

# Fisherfolk and Farmers: Carbon and Nitrogen Isotope Evidence from Middle Horizon Ancón, Peru

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**ABSTRACT** This study investigates human dietary patterns and economic trends at the coastal site of Ancón, Peru during the Andean Middle Horizon (550AD–1000AD) using stable isotopic data from 32 individuals buried at the site.  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  results from human bone collagen and  $\delta^{13}\text{C}$  from human tooth enamel and bone carbonate indicate that inhabitants consumed a mixed diet composed primarily of marine protein and  $\text{C}_4$  resources, with only marginal reliance on  $\text{C}_3$  foods. Over time, Ancóneros appear to have relied more heavily on  $\text{C}_4$  resources, particularly maize, despite the fact that the crop could not have been grown locally. These results are notable given that  $\text{C}_3$  rather than  $\text{C}_4$  or marine foods dominate the site's archaeological record. These data suggest that Ancón's inhabitants either had access to more fertile land up-valley where maize could be cultivated successfully or that they engaged in trade relationships with their valley neighbours. A third possibility is that increased maize consumption at Ancón during the Middle Horizon resulted from Wari imperial influence and interregional exchange. Comparisons of  $\delta^{13}\text{C}$  values in enamel and bone carbonate from Ancón individuals indicate that  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values are significantly more positive than  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values. This suggests that the diets of young children were systematically enriched in  $^{13}\text{C}$  compared to that of adults, perhaps as a result of nursing activity and/or differential dietary practices among various age groups at the site. Copyright © 2009 John Wiley & Sons, Ltd.

*Key words:* carbon isotopes; nitrogen isotopes; Ancón; Wari; Andes

## Introduction

Carbon and nitrogen isotope analyses of archaeological skeletal remains are based on the well-tested premise that isotopic ratios in human bones and teeth will reflect the isotopic composition of an individual's diet (DeNiro & Epstein, 1978, 1981; Schoeninger & DeNiro, 1984). Carbon and nitrogen isotope ratios vary by food category and these differences are recorded in human body tissue. Archaeologists capitalise on this principle to reconstruct dietary patterns and subsistence strategies among past human populations (Hobson & Collier, 1984; Farnsworth *et al.*, 1985; Ambrose & DeNiro, 1986; Walker & DeNiro, 1986; Muldner & Richards, 2005; Keenleyside *et al.*, 2006; Bocherens *et al.*, 2007), investigate human evolutionary trends (Lee-Thorp &

van der Merwe, 1987; Lee-Thorp *et al.*, 1994; Schoeninger, 1995), and document the emergence of social inequality in the past (Schurr & Schoeninger, 1995; Ubelaker *et al.*, 1995).

Carbon and nitrogen isotope analyses are employed here to determine ancient diet of a subset of Middle Horizon (550AD–1000AD) individuals from the coastal site of Ancón, Peru. Numerous archaeologists have noted a significant decrease in marine shell and fish remains at the site during the Middle Horizon compared to earlier and later time periods and have suggested that seafood was no longer consumed locally to the degree that it had been in the past. Instead, they argue that marine resources were exported to sites further inland, perhaps as a result of increased trade and exchange during the Middle Horizon.

Despite more than a century's worth of research at Ancón and the recovery of more than 3000 human burials (Kaulicke, 1997), this hypothesis has yet to be tested using skeletal remains. The current study provides the first isotopic evidence for Middle Horizon

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diet at Ancón through an analysis of 32 individuals interred at the site. Given the sharply delineated nature of resource zones in the Andes (Murra, 1975; Tomczak, 2003), the dietary data generated in this investigation should provide a refined sense of the potential economic networks that may have existed between inland areas and Ancón in ancient times.

## The site of Ancón

The archaeological site of Ancón is located 40 km north of Lima, on Peru's Central Coast (Figure 1). Ancón is desert-like in appearance, but sits only a few metres from the resource-rich Bay of Ancón. Based on the extensive presence of ancient shell middens, fish remains, fish hooks and fishing nets at the site (Corbett, 1951; Willey & Corbett, 1954; Uhle, 1968 [1912]), Ancón's economy appears to have been primarily maritime-based for much of its history.

Despite easy access to seafood, ancient Ancóneros apparently consumed terrestrially grown foods as well. Remains of  $C_3$  plants and fruits such as lúcuma (*Pouteria lucuma*), quinoa (*Chenopodium quinoa*), maní (*Arachis hypogaea*), avocado (*Persea americana*) and beans have been identified at Ancón (Dorsey, 1894; Bonavia, 1962; Muelle & Ravines, 1973; Menzel, 1977; Leon del Val, 1994; Kaulicke, 1997), along with small amounts of maize, a  $C_4$  plant (Dorsey, 1894; Bonavia, 1962; Menzel, 1977; Leon del Val, 1994; Kaulicke, 1997).



Figure 1. Map of Ancón, Peru and other sites mentioned in text.

The bulk of these foods, particularly corn, likely were imported from the neighbouring Chancay and Chillón River Valleys since there was not enough water at the site for intensive cultivation (Uhle, 1968[1912]; Menzel, 1977; Kaulicke, 1997). Terrestrial protein may have contributed to ancient diet as well, as llama (*Lama glama*) and guinea pig (*Cavia porcellus*) remains have been found in archaeological deposits (Uhle, 1968[1912]; Kauffmann Doig, 1994).

Ancón's initial settlements span the Andean Pre-ceramic through Early Horizon periods (2250 BC-200 BC) (Muelle & Ravines, 1973), and were concentrated on the hilly slopes beyond the modern town of Ancón. The early economy appears to have revolved around marine exploitation as evidenced by extensive shell middens from this time, some of which reached depths of over 8 m (Corbett, 1951; Willey & Corbett, 1954; Matos Mendieta, 1968).

Sometime during the Middle Horizon economic and social practices among ancient Ancóneros changed. Shell and fish remains, once dominant at the site, are rare in Middle Horizon occupation refuse (Uhle, 1968[1912]; Menzel, 1977; Ravines, 1977). Ancón's hillside settlements are abandoned and occupation is re-established in the Necropolis of Ancón—a large, flat plain north of the modern village that contained thousands of human burials as well as occupation refuse, house posts, wall foundations and other cultural debris (Uhle, 1968[1912]). Ancón mortuary practices change, too, from relatively simple, single interments with few grave goods to elaborate burials in deep chambered tombs (Ravines, 1977, 1981; Kaulicke, 1997; Segura, 1997). Among Middle Horizon funerary patterns, there is a marked display of wealth absent in earlier tombs and an increased presence of imperial-style objects associated with the highland Wari empire (Menzel, 1977; Ravines, 1977; Kaulicke, 1997; Slovak, 2007).

Many scholars interpret these changes as evidence for increased trade and exchange between Ancón and the Wari polity during the Middle Horizon, and Ancón often is depicted as intensively involved in the harvesting and exportation of marine resources (Menzel, 1977; Ravines, 1977). Such activities certainly might explain the changes in economic practices that have been documented at the site during this time, namely the marked decrease in shellfish debris compared to earlier and later time phases and the notable increase in foreign-style artifacts. Intensive trade and exchange also might have affected the diets of ancient Ancóneros themselves. If marine foods were increasingly being funneled out of the local community, Ancón's inhabitants may have relied more heavily on terrest-

rially grown foods and/or animal protein and less upon marine resources. In order to assess whether ancient Ancóneros' diets reflect this pattern, this study employs carbon isotope analysis of enamel and bone carbonate and carbon and nitrogen isotope analysis of human bone collagen.

