Domesticate Animal Herding and Procurement Strategies, and the Childhood Origin of the Human Mandible (SP1) from the Bronze Age Site of Punta di Zambrone, Calabria, Italy: a Laser Ablation Sr Isotope Study

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Abstract: Strontium isotope analysis of human and faunal enamel has the potential to reveal human childhood origins, and herding and other movements of animals. Recent developments have included laser ablation sampling which dramatically increases the spatial resolution at which strontium isotopes can be measured. This allows time series of changes in strontium isotopes to be measured along the growth axis of enamel, giving a highly time-resolved measure of human and animal movement. Here we present the method and some examples of its application including our work attempting to identify the provenance of humans at the Bronze Age harbour site of Punta di Zambrone (possible inhabitants of the settlement or foreigners related to its destruction?) and to identify the extent of the agricultural catchment of this settlement through analysis of cattle, pig and other animal teeth. The Sr isotope results show a faunal catchment of approximately 15km, stretching from the coast to the inland high plain, and suggest that the individual SP1 was most likely local to the region.

Keywords: Animal teeth, human teeth, laser ablation, Early Bronze Age Calabria, Punta di Zambrone, Recent Bronze Age Calabria, Strontium isotope analyses

Introduction and Aims

Here we present the laser ablation Sr isotope analysis of tooth enamel of various fauna and a human mandible (SP1) from the Early and Recent Bronze Age layers at Punta di Zambrone.

The coastal settlement of Punta di Zambrone is located on the Poro promontory in the western part of southern Calabria. Marco Pacciarelli and his local collaborators have investigated this geomorphologically very diverse region for several decades by means of archaeological surveys and excavation campaigns.⁷ The results of these activities allow the reconstruction of the settlement history of this specific micro-region in detail and form the background against which we can evaluate the results of the three-year excavation project centred at Punta di Zambrone.⁸ In the northern

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⁷ Pacciarelli 2000, 72–85.

⁸ For an overview on this project, see Jung and Pacciarelli in the present volume.

section of the Recent Bronze Age (RBA) fortification ditch we excavated a sequence of ashy fill layers very rich in artefacts, animal bones and charred botanical remains. In addition, these layers yielded dispersed human bone fragments that could be partially recomposed and belong to two different individuals, one of which retains its mandible with three molars (M1–M3) and a canine.⁹ The find position of these bones is a secondary one, as they had been deposited together with the ashy sediment in the fortification ditch in an intentional backfill at the end of the settlement's use. Earlier, the ashes had accumulated elsewhere – most probably close to the fortification wall, over an extended period while the settlement was inhabited.¹⁰ Consequently, we can use the contents of the ashy layers to reconstruct the Recent Bronze Age history of the site. Early Bronze Age (EBA) finds come from the fill of a long rock-cut feature running close and almost parallel to the RBA ditch.

We aim to determine the animal management strategies of the site (e.g. importation vs local husbandry) and to determine if SP1 was 'local' to his/her place of burial or had spent his/her childhood (to the age of about 14) in an area of differing strontium geology.

Strontium isotopes (measured as ⁸⁷Sr/⁸⁶Sr) are passed unchanged into tooth enamel via the diet, preserving the mean ⁸⁷Sr/⁸⁶Sr of the parent rocks that form the soils of the dietary catchment. Since ⁸⁷Sr is radiogenic and is produced from the radioactive decay of ⁸⁷Rb (t_{y_2} = 48.8 By), regions of older geology (or geologies with high ⁸⁷Rb) will have higher ⁸⁷Sr/⁸⁶Sr than areas of younger geology. Hence, tooth enamel that formed from dietary catchments that have different parent geologies will have different ⁸⁷Sr/⁸⁶Sr, enabling the identification of different childhood origins for individuals and fauna.¹¹

The most common application of Sr isotope analysis on archaeological material has been to identify individuals who are 'non-local', that is they spent their childhood (when the enamel of their teeth was mineralising) in a different strontium isotope catchment to the location of their burial. The 'local' Sr isotopic range can be estimated from Sr isotopic values of non-migratory fauna, sediments, flora and drinking water from the vicinity of the site where the tooth samples were recovered.¹² Where Sr isotopic values in an individual's teeth lie outside this range, they are deemed to be non-local, and must have migrated to the site sometime between the end of enamel mineralisation (e.g. c.14 years old for M3) and their death. Among many such studies, this technique has been useful in understanding degrees of human mobility in the past,¹³ patterns of exogamy and kinship,¹⁴ and animal migration.¹⁵

