosoma cruzi*. An estimated 6–7 million people today live with Chagas Disease in Latin America (WHO, 2017). Ancient DNA analyses suggest that around 40% of ancient populations from Chile and Peru were infected with *T. cruzi* (Aufderheide et al., 2004).

#### 7. Determination of disability

Chagas Disease has two phases; an acute phase and a chronic phase. Infection occurs when infected triatomine bugs puncture the skin to feed on blood and simultaneously deposit faeces or urine containing trypomastigotes of *T. cruzi* into the wound. In the acute phase general malaise is common, as is the enlargement of glands, the liver and spleen, followed by vomiting, diarrhoea, anorexia and fever. Meningoencephalitis with fever, convulsions, and/or loss of consciousness has also been associated with the acute phase of Chagas Disease (WHO, 2017, 2002).

After the acute phase ends, the infected individual enters an indeterminate form of the disease. Several studies of autonomic function have demonstrated changes in both the sympathetic and parasympathetic nervous system by damaging motor neurones. Cardiac abnormalities are also evident in some asymptomatic individuals with the chronic indeterminate form of Chagas disease (WHO, 2017, 2002).

Chagasic cardiomyopathy is the most significant clinical manifestation of chronic Chagas Disease. It is present in approximately 10–30% of modern patients. The most common cardiac complications of chronic Chagas Disease are left ventricular dilation and dysfunction, aneurysm, congestive heart failure, blood clots, abnormal heart rhythm, and sudden cardiac death (Rassi et al., 2010). Typical symptoms of congestive heart failure include lower limb oedema, feeling of weakness, chest pain and dizziness. Enlargement of the heart because of damage to cardiac muscle can cause pulmonary embolisms and embolisms in other organs, usually the brain, spleen, and kidneys (Malik et al., 2015; Rassi et al., 2010, 2007).

Of particular note is the fact that infection by T. cruzi can result in the destruction of the autonomic enteric innervation that leads to the dysfunction of the digestive system. The initial main symptom, almost always present, is difficulty swallowing. As the disease progresses this is followed by thoracic pain, active and passive regurgitation, heartburn, hiccups, cough, excessive production of saliva, enlargement of the salivary glands and emaciation (WHO, 2002). The paralysis of the digestive system can result in megaoesophagus and megacolon. The typical symptoms of megaoesophagus include difficulty swallowing and regurgitation of food. Malnutrition is a complication of megaoesophagus. Symptoms of megacolon include constipation, abdominal pain, and excessive gas, with impacted calcified faeces (faecaloma) in extreme cases (Oliveira et al., 1998). The physical manifestations of Chagas - cardiomegaly, megaoesophagus and megacolon with impacted coprolites have been documented in spontaneously mummified individuals from the Andean region (Aufderheide et al., 2004; Reinhard et al., 2003; Rothhammer et al., 1985).

Based on the age at death of IQU-95 T3, Arriaza et al. (2008), suggest that this individual had probably lived for a long period with Chagas Disease, so was probably in the chronic stages of the disease. Most of the internal organs in this individual had completely decomposed or were not sufficiently preserved, so it is not possible to determine whether there were any physical manifestations of Chagas Disease that may indicate what symptoms this individual may have

had. Modern clinical observations have indicated that difficulty swallowing (dysphagia) is extremely common, but oesophageal abnormalities are only observable radiologically in 7-10% of people living with chronic Chagas Disease (Oliveira et al., 1998). Given what we know about Chagas Disease in modern cases, at the very least IQU-95 T3 would have likely had difficulty swallowing, and pain on swallowing, as well as intermittent stomach cramps, diarrhoea and regurgitation. The effect of dysphagia and regurgitation can result in weight loss and malnutrition (Bastien, 2003; Guevara et al., 1997; Oliveira et al., 1998). Bastien worked with rural communities in Bolivia living with the burden of Chagas' Disease. He described it as "a debilitating disease at all stages [which] creates a downward spiral of productivity (Bastien, 2003, p. 168). It is very likely that IOU-95 T3 suffered from intermittent periods of sickness relating to Chagas' Disease over a period of years which may have made it difficult to fully participate in community life. This individual does exhibit markers of occupational activity, namely occupational deformation of the mandibular dentition and bilateral flattening of the femoral condyles, which Aufderheide suggests are squatting facets (Aufderheide, 1995) so was likely contributing in some way although this activity may pre-date infection or have occurred during an asymptomatic phase of the disease. Given the ubiquity of Chagas' Disease in modern South American populations and what has been found in palaeopathological studies, her illness would have not been unusual and there would likely have been a proportion of the community with Chagas symptoms.