### Stable isotope analysis and dietary reconstruction

Carbon and nitrogen isotope values ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , respectively) have been used to assess the relative amounts of marine and terrestrial foods in prehistoric diets (DeNiro & Epstein, 1978, 1981; Schoeninger & DeNiro, 1984; Walker & DeNiro, 1986; Ubelaker *et al.*, 1995). Carbon isotope ratios vary significantly between plants that possess the  $\text{C}_3$ ,  $\text{C}_4$  or CAM photosynthetic pathway.  $\text{C}_3$  plants exhibit  $\delta^{13}\text{C}$  values between  $-22$  and  $-35\text{‰}$  with an average value of  $-26.5\text{‰}$ , while  $\text{C}_4$  plants have  $\delta^{13}\text{C}$  values ranging from  $-9$  to  $-16\text{‰}$  with a mean value of  $-12.5\text{‰}$  (van der Merwe & Tschauner, 1999). Plants that fix  $\text{CO}_2$  by the Crassulacean acid metabolism (CAM) pathway have  $\delta^{13}\text{C}$  values that fall in between those for  $\text{C}_3$  and  $\text{C}_4$  plants (Katzenberg, 1992). Humans who consume these plants retain a record of the  $\delta^{13}\text{C}$  signature of their food source, which is recorded in human tissue.

The carbon isotope composition of human tissue is systematically enriched compared to  $\delta^{13}\text{C}$  values in diet (DeNiro & Epstein, 1978). Carbon isotope values in bone collagen and carbonate respectively will be 4‰ and 11‰ more positive than the  $\delta^{13}\text{C}$  (‰) in plants consumed (Koch *et al.*, 1994). Additionally, there is a fractionation factor in collagen values related to trophic level such that carnivore  $\delta^{13}\text{C}$  collagen values are approximately 1.5‰ more positive than the prey they consume (van der Merwe & Tschauner, 1999).

Carbon isotope values from bone collagen reflect the lifespan of carbon atoms in collagen, which may be as long as 25 years or equal to the lifetime of the individual (White & Schwarcz, 1994). Collagen, which constitutes 90% of the organic component of bone, is comprised of amino acids that derive from ingested food (Chisholm *et al.*, 1982), and preferentially reflects the protein component of human diet (Ambrose, 1993; Lee-Thorp & Sponheimer, 2003). In contrast, carbonate in bone mineral derives from blood bicarbonate which, in turn, reflects total dietary intake, including protein, carbohydrates and lipids (Ambrose, 1993; Ambrose & Norr, 1993).

The offset between carbonate and collagen carbon isotope values ( $\Delta^{13}\text{C}_{\text{carb-coll}}$ ) has been shown to yield

important dietary information. Initial studies by Krueger & Sullivan (1984) suggested that offsets varied depending on whether an individual consumed an omnivorous, carnivorous or herbivorous diet. Additional research generally confirmed this pattern, although an overlap in offset values was noted among certain classes of carnivores and herbivores (Lee-Thorp *et al.*, 1989). Later laboratory experiments by Ambrose & Norr (1993) demonstrated that offsets tended to vary according to whether individuals consumed a mono-isotopic diet (when carbon isotope ratios among protein and non-protein components of the diet are the same) or a polyisotopic diet (when carbon isotope ratios among protein and non-protein components of the diet differ). Animals who consumed protein sources with less negative  $\delta^{13}\text{C}$  values than the remainder of their diet yielded  $\Delta^{13}\text{C}_{\text{carb-coll}}$  values less than 4.4‰, while those individuals who consumed protein resources with more negative  $\delta^{13}\text{C}$  values than their whole diet had  $\Delta^{13}\text{C}_{\text{carb-coll}}$  values greater than 4.4‰ (Ambrose & Norr, 1993; Ambrose *et al.*, 1997).

While this latter approach is informative when the components of ancient and modern diet are known, research by Kellner & Schoeninger (2007) has demonstrated that drastically different diets (e.g. pure  $\text{C}_3$  diets versus marine/ $\text{C}_4$  diets) can produce identical  $\Delta^{13}\text{C}_{\text{carb-coll}}$  values. In order to reduce ambiguity in the reconstruction of ancient diet, Kellner & Schoeninger (2007) advocate the use of a carbon isotope model based on three regression lines, where each line corresponds to a protein source ( $\text{C}_3$ ,  $\text{C}_4$  or marine) and an individual's placement on each line indicates energy source ( $\text{C}_3$ ,  $\text{C}_4$  or mixed). Based on the results of their study (Kellner & Schoeninger, 2007), it appears that a bivariate plot of  $\delta^{13}\text{C}_{\text{collagen}}$  and  $\delta^{13}\text{C}_{\text{apatite}}$  values more accurately reflect ancient dietary composition than using  $\delta^{13}\text{C}_{\text{collagen}}$ ,  $\delta^{13}\text{C}_{\text{apatite}}$  or  $\Delta^{13}\text{C}_{\text{carb-coll}}$  values alone.

In instances where prehistoric human populations had access to marine foods and  $\text{C}_4$  plants,  $\delta^{13}\text{C}$  values alone are inadequate for determining prehistoric diet because marine and  $\text{C}_4$  foods yield indistinguishable  $\delta^{13}\text{C}$  values (Schoeninger & DeNiro, 1984; Schoeninger, 1995). In these cases, nitrogen isotopes have been used to discriminate between marine and terrestrial food sources.

Organisms are grouped into three major divisions based on their nitrogen isotope ratios: nitrogen fixing plants ( $\text{N}_2$ -fixing), terrestrial plants that utilise nitrates from non-atmospheric sources, and marine foods that are not based on  $\text{N}_2$  fixation (Price *et al.*, 1985). In almost all instances, marine vertebrates have higher  $\delta^{15}\text{N}$  values than terrestrial vertebrates at similar

trophic levels, permitting the differentiation between marine versus  $C_4$  protein in human and non-human animal diets (Schoeninger & DeNiro, 1984; Walker & DeNiro, 1986). On average, human  $\delta^{15}N$  collagen values among ancient maize consumers range between 9 and 10‰, while those for marine eaters and/or fisher-gatherers range between 14 and 15‰ (Schoeninger *et al.*, 1983; DeNiro, 1987). Low trophic level marine organisms such as shellfish however, have significantly lower  $\delta^{15}N$  values than marine vertebrates. Isotopic values from South African species of mussels and other filter feeders yielded  $\delta^{15}N$  values ranging from 7.1–9.5‰ (Sealy *et al.*, 1987). Shellfish-based diets, therefore, will have noticeably different  $\delta^{15}N$  values from those based primarily on fish or marine mammals—a factor which should be accounted for in dietary reconstructions.