However, recent developments in laser ablation sampling allow the measurement of Sr isotopes along the growth axis of tooth enamel at very high spatial resolution.¹⁶ Since enamel forms incrementally, measurements along the enamel provide a time series of Sr isotopes and enable the reconstruction of movement over the time the enamel is mineralising. Intra-tooth sampling of Sr has previously been done by drilling sequential samples from the enamel,¹⁷ though it has rarely produced more than 5 or 6 measurements per tooth. Laser ablation Sr isotope analysis, on the other hand, can produce hundreds of isotopic measurements per millimetre, allowing the detection of movement over far shorter timescales than drilling. It is therefore ideal for detecting movement over shorter (e.g. seasonal) timescales as required to identify certain animal husbandry regimes such as transhumance.

¹⁴ E.g. Haak et al. 2008.

⁹ See Kanz et al., this volume.

¹⁰ Pottery sherds and human bones exhibit refitting joins throughout the ash layers. For further information on the character of the ashy layers and their depositional history, see the contribution by Jung and Pacciarelli in the present volume.

¹¹ E.g. Bentley 2006.

¹² E.g. Bentley et al. 2004.

¹³ E.g. Price et al. 2004.

¹⁵ Pellegrini et al. 2008.

¹⁶ De Jong et al. 2010; De Jong 2013; Lewis et al. 2014.

¹⁷ E.g. Bentley – Knipper 2005.



Fig. 1 Location of the water sample in the region of Punta di Zambrone (map: A. Buhlke)

Samples and Sampling

In order to define the 'local' range to the site, enamel from non-migratory archaeological fauna (a dog and a badger¹⁸) were sampled, along with human and faunal dentine – which in most cases takes up Sr from the burial environment and therefore reflects the 87Sr/86Sr of the immediate site. Water samples were collected, both local to the site, and further afield to provide a Sr isotope 'map' of the region (Fig. 1). These also included a small number of water samples from Achaea, Greece (Fig. 2), since Mycenaean pottery found at Punta di Zambrone turned out to be produced in that region and in further regions mainly in western Greece.¹⁹ The human teeth sampled (SP1 – M1, M2 and M3) were mandibular permanent molars and provide a time series of Sr isotopes from approximately birth to about the age of 14 for this individual.²⁰ A selection of domesticated fauna (pigs, sheep, cattle, horse) were sampled to look at animal procurement strategies (Tab. 1). We especially wanted to test whether the hinterland of the site, rising from the coastal strip and extending over hilly terraces up to the Poro high plain or plateau (with elevations of more than 600m asl), formed an economically integrated system during the different habitation periods of Punta di Zambrone.²¹ This would help to understand the role the site may have played as a harbour for an economically and perhaps politically structured system of settlements.

¹⁸ Larivière – Jennings 2009, 624.

¹⁹ Jung et al. 2015; Jung et al., this volume.

²⁰ AlQahtani et al. 2010.

²¹ As Pacciarelli (2000, 78–84 with figs. 42–44) has proposed for EBA 2, MBA 1–2 and the RBA.



Fig. 2 Water sample locations in the central plain of Achaea (map: A. Buhlke)