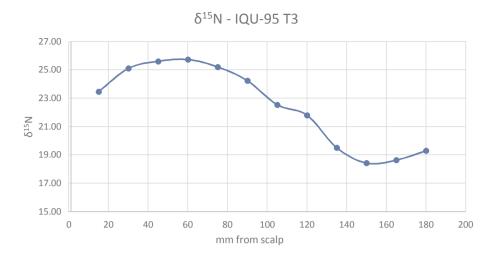
The physical examination of the bones of this individual and the isotopic analysis of hair hint at the possible metabolic state of this individual. As indicated above, the individual may have had osteopenia, a generic description that identifies the occurrence of a disproportionate amount of bone mass (Ortner, 2003). A wide range of diseases, hormonal conditions, malnutrition and early childbirth and breastfeeding can all contribute to the development of osteopenia (Brickley and Ives, 2008; Curate, 2014; Ortner, 2003). For IQU-95 T3 the presence of osteopenia suggests a history of physiological stress.

Carbon and nitrogen stable isotope values reflect dietary input and metabolic or physiological stress at the time of formation (see Fig. 2). Stable light isotope analysis of the hair from IQU-95 T3 revealed increasing nitrogen values starting  $\approx$  8 months prior to death, from 19.5‰, to a peak of 25.7‰,  $\approx$  4 months prior to death, a shift of  $\approx$  6‰. High nitrogen values such as these are not unusual in this particular region, due to marine dietary input (Díaz-Zorita Bonilla et al., 2016; King et al., 2018) and the use of guano as manure in agriculture (Poulson et al., 2013; Szpak et al., 2012).

In the same time period, the carbon values become more negative, from – 11.9‰  $\approx$  9 months prior to death to – 15.6‰  $\approx$  3 months before death. Whilst dietary input may account for the change in  $\delta^{15} N$ and  $\delta^{13} C$  values, it is important to note that similar trends have been observed in nutritionally stressed adults (Beaumont and Montgomery, 2016). Segmental analysis of hair from people who had died as a result of starvation showed that  $\delta^{15}N$  values rise as body mass index fell, and that  $\delta^{13}$ C values to be in phase with BMI. This observed drop in  $\delta^{13}$ C was thought to be the result of the breakdown of fat deposits, which are approximately 3% lower than other body tissues such as muscle (Beaumont and Montgomery, 2016; Neuberger et al., 2013). Significantly elevated nitrogen levels have also been observed in adult female burials from Sudanese Nubia with osteopenia; given what is now known about metabolic processes it is likely these signatures are reflecting underlying health problems (White and Armelagos, 1997). Given that Chagas Disease commonly has gastrointestinal complications which can result in malnutrition, the isotopic signatures observed in the hair of IQU-95 T3 could indicate a period of illness rather than merely a dietary change.

#### 8. Model of care

Chagas Disease has been described as being disabling at every stage



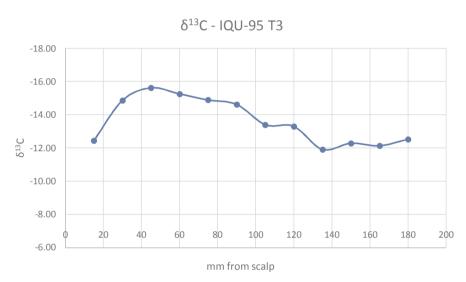


Fig. 2.  $\delta^{15} N$  and  $\delta^{13} C$  values from segmental hair analysis of IQU-95 T3.

of the disease (Bastien, 2003). In the case of IQU-95 T3 it is not possible to tell what stage of Chagas Disease she was in when she died. It is possible she was in the chronic phase based on her advanced age at death (Arriaza et al. (2008). However, the viscera that show changes consistent with chronic Chagas Disease (heart, colon) were either poorly preserved, or did not survive at all. However, only 30% of chronically infected people develop cardiac changes and 10% develop associated conditions such as megacolon (WHO, 2017). Consequently, the absence of these in a naturally mummified individual does not preclude them having lived with Chagas Disease for many years prior to death.