## Materials and methods

Thirty-two adult individuals from the Necropolis of Ancón were analysed in the current study. The sample comprised skeletons from three projects: Cirilo Huapaya Manco's 1947 excavations sponsored by the Museo Nacional de Antropología y Arqueología, Luis Ccosi Salas and Marino Gonzalez's 1950–1953 Inspección de Monumentos Arqueológicos project, and Federico Kauffmann Doig's Proyecto Tumbas de Ancón in 1994. The sample represents those individuals that securely could be dated to the Middle Horizon based upon stratigraphic and stylistic associations. Additionally, only adult specimens were selected for analysis, since one of the additional goals of our analysis was to compare childhood and adult strontium isotope signatures from the same individual, the results of which are presented in a separate publication (Slovak *et al.*, 2009).

The sample was divided according to time period: Early Middle Horizon (EMH) and Late Middle Horizon (LMH) burials, which correspond to distinct Middle Horizon occupation phases at the site. EMH burials are found in an earlier, deeper stratum than LMH burials and are associated with Nieveria, Chakipampa and imperial Wari ceramic styles, which traditionally date to the first half of the Middle Horizon (Menzel, 1964). LMH tombs were separated from their earlier counterparts by a thick layer of culturally sterile, alluvial soil and often contained pottery and textiles bearing Teatino and Huara influences—two cultural traditions associated with the latter Middle Horizon on the Central Coast (Bonavia, 1962). In total, 9 EMH

skeletons and 23 LMH skeletons were included in the present study.

Age and sex estimation for the Ancón skeletons was based on standard osteological parameters (Phenice, 1969; Acsadi & Nemeskeri, 1970; Meindl & Lovejoy, 1985; Brooks & Suchey, 1990; Buikstra & Ubelaker, 1994) and the results are reported in Table 1. Approximately 2 g of cortical bone and 10–15 mg of tooth enamel were collected from the majority of skeletons for carbon and nitrogen isotope analysis ( $n = 20$ ). In some cases post-cranial material was missing or in very poor condition and only tooth enamel was collected ( $n = 5$ ), while in others very little tooth enamel remained and only bone samples were taken ( $n = 7$ ). Bone samples were taken from the mid-diaphyseal shaft of individuals' long bones, while enamel samples were collected from either the third, second or first molar in that order depending on the condition, placement and integrity of the tooth.

Bone and enamel samples initially were abraded using a Dremel Multipro drill (Model 395) outfitted with a 0.5 mm inverted cone tip in order to remove adhering matter and superficial contamination. Collagen was extracted from 27 human bone samples following the method outlined in Tuross *et al.* (1988) and summarised in Koch *et al.* (1994), whereby 0.5N HCl was used to demineralise the bone. Acid was changed every 2 days until the reaction ceased and a white or yellow collagen replica of the bone specimen remained. Three of the samples yielded no collagen (specimens CF-14-III, A1-1333 and A1-2257). The remaining 24 samples were transferred to acid-cleaned microcentrifuge tubes and washed to neutrality with distilled water. Samples were freeze-dried and ground to a loose powder for further analysis.

Bone samples from 21 individuals were prepared for carbon isotope analysis of bone carbonate. Powdered bone was soaked in 2 ml of diluted  $H_2O_2$  (3%), left overnight, and rinsed and sonicated with deionised water. This process was repeated daily until organics had been removed and each sample had stopped bubbling. Enamel from 25 Ancón skeletons was prepped for analysis following the procedures of MacFadden & Cerling (1996) and Koch *et al.* (1997). Briefly, samples were treated with 1 ml of  $H_2O_2$  (30%), left overnight, then sonicated with deionised water. Samples were soaked in 1 ml of 0.1N acetic acid for 24 h, rinsed twice with distilled water and dried down completely.

Bone and enamel samples were analysed at the Stanford Stable Isotope Laboratory. Collagen samples were converted to  $CO_2$  and  $N_2$  gases using a Carlo Erba NA 1500 Elemental Analyzer. The resulting  $CO_2$  and  $N_2$  gases were transferred to a Finnigan Delta plus IRMS via a Finnigan ConFloII interface and analysed.

Table 1. Ancón stable carbon, nitrogen and oxygen isotope data

Burial #	Time period	Sex	Age (years)	Tooth	Bone	$\delta^{13}\text{C}_{\text{carb\_enamel}}$	$\delta^{13}\text{C}_{\text{carb\_bone}}$	$\delta^{13}\text{C}_{\text{coll}}$	$\delta^{13}\text{C}_{\text{carb-coll}}$	$\delta^{15}\text{N}$	C/N	$\delta^{18}\text{O}$
A1/P6136	EMH	M	20–34	RM <sub>3</sub>	Tibia	-5.6		-11.2		15	3.37	-7.6
A1/P6504	EMH	M	20–34		Humerus			-10.8		14.8	3.29	
A1/P6677	EMH	M	35–49		Radius		-7.9	-12.2	4.3	13	3.31	
A1/P6865	EMH	F	35–49	RM <sub>3</sub>	Humerus	-4.3		-9.6		14.9	3.31	-7.8
A1/P7854	EMH	M	50+	RM <sup>3</sup>	Humerus	-4.2		-8.5		14.7	3.22	-7.9
A1/P7968	EMH	F?	20–34	RM <sub>2</sub>	Tibia	-5.3	-6.5	-12.4	5.9	15	3.4	-7.2
A1/P8247	EMH	F?	15–19		Radius		-4.4	-10.6 <sup>a</sup>	6.6 <sup>a</sup>	16.4 <sup>a</sup>	3.72	
A1/P8375	EMH	M	20+	LM <sub>3</sub>		-5						-8.8
A1/P8406	EMH	F	20–34	LM <sup>2</sup>	Humerus	-3.7	-5.3	-10.8 <sup>a</sup>	5.5 <sup>a</sup>	17.1 <sup>a</sup>	3.8	-5.4
A1/1333	LMH	M	20–34	LM <sub>3</sub>	Radius	-4.6	-6.6					-7.1
A1/1345	LMH	F	35–49	LM <sup>2</sup>	Tibia	-4.2	-4	-9.2	5.2	14.5	3.34	-7.0
A1/1461	LMH	?	50+		Ulna			-10.7		13.6	3.44	
A1/1509	LMH	?	15–19	RM <sub>2</sub>	Tibia	-3.9	-4.3	-10.4	6.1	14	3.59	-6.5
A1/1554	LMH	?	35–49	RM <sup>1</sup>		-3.2						-7.1
A1/1569	LMH	F	35–49		Fibula		-4.5	-10.1 <sup>a</sup>	5.5 <sup>a</sup>	14.1 <sup>a</sup>	3.75	
A1/1572	LMH	?	20–34	LM <sub>2</sub>	Fibula	-5.6	-5.5	-9.9	4.3	14.1	3.37	-6.9
A1/1645	LMH	M	35–49	LM <sup>3</sup>	Radius	-3.7	-5.4	-11.3 <sup>a</sup>	6.0 <sup>a</sup>	16.9 <sup>a</sup>	4.67	-7.2
A1/1703	LMH	F	35–49	LM <sub>3</sub>	Fibula	-4.7	-6.7	-11.2	4.5	13.1	3.4	-7.1
A1/1939	LMH	F?	20+	RM <sup>2</sup>	Radius	-4.6	-5.9	-11.0 <sup>a</sup>	5.1 <sup>a</sup>	15.6 <sup>a</sup>	3.8	-7.3
A1/1979	LMH	F	20–34		Humerus			-10.7		13.4	3.42	
A1/2250	LMH	F?	20–34	LM <sub>3</sub>		-1.8 <sup>b</sup>						-7.8 <sup>b</sup>
A1/2257	LMH	M?	20–34	LM <sub>3</sub>	Humerus	-3.9 <sup>b</sup>	-5.3					-7.2 <sup>b</sup>
A1/2271	LMH	?	15–19	LM <sup>2</sup>	Radius	-3.7	-4.2	-10.2 <sup>a</sup>	6.0 <sup>a</sup>	16.7 <sup>a</sup>	4.14	-6.5
A1/P7224	LMH	?	20–34	LM <sub>2</sub>		-4						-7.1
CF-03-II	LMH	F?	20–34	LM <sub>3</sub>	Fibula	-3.7	-4.9	-10	5.1	13.5	3.34	-6.6
CF-08-I	LMH	M?	20–34	RM <sub>3</sub>	Fibula	-4.2	-5.4	-10.1	4.7	14.9	3.33	-7.4
CF-08-II	LMH	M?	35–49		Radius		-4.8	-10.2	5.4	14.1	3.31	
CF-08-Y	LMH	F	50+	LM <sup>2</sup>	Tibia	-3.1	-4.9	-10.5	5.5	14	3.28	-6.4
CF-14-I	LMH	F	35–49	RM <sub>3</sub>	Humerus	-6.4 <sup>b</sup>	-6.4	-11.8 <sup>a</sup>	5.4 <sup>a</sup>	14.5 <sup>a</sup>	4.13	-8.6 <sup>b</sup>
CF-14-III	LMH	F	35–49	LM <sup>3</sup>	Radius	-5	-6.3					-7.6
CF-14-X	LMH	F	50+	RM <sub>2</sub>	Fibula	-6.1	-6.1	-11	4.9	13.3	3.29	-7.8
CF-18	LMH	M	15–19	RM <sup>2</sup>		-2.7						-5.9