Lab ID	Species/age/tooth	Site code(s)	Date	Notes
SP1 (M1, M2, M3)	Human mandible M1, M2 and M3	Area C, PZ95/P9 (= DNA sample D041625)	RBA 2	Early adult 20–30 years. M2 section incomplete
SP2	Pig, left maxilla, M3?	Area C, PZ95/P8	RBA 2	
SP3	Horse, right maxilla, M1	Area C, PZ66FFGG10	RBA 2	
SP4	Red Deer, right mandible, I2	Area C, 66cCC12	RBA 2	
SP5	Pig, right maxilla, M3	Area B, PZ83/P3	EBA 2	
SP6	Probably domestic pig	Area C, PZ66cBB11	RBA 2	
SP7	Badger, c. 1 year, left maxilla, M1	Area C, PZ66DD11	RBA 2	
SP8	Sheep, right mandible, P4	Area C, PZ66EE11	RBA 2	
SP9	Dog, right mandible, M1	Area C, PZ95DD8	RBA 2	
SP12	Bovine, > 2 years, right maxilla, P3	Area B, PZ86cF3	EBA 2	
SP13	Pig, > 1.5 years, 4 teeth, possi- bly from the same individual	Area B, PZ191I2	EBA 2	
SP14	Pig, > 1.5 years, left mandible, P2	Area C, PZ129BBCC12	RBA 2	
SP15	Sheep/goat, > 4 years, right maxilla, M2	Area C, PZ129bDD11	RBA 2	certainly from an ani- mal other than SP16
SP16	Sheep/goat, right maxilla, M1 or right mandible, M2, both >1 year	Area C, PZ129bDD10	RBA 2	certainly from an ani- mal other than SP15
SP17	Bovine, > 0.5 years, right maxilla, M1	Area C, PZ129bCC10-11	RBA 2	

Tab. 1 List of tooth samples analysed

Longitudinal sections of approximately 1.5mm thick of enamel and some incidental dentine were removed using a hand drill and diamond cutting disk. As far as possible, the samples represent a section of the complete length of the enamel from the crown to the cervix. Samples were cleaned in an ultrasonic bath in $18M\Omega$ H₂O for ten minutes and dried overnight in an oven at 60°C.

Analysis

The samples were mounted in the laser cell by pressing them into pressure-sensitive adhesive putty (Blu-tack). Sr isotopic analysis was performed on a Thermo Fisher Neptune multi collector ICP-MS with a New Wave 193nm ArF homogenised excimer laser, using the oxide reduction technique of De Jong.²² The measurement of Sr isotopes by laser ablation has only recently been made reliable. The primary difficulty has been the molecular interference on ⁸⁷Sr of ⁴⁰Ca³¹P¹⁶O⁺ which is the primary constituent of the enamel matrix. Other potential problems come from doubly charged rare earth elements which give mass to charge ratios of between 84 and 88, calcium-calcium and calcium-argide dimers which can interfere with ⁸⁴Sr, ⁸⁶Sr and ⁸⁸Sr, in addition to potential ⁸⁷Rb and ⁸⁶Kr interferences. Our approach has been to minimise oxide formation (monitored as ²⁵⁴(UO)^{+/238}U⁺) through careful control of plasma conditions, and to monitor and reject teeth that have significant rare earth concentrations which we consider to indicate diagenetic alteration. We correct for the ⁸⁶Kr using an on peak gas blank and for rubidium interference using the natural ⁸⁷Rb/⁸⁵Rb ratio of 0.385617. A small positive offset from known values ⁸⁷Sr/⁸⁶Sr of calcium phosphate standards is usually observed because the oxide interference is not completely eliminated, but it is normally well within the precision of a typical measurement.

Time series of strontium isotopes are obtained as continuous data by moving the tooth along its growth axis (at 15 or 25μ ms⁻¹ depending on the size of the tooth) as the laser pulses with a repetition rate of 10Hz and spot size of 150 μ m, giving a fluence of c. 8.6Jcm⁻², and collecting strontium isotopic data every 2 seconds. Thus a single measurement represents the Sr isotopic signal integrated over 30 or 50 μ m of tooth enamel, although in practice we present the data as a 5-point moving average. The human (SP1) dentine associated with M1, M2 and M3, the pig dentine (SP2) and the enamel from the badger (SP7) and the dog (SP9) were analysed as 'spots'. The laser, with a spot size of 150 μ m, was pulsed at 10Hz for 90s without moving the sample, effectively 'drilling' with the laser and sampling a few tens of microns of the tooth orthogonal to the axis of growth. Where appropriate, spots were repeated at different locations on each of the tissues.

Repeat analysis of an in-house ashed bovine pellet standard (BP1) bracketing every third analysis, showed an offset of $+89\pm98$ ppm (1 sigma) for the laser ablation analyses over TIMS values. This is about the same order as the within-tooth variation for homogenous teeth, but well within the total variation between the teeth of c. 1000 ppm, and is therefore considered insignificant to our interpretation of the results.