In a modern context, Chagas Disease remains a disease of poverty, with high morbidity and mortality in the Americas and increasingly around the world as a result of migration (Dias et al., 2002). Palaeopathological work has shown that there was a high incidence of Chagas Disease in ancient populations, so we can deduce that IQU-95 T3 would have probably exhibited symptoms that were not unusual in her community, given the chronic nature of this condition. Periods of sickness would have required care from either her family or extended community, such as aiding sanitation, and perhaps providing different, more digestible food and care during bouts of sickness.

Chemical analyses show that she was consuming coca in some form before she died. Qualitative results from the LC–MS/MS analysis indicated the presence of BZE in the hair of this individual. An earlier

radioimmunoassay of a hair sample from IQU-95 T3 also revealed coca metabolites (Arriaza et al., 2008). Given what we know about the therapeutic value of coca, it is likely that this was chewed to help with some of the gastrointestinal side effects of Chagas Disease. However, it is also possible that she had chewed coca for an extended time over the course of her life, as suggested by the loss of all six molars. Placing the results of the biomolecular analysis in this context helps us interpret the role of coca in past communities and opens up new research questions into direct methods of investigating and contextualising the use of psychoactive plants by ancient communities.

Coca is widely reported from funerary contexts in this region, despite not growing on the coast today. and has a traditional use in treating gastrointestinal complaints (Biondich and Joslin, 2016; Weil, 1995, 1981, 1978) for which it would have probably been fairly effective, given that cocaine is an effective anaesthetic. Only bulk analysis of a hair sample was carried out, so it is not possible to speculate on the timing of coca ingestion. However, it should be noted that IQU-95 T3 had lost all six molars, a pattern seen in habitual coca chewers (Indriati and Buikstra, 2001). It is not possible to infer that coca would have only been used to treat the gastrointestinal symptoms of Chagas Disease, given the importance and symbolism of coca in traditional lifeways to both ancient and modern Andean communities.

#### 9. Interpretation

Chemical analyses, including that of scalp hair has broadened the picture significantly; we can see that this individual lived with Chagas Disease, a debilitating parasitic infection that would likely have affected around 40% of the people in her community, based on prevalences detected in ancient populations from this region (Aufderheide et al., 2004). Stable light isotope analyses of hair indicate a period of physiological stress prior to death. Arriaza et al. (2008) noted that there were no obvious signs of physiological stress, but the hair analysis further support to the likely experience of this particular individual. Periods of illness such as those experienced with Chagas Disease can be debilitating and would have required at least some longstanding support from her community.

#### 10. Conclusion

The traditional mummy autopsy approach used by Aufderheide (Arriaza et al., 2008; Aufderheide, 1995) revealed that this individual had lightweight vertebrae, suggestive of osteopenia and healed lung adhesions. Other than the lungs, the viscera were not well preserved, preventing further interpretation of the history of morbidity for this individual.

Hair can survive in a wide range of depositional contexts. With carefully considered research questions, hair and other soft tissues can provide a huge amount of valuable information. Given that the sampling of hair is minimally invasive, it offers an important adjunct to complement traditional osteology and palaeopathology when using incremental stable isotope analysis, drug analysis and whole genome sequencing. When the osteological analyses and results of hair analyses are integrated into the bioarchaeology of care framework the combined results have potential to offer a very detailed biography of the health of an individual alongside other contextual evidence from archaeological, historical, social and political perspectives.

We can use this approach to help understand the role that individuals such as IQU-95 T3 would have had within their society – by examining their health status, how they were cared for, and what kind of medicinal or surgical interventions they may have had. These findings continue to help us understand how societies view long term illness and disability. Increased awareness of these issues will help researchers to further develop and refine such research questions and priorities.

#### Acknowledgement

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