<sup>a</sup> Results excluded from study because C/N ratios fall outside of acceptable atomic range.

<sup>b</sup> Results excluded from study because samples were unbalanced during analysis.

Analytical precision for nitrogen and carbon in collagen is  $\pm 0.2\text{‰}$  and  $\pm 0.3\text{‰}$ , respectively.

Enamel and bone samples were acidified with two drops of 100% purified phosphoric acid. Oxygen was converted to  $\text{CO}_2$  and analysed on a Finnigan MAT 252 IRMS interfaced with a Finnigan MAT Kiel III carbonate device. A series of standards of known isotopic composition also were run through the carbonate device to assure proper reference gas calibration and sampling protocol. Precision for carbon isotopes in bone carbonate was 0.05 and 0.1‰ in tooth enamel.

As reported elsewhere (Slovak *et al.*, 2009), a random subset of the Ancón bone samples ( $n = 5$ ) were monitored for diagenetic contamination of bone mineral using Fourier transform infrared (FTIR) spectroscopy. FTIR measures the absorption of infrared radiation by a bone or tooth sample at its molecular level and permits an assessment of the overall mineralogy and crystallinity of skeletal materials (Sillen, 1989; Shemesh, 1990; Wright & Schwarcz, 1996; Hoppe *et al.*, 2003).

Results from this study (Slovak *et al.*, 2009) indicate that the carbonate composition of the Ancón samples remained unaltered and resembled that of modern bone.

Diagenetic alteration among the collagen samples was monitored using C/N ratios (DeNiro, 1985). Collagen samples with C/N ratios above or below the 2.9–3.6 range established for modern bone were excluded from analysis.

## Results

As discussed above, the mineral component of the Ancón bone samples appears pristine. The organic phase of bone, however, is more susceptible to diagenetic alteration. Seven of the twenty-four bone collagen samples yielded C/N ratios higher than the acceptable range found in modern bone (2.9–3.6) (DeNiro, 1985) (Table 1). While four of these samples are relatively close to the normal range of values (C/

$N = 3.75\text{--}3.8$ ), three of the samples yielded C/N ratios greater than 4.1. These seven individuals were excluded from the current analysis. Additionally, based on data quality criteria three of the enamel samples (CF-14-I, A1-2257, and A1-2250) yielded unreliable carbon carbonate isotope values. While these values are not included in the present analysis, all of the carbon results are reported in Table 1.

In total, reliable carbon isotope ratios from enamel ( $\delta^{13}\text{C}_{\text{carb\_enamel}}$ ) and bone carbonate ( $\delta^{13}\text{C}_{\text{carb\_bone}}$ ) were obtained for 22 and 21 individuals, respectively and reliable carbon and nitrogen isotope values from collagen ( $\delta^{13}\text{C}_{\text{coll}}$  and  $\delta^{15}\text{N}$ ) were produced for 17 skeletons (Table 1). All carbon and nitrogen results are reported in per mil (‰) notation relative to PDB Belemnite, the standard for carbon (Chisholm *et al.*, 1982), and AIR, the standard for nitrogen (Mariotti, 1983).

## Carbon

$\delta^{13}\text{C}_{\text{carb\_enamel}}$  values among the Ancón sample range from  $-2.7$  to  $-6.1$ ‰ with a mean of  $-4.3$ ‰.  $\delta^{13}\text{C}_{\text{carb\_bone}}$  results have a mean value of  $-5.5$ ‰ and a range of  $-4.0$  to  $-7.9$ ‰. Results of the *t*-test indicate that  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  and  $\delta^{13}\text{C}_{\text{carb\_bone}}$  are significantly different (*t* test,  $p < 0.0002$ ) (Figure 2). Given that

tooth enamel forms during an individual's childhood (Hillson, 1996) while bone mineral regenerates throughout an individual's lifetime (Parfitt, 1983), these results suggest that the diets of young children were enriched in  $^{13}\text{C}$  compared to that of adults.

$\delta^{13}\text{C}_{\text{carb\_enamel}}$  and  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values do not vary significantly by sex. Additionally,  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values do not differ significantly by time period, although admittedly the number of Early Middle Horizon (EMH) individuals is quite small ( $n = 4$ ) compared to the number of LMH individuals ( $n = 17$ ). Nonetheless there does appear to be greater dietary variation in the first half of the Middle Horizon than the later half, with  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values ranging from  $-4.4$  to  $-7.9$ ‰ in the EMH and  $-4.0$  to  $-6.7$ ‰ during the LMH (Figure 3)

$\delta^{13}\text{C}_{\text{coll}}$  values among Ancón individuals vary from  $-8.4$  to  $-12.4$ ‰ with a mean value of  $-10.5$ ‰. As with the  $\delta^{13}\text{C}_{\text{carb}}$  results, there is no significant difference in  $\delta^{13}\text{C}_{\text{coll}}$  values by sex or time period, although there is a greater range of  $\delta^{13}\text{C}_{\text{coll}}$  values during the EMH ( $-8.4$  to  $-12.4$ ‰) than during the LMH ( $-9.2$  to  $-11.2$ ‰) (Figure 4).

Comparisons of  $\delta^{13}\text{C}_{\text{coll}}$  and  $\delta^{13}\text{C}_{\text{carb\_bone}}$  from the same individuals show that the two are significantly correlated, i.e. as  $\delta^{13}\text{C}_{\text{coll}}$  values increase so do  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values ( $R^2 = 0.719$ ,  $p < 0.001$ ). The mean

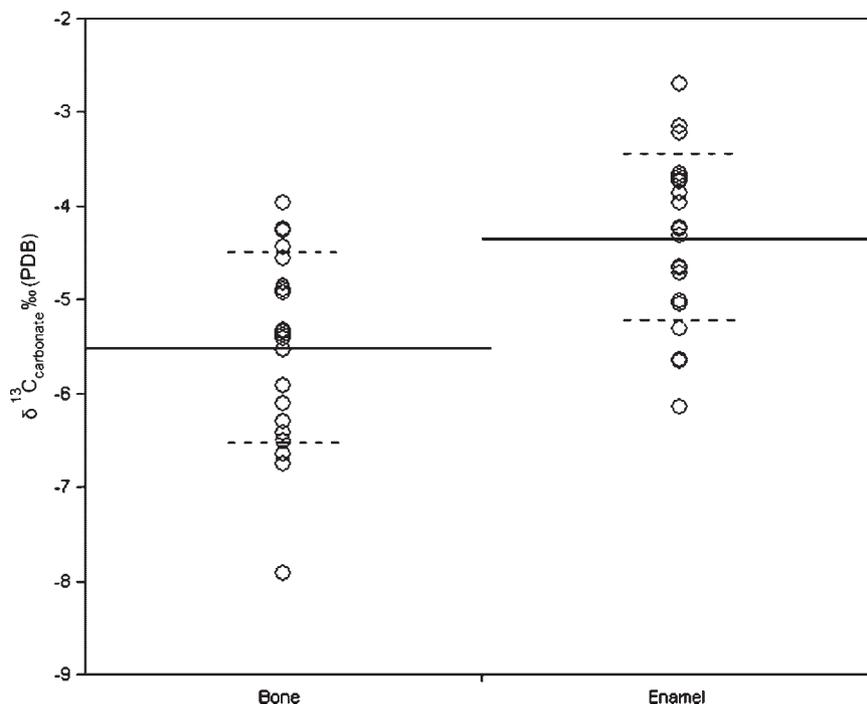


Figure 2.  $\delta^{13}\text{C}$  values from bone and enamel carbonate for Ancón individuals. The solid lines indicate the mean values for bone and enamel and the dotted lines indicate one standard deviation from these means.

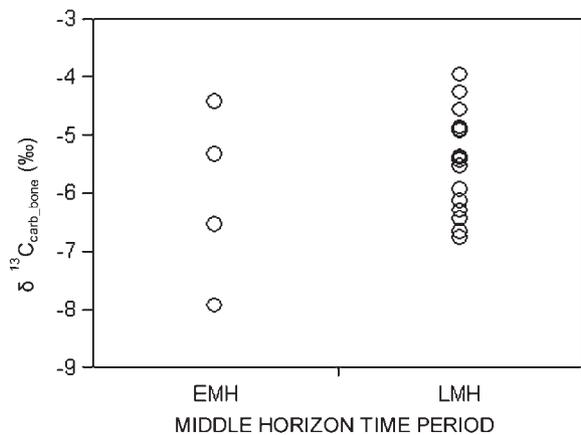


Figure 3.  $\delta^{13}\text{C}$  values from bone carbonate for Ancón individuals plotted by time period (EMH, Early Middle Horizon; LMH, Late Middle Horizon).

difference between  $\delta^{13}\text{C}_{\text{carb\_bone}}$  and  $\delta^{13}\text{C}_{\text{coll}}$  values ( $\Delta^{13}\text{C}_{\text{carb\_coll}}$ ) among the Ancón sample is 5.1‰ with a range of 4.3 to 6.1‰, indicating that the majority of ancient Ancóneros likely consumed protein resources with more negative  $\delta^{13}\text{C}$  values than the remainder of their diet, as would be expected among populations whose diets consisted of  $\text{C}_4$  plants in addition to marine and/or  $\text{C}_3$  proteins (Tomczak, 2003; Finucane *et al.*, 2006).

When plotted along Kellner & Schoeninger's (2007) regression line model (Figure 5), it is clear that the majority of isotope values cluster around the marine protein line, which is not unexpected given Ancón's proximity to the sea. However, some values plot closer to the  $\text{C}_4$  protein line, suggesting that these inhabitants' diets included significant amounts of  $\text{C}_4$

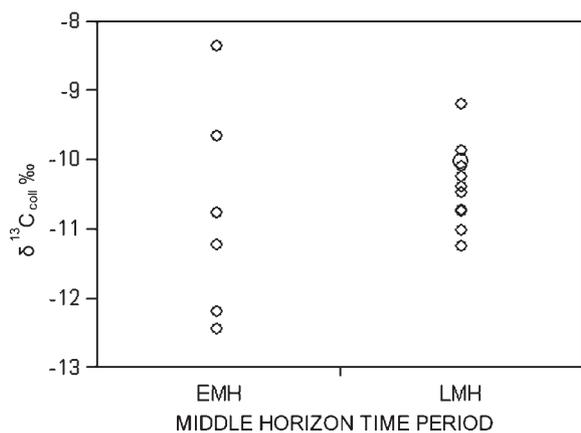


Figure 4.  $\delta^{13}\text{C}$  values from bone collagen for Ancón individuals plotted by time period (EMH, Early Middle Horizon; LMH, Late Middle Horizon).

protein as well. Additionally, dietary energy appears to have come predominantly from  $\text{C}_4$  sources. This is somewhat surprising given that the bulk of paleobotanical evidence documented at the site represents  $\text{C}_3$  rather than  $\text{C}_4$  foods (Dorsey, 1894; Bonavia, 1962; Muelle & Ravines, 1973; Leon del Val, 1994; Kaulicke, 1997), and indicates that site inhabitants may have relied heavily on maize despite the fact that this crop could not have been grown locally (Uhle, 1968[1912]; Menzel, 1977; Kaulicke, 1997).

## Nitrogen

Nitrogen isotope ratios from bone collagen are reported in Table 1 and vary from 13.0 to 15.0‰ with a mean value of 14.1‰. These values fall within the range of marine consumers in the Andes and elsewhere (see e.g., Schoeninger *et al.*, 1983; DeNiro, 1987; Ericson *et al.*, 1989; Tomczak, 2003), and support the conclusions drawn from the carbon isotope results above. Ancón's inhabitants appear to have consumed seafood throughout the Middle Horizon, although an ANOVA test reveals a statistically significant difference ( $p \leq 0.05$ ) in mean  $\delta^{15}\text{N}$  values between EMH and LMH populations (Figure 6). With the exception of one individual, all of the EMH inhabitants exhibit  $\delta^{15}\text{N} \geq 14.5\text{‰}$  ( $n = 6$ ) while the majority of the nitrogen isotope values among LMH individuals fall below 14.0‰ ( $n = 11$ ). Additionally, there is greater variability in  $\delta^{15}\text{N}$  values among the LMH sample than among EMH individuals, particularly if the one EMH outlier is excluded (Figure 6). The decline in  $\delta^{15}\text{N}$  over time coupled with greater variability in  $\delta^{15}\text{N}$  values during the LMH suggest that site inhabitants may have relied less on marine protein and increasingly supplemented their diets with various amounts of  $\text{C}_4$  plants and terrestrial protein.

## Discussion

The carbon and nitrogen isotope data from bone collagen and bone carbonate suggest that ancient Ancóneros consumed a mixed diet of marine protein and  $\text{C}_4$  foods throughout the Middle Horizon, with only a marginal reliance on  $\text{C}_3$  resources. High nitrogen isotope values among all individuals indicate a clear dependence on seafood; however in some instances, site inhabitants also relied on  $\text{C}_4$  protein potentially derived from maize and/or maize-fed animals such as llamas (see e.g. Finucane *et al.*, 2006). Additionally, it appears that most individuals relied on  $\text{C}_4$  energy sources rather than  $\text{C}_3$  sources (Figure 5). As noted

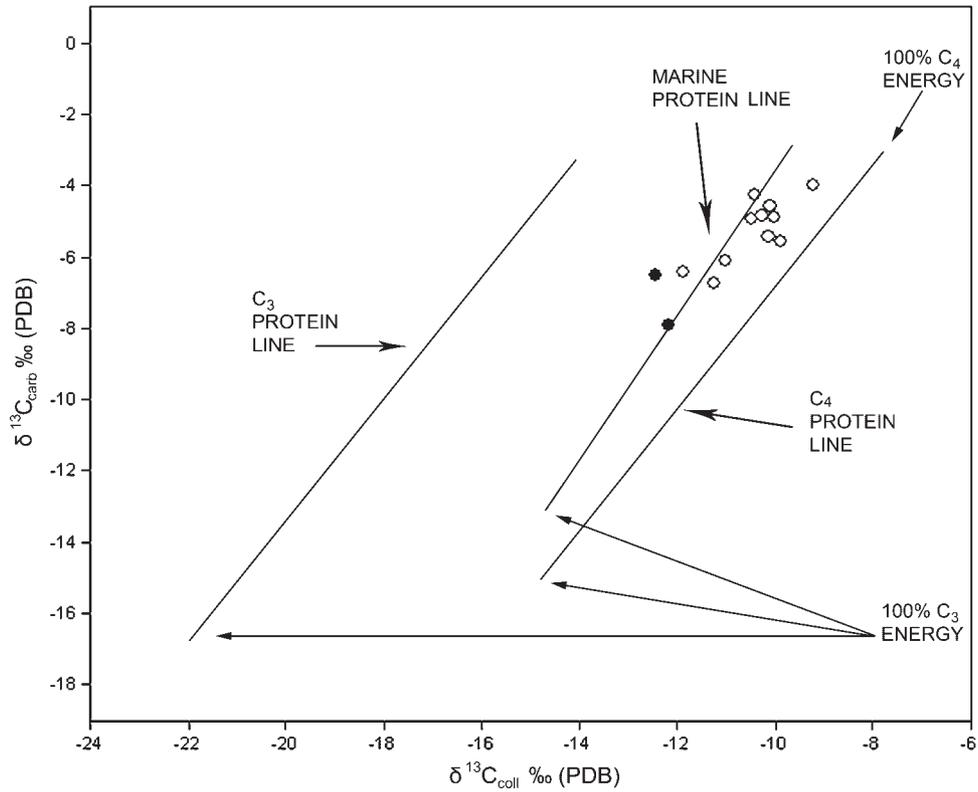


Figure 5.  $\delta^{13}\text{C}_{\text{carb}}$  plotted against  $\delta^{13}\text{C}_{\text{coll}}$  using Kellner & Schoeninger's (2007) 3-regression line model. Dark circles correspond to Early Horizon Ancón skeletons; light circles correspond to Late Middle Horizon Ancón skeletons.

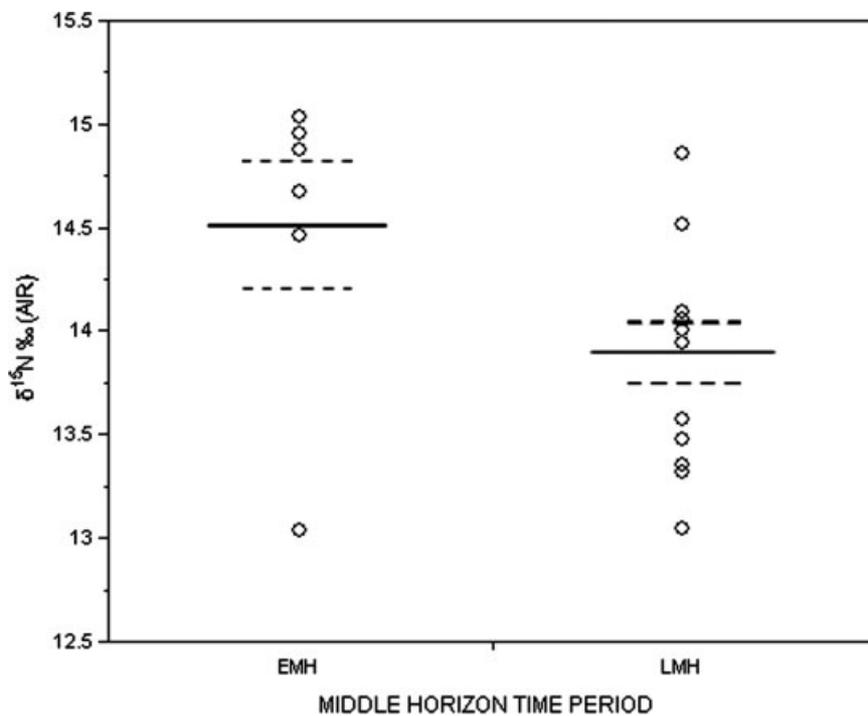


Figure 6.  $\delta^{15}\text{N}$  values from bone collagen for Ancón individuals plotted by time period (EMH, Early Middle Horizon; LMH, Late Middle Horizon). The solid lines indicate the mean  $\delta^{15}\text{N}$  values for both time periods and the dotted lines indicate one standard deviation from these means.

earlier, this pattern contrasts with paleobotanical evidence from the site, which is dominated by the presence of C<sub>3</sub> foods such as *lúcuma*, quinoa, *maní*, avocado and beans (Dorsey, 1894; Bonavia, 1962; Muelle & Ravines, 1973; Leon del Val, 1994; Kaulicke, 1997).

On the other hand, the isotopic evidence accords well with dental data collected from the Ancón skeletons (Slovak, 2007). Of the 32 individuals analysed here, 27 skeletons display at least one carious lesion and of these seven exhibit at least one dental abscess. The increased presence of dental caries and abscesses among human populations has been linked to a greater reliance on fermentable carbohydrates, particularly corn (Larsen, 1983; Navia, 1994). The near ubiquitous presence of dental caries among the Ancón sample suggests that the majority of Ancóneros consumed maize throughout the Middle Horizon, as suggested by the carbon isotope results.

The degree to which C<sub>4</sub> foods were incorporated into Ancón diet appears to have varied over time, however. During the EMH,  $\delta^{13}\text{C}_{\text{carb\_bone}}$  and  $\delta^{13}\text{C}_{\text{coll}}$  values are widely dispersed (Figures 4 and 5), while  $\delta^{15}\text{N}$  values remained relatively homogenous (Figure 6). With the exception of one outlier, all of the individuals have  $\delta^{15}\text{N}$  signatures that cluster between 14.5 and 15‰.

The variation in  $\delta^{13}\text{C}_{\text{carb\_bone}}$  and  $\delta^{13}\text{C}_{\text{coll}}$  values during the EMH potentially indicates that some individuals in the population had increased access to maize while other members of the population consumed this plant to a limited degree. This pattern may reflect varied dietary practices or preferences of individual consumers, but it may also reflect socio-economic or status differentiation at the site. It is notable that the individual with the highest  $\delta^{13}\text{C}_{\text{coll}}$  value (−8.5‰) in the entire sample irrespective of time period was interred in the only EMH tomb designated as elite based on a variety of cultural criteria (Slovak, 2007), while the remainder of EMH individuals included here were found in non-elite tombs. The relative uniformity in  $\delta^{15}\text{N}$  values suggest that, unlike maize, marine resources were equally distributed throughout the population. Furthermore, the overwhelmingly positive  $\delta^{15}\text{N}$  values likely indicate that Ancón's EMH population derived most of its dietary protein from marine foods, rather than from other sources.

During the LMH, the carbon and nitrogen isotope patterns are reversed.  $\delta^{13}\text{C}_{\text{carb\_bone}}$  and  $\delta^{13}\text{C}_{\text{coll}}$  values fall closely together and are enriched in  $^{13}\text{C}$  while  $\delta^{15}\text{N}$  signatures are variable and less positive relative to EMH isotope values. The increase in  $\delta^{13}\text{C}$  values over time as well as their relative homogeneity during the LMH indicate that C<sub>4</sub> resources, especially maize, were widely consumed. The relatively high  $\delta^{15}\text{N}$  values

exhibited by most LMH individuals demonstrates that marine foods continued to contribute to ancient diet; however the statistically significant decrease in  $\delta^{15}\text{N}$  signatures from the EMH to the LMH indicates that dietary protein likely came from a mixture of C<sub>4</sub> and marine foods.

The reliance on corn as the primary energy, and in some cases protein, component of Ancón diet, particularly during the LMH, is surprising given that like many other crops it could not be grown at the site owing to lack of arable land and fresh water. It is possible that ancient Ancóneros either had access to fertile land several kilometres up-valley where they could grow maize themselves or were involved in intense trade relationships with their valley neighbours. It is also possible that site inhabitants were involved in a large-scale interregional trade network, one that may have included the powerful Wari empire.

As discussed earlier, the economic and cultural character of Ancón's Middle Horizon settlement is unique, particularly when compared to earlier and later occupations at the site. Fish and shellfish remains are scarce in Middle Horizon strata, funerary practices transform, and Wari cultural influences appear. Furthermore, recent research using strontium isotope analysis of Middle Horizon Ancón skeletal material has highlighted the presence of at least one probable migrant from the Wari/Ayacucho area at this time (Slovak *et al.*, 2009). It is possible that ancient Ancóneros' marked dependence on maize throughout the Middle Horizon is indicative of Wari-Ancón economic interaction.

The intensification of maize cultivation and consumption during times of state and empire expansion has been documented throughout Andean prehistory (Morris, 1979; Hastorf, 1990; Hastorf & Johannessen, 1993), including the Middle Horizon. Schreiber (1992), for example, has argued that one of the primary motivations behind Wari's expanding empire was increased access to maize. Finucane *et al.* (2006) documented that Wari individuals residing at the highland site of Conchopata consumed almost exclusively maize or maize-fed animals during the Middle Horizon, while Cook & Glowacki (2003) have argued that maize consumption, particularly in the form of *chicha* or beer, was integral to Wari feasting events. It is possible that in exchange for marine resources, Ancón was incorporated increasingly into the Wari economic and cultural sphere of influence. This explanation could account for the overall increase in  $\delta^{13}\text{C}$  values and decline in  $\delta^{15}\text{N}$  values over time and accords well with the presence of Wari style artifacts and relative absence of marine debris in the site record during the Middle Horizon.

It is also plausible, however, that Ancón individuals' increased dependence on maize over time was prompted by reasons independent of imperial interaction. In a typical year the cool Humboldt Current runs northward along Peru's coast, creating nutrient-rich water conducive to marine life. During El Niño events, however, unusually warm surface ocean temperatures and seasonal meteorological fluctuations cause marine life to head south in search of cooler water. In recent years, the El Niño phenomena has wrecked havoc on the Peruvian fishing industry and has negatively affected marine mammal survival rates in the area (Limberger, 1990; Wyrski *et al.*, 1976). It's likely that severe El Niño episodes in the past had similar adverse consequences for ancient coastal dwellers and sea life (DeVries, 1987).

Strong El Niño events often bring with them periods of intense rainfall, increased temperatures and severe flooding (Goldberg *et al.*, 1987; Quinn *et al.*, 1987). Two such floods, one around 500 AD and the other around 1100 AD, have been documented on Peru's north coast, and have been explained as the result of catastrophic El Niño events (Nials *et al.*, 1979a, 1979b). While little is known about the effects of El Niño at Ancón, severe flooding episodes dating to the latter half of the Middle Horizon have been documented at the site (Dorsey, 1894; Carrion Cachot, 1948; Patterson, 1966; Uhle, 1968[1912]). It is possible that Ancón's inhabitants experienced a number of El Niño-related environmental changes during the LMH, including an overall decrease in the type and abundance of marine life present in the Bay of Ancón. Such an event would have forced Ancóneros to rely less heavily on marine foods and depend more on terrestrial resources both for energy and dietary protein, which in turn would result in the overall decrease in  $\delta^{15}\text{N}$  values and increase in  $\delta^{13}\text{C}$  values discussed earlier.

In addition to changes in dietary patterns over the course of the Middle Horizon, carbon and nitrogen isotope data from the site of Ancón indicate that diet varied within the lifetime of the individual. Enamel carbonate values from Ancón individuals are significantly higher than bone carbonate values, suggesting that the diets of young children and juveniles were enriched in  $^{13}\text{C}$  compared to that of adults (Figure 2). A number of paleodietary studies have documented a similar decline in  $\delta^{13}\text{C}$  values from infancy to adulthood, albeit in bone collagen rather than enamel and bone carbonate (Katzenberg *et al.*, 1993; Dupras *et al.*, 2001; Richards *et al.*, 2002; Fuller *et al.*, 2006; Dupras & Tocheri, 2007; Prowse *et al.*, 2008). According to these studies, enriched  $\delta^{13}\text{C}_{\text{coll}}$  values among infants and young children likely were due to either a trophic level

effect associated with breastfeeding or to the increased consumption of  $\text{C}_4$ -based foods such as maize or millet gruel during the weaning process.

Previous studies that have focused on carbon isotope values in enamel carbonate have noted a somewhat different trend. Rather than decreasing over time from infancy to adulthood as was the case with  $\delta^{13}\text{C}_{\text{coll}}$ ,  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values tended to increase from infancy to late childhood. Among their study of Kaminaljuyú skeletons, Wright & Schwarcz (1998) discovered that  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values in later-developing permanent teeth (specifically premolars and third molars) were significantly higher than  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values in early forming teeth. Similarly, Dupras and Tocheri (2007) noted a statistically significant increase in carbon apatite values in later-forming teeth (i.e. premolars and second molars) than in earlier-forming teeth (i.e. incisors, canines and first molars) among a sample of juvenile and adult individuals from Kellis, Egypt. Both sets of authors (Wright & Schwarcz, 1998; Dupras & Tocheri, 2007) linked these elevated values to a potential shift in diet from one based exclusively on breast milk to a diet that was increasingly supplemented with solid foods.

Unfortunately it is not possible to directly compare the results of the current sample with those from Kaminaljuyú or Kellis, as the current study lacks carbon isotopic data from early forming permanent dentition. All of the teeth but one in the present study represents either second or third molars. Enamel formation in second molars occurs around 2–3 years of age and is completed by 7 years of age, while the third molar initially forms between 7 and 13 years of age (Hillson, 1996). There is no statistically significant difference between second and third molar mean  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values ( $-4.26\text{‰}$  and  $-4.50\text{‰}$ , respectively) in the present sample. If the introduction of solid foods to the diets of Ancón children is responsible for the elevated  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values, then this supplementation likely occurred sometime around 2 years of age, and continued through late childhood (or between the time of second and third molar enamel formation).

That said, the difference in  $\delta^{13}\text{C}_{\text{carb\_enamel}}$  values noted in the current study is not between deciduous and permanent dentition nor between early forming versus late-forming dentition; rather the difference is between late-forming teeth and bone from adult individuals. This suggests that the overall composition of an individual's diet shifted sometime between childhood and adulthood. It may be that children were fed a maize-based weaning gruel, as has been suggested for populations elsewhere (see e.g. Wright & Schwarcz, 1998; Katzenberg *et al.*, 1993). Such a diet

would result in elevated  $\delta^{13}\text{C}$  values among individuals owing to the plant's low lipid content and overall  $^{13}\text{C}$  enrichment (Wright & Schwarcz, 1998).

In addition to carbon isotope data, oxygen isotope signatures ( $\delta^{18}\text{O}$ ) were generated for the Ancón individuals studied here and potentially shed light on childhood dietary practices at the site (Table 1). Oxygen isotope ratios in an individual's skeletal material primarily reflect the isotopic composition of the water that an individual consumes during his or her lifetime (Longinelli, 1984; Koch *et al.*, 1989), which in turn reflects a specific ecological environment (Dupras & Schwarcz, 2001). Oxygen isotopes from local water sources are incorporated into enamel phosphate and carbonate via body water at the time of mineralisation in childhood (Luz *et al.*, 1984). While breastfeeding, an infant receives all of the water that he or she needs from a mother's breast milk, which is significantly enriched in  $^{18}\text{O}$  compared to drinking water (Dupras and Tocheri, 2007). As a result, tissues that form during the time that an infant and child breastfeeds display more positive  $\delta^{18}\text{O}$  signatures than those tissues that form after breastfeeding has ceased. A number of archaeological studies have capitalised on this principle to investigate breastfeeding, complimentary feeding and weaning practices among past populations (Wright & Schwarcz, 1998; White *et al.*, 2004; Dupras & Tocheri, 2007; Turner *et al.*, 2007).

A comparison of  $\delta^{18}\text{O}$  values between second and third molars in the Ancón sample demonstrates that the former type of tooth is significantly more positive than the latter type (*t* test,  $p < 0.01$ ). On average, second molars are 0.8‰ heavier than third molars in  $^{18}\text{O}$ . While there is some variability in  $\delta^{18}\text{O}$  values among individuals, overall children between the ages of 2 and 6 years consumed water from a source enriched in  $^{18}\text{O}$ , while older children (7 years+) derived water from sources that were isotopically lighter in  $^{18}\text{O}$ . The most probable explanation for the discrepancy in  $\delta^{18}\text{O}$  values between second and third molars is that younger children received some of their water intake via maternal breast milk while older children did not. Instead, these latter individuals likely consumed water from springs at the site.

While the oxygen isotope data do not bear directly on the carbon isotope results presented here, they do suggest that Ancón children were weaned off of breast milk sometime between second and third molar enamel formation. During the weaning period children likely would have been fed a special weanling diet, which may have consisted primarily of maize gruel as discussed above. Over the lifespan of an individual, diet likely diversified to include  $\text{C}_4$ ,  $\text{C}_3$  and marine

foods. The latter two food sources are less enriched in  $^{13}\text{C}$  than  $\text{C}_4$  plants, and probably contributed to the overall decrease in  $\delta^{13}\text{C}_{\text{carb\_bone}}$  values that were present among adults in the sample. Ultimately it remains unclear whether the difference in mean  $\delta^{13}\text{C}_{\text{carb}}$  values between children and adult isotope signatures at Ancón is the result of breastfeeding and weaning practices, different diets among age groups, or to some other effect. Additional research on Ancón skeletons, particularly infants and young children, as well as the inclusion of isotopic data from deciduous and early forming permanent teeth may help to clarify the issue further.

## Conclusion

At the start of this paper, it was suggested that the significant decrease in marine remains documented at Ancón during the Middle Horizon corresponded to a decline in the amount of seafood consumed by site inhabitants. It was further suggested that this shift in diet resulted from increased trade and exchange between Ancón and inland communities, with Ancón serving as an important centre for marine harvesting and exportation and potentially under the political sway of the Wari empire. Based on the isotope results reported here, the first half of the hypothesis appears tenable. Fluctuations in  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values between early and later Middle Horizon populations suggest that terrestrial and marine resources were consumed to varying degrees over time. Elevated  $\delta^{13}\text{C}_{\text{coll}}$  and  $\delta^{13}\text{C}_{\text{carb}}$  signatures and significantly lower  $\delta^{15}\text{N}$  values among LMH individuals relative to EMH inhabitants indicate that maize represented an increasingly important dietary staple at Ancón, while marine foods were consumed to a considerably lesser degree.

While a change in diet over time at Ancón is apparent, the reason behind such a shift remains unclear. The high consumption of maize, particularly during the LMH, suggests that the crop increasingly was brought into the community, potentially as a result of Wari imperial influence, trade with lower-lying inland communities, or as a result of El Niño-induced climatic stress. In order to more adequately assess the range of possibilities, however, pre- and post-Middle Horizon Ancón skeletons should be analysed. Additionally, carbon and nitrogen isotope ratios from local Ancón flora and fauna must be determined to further refine the interpretations presented here.

Finally, differences in  $\delta^{13}\text{C}_{\text{carb}}$  values between enamel and bone from the same individual indicate that children consumed a diet enriched in  $^{13}\text{C}$  compared to adults.

Based on oxygen isotope data from the Ancón skeletons, this difference is likely related to a special weanling diet, although research must be conducted on infant and juvenile Ancón skeletons to evaluate this possibility further. In conclusion, this study has clarified aspects of dietary practices at Ancón and shed light on potential trade and political networks that may have functioned between the central sierra and coastal communities during the Middle Horizon.

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