Water samples were analysed by conventional Thermal Ionisation Mass Spectrometry (TIMS) methods following strontium separation on Sr-Spec columns and loading onto tantalum filaments with a tantalum activator solution. The samples were analysed at a ⁸⁸Sr beam size of 2V on a Thermo Fisher Triton Plus. The long-term average for NIST987 for the instrument is 0.710244 \pm 0.000019 (2 s.d.) on 180 analyses.

Results and Discussion

Table 2 shows the results of the dentine and analyses used to define the local range. Table 3 gives the locations and results of the analyses of the water samples. Water samples from within a radius of about 6km from the site gave Sr isotopic values indistinguishable from the local range defined

²² De Jong et al. 2010; De Jong 2013; Lewis et al. 2015.

by the dentine and enamel samples. The exception to this is the water taken from a fountain close to the site itself (WP7) which gave the most radiogenic Sr, completely at odds with the dentine measurements. The dentine is very consistent between samples, so it is likely that the water sample is unrepresentative of the local Sr isotopic range, either because of modern contamination of the water source, or because of an error in sampling or analyses.²³ We therefore exclude this sample in our interpretation of the results.

Lab ID	Tissue	⁸⁷ Sr/ ⁸⁶ Sr
SP1	M1 – dentine	0.708770 ± 42
	M1 – dentine	0.708764 ± 26
	M2 – dentine	0.708679 ± 25
	M3 – dentine	0.708715 ± 51
	M3 – dentine	0.708718 ± 40
SP2	Dentine	0.708909 ± 21
SP7	Enamel 1	0.708934 ± 19
	Enamel 2	0.708766 ± 14
	Enamel 3	0.708797 ± 17
SP9	Enamel 1	0.708566 ± 31
	Enamel 2	0.708568 ± 31
	Enamel 3	0.708560 ± 30

Tab. 2 Results of laser 'spot' analyses of Sr isotopes of dentine and enamel. Errors at 2 SE.

Lab ID	Location	⁸⁷ Sr/ ⁸⁶ Sr
WP1	Aqua Pirara	0.708374 ± 0.000014
WP2	Bandino	0.709007 ± 0.000013
WP3	San Michele	0.709264 ± 0.000012
WP4	Sorgente Ambrosia	0.708733 ± 0.000015
WP5	Daffinacello	0.708552 ± 0.000014
WP6	San Giovanni "La Velli"	0.708830 ± 0.000014
WP7	Marina de Zambrone	0.709565 ± 0.000012
WP8	San Cono	0.708904 ± 0.000011
AC1	Spring close to the Mycenaean cemetery of Voúdeni	0.707959 ± 0.000016
AC2	Spring close to the Mycenaean settlement of Chalandrítsa	0.708049 ± 0.000016
AC3	River Píros close to the village Chaikáli	0.708311 ± 0.000016
AC4	Yianiskári, plant sample (not far from the beach)	failed
AC5	Teichos Dymaion, plant sample (close to the Mycenaean citadel)	failed

Tab. 3Results of the water analyses from around Punta di Zambrone (WP) and from the central plain of Achaea (AC).Errors at 2 SE.

²³ We suppose that the first hypothesis is correct, as this specific fountain has been restructured in recent years by adding modern pipes and installations. All the other water samples come from old fountains used by the local population for decades and retain their traditional layout. We thank Francesco Rombolà for taking us to the different fountains in the region.



Fig. 3 Results of the Sr isotope analysis of SP1. The black lines represent the 5-point moving mean of individual isotopic measurements, with error bars representing the standard error. The broken lines represent the 'local' Sr isotopic range as defined by the dentine, badger and dog samples. Also shown are the water samples from the region (blue) and from Achaea (red) (graphics: A. W. G. Pike)

Is SP1 a Local?

The laser ablation Sr isotopes of the three molars of individual SP1 are shown in Fig. 3. The molars represent a time series of isotopes from approximately birth to around 14 years of age. There is little variation in Sr isotopic ratios within the three teeth indicating that this individual moved very little during the time of enamel formation (i.e. the first 14 years of life), or if he/she moved he/she did so to an area of similar strontium geology. The human dentine, used as a proxy for the Sr isotopes in the groundwater immediate to the site, cluster tightly around the mean values for the three tooth enamel samples. The strontium isotopic composition in human teeth, however, reflects the strontium of the whole dietary catchment with a smaller input from drinking water. A better estimate of the 'local' strontium isotopic range therefore comes from non-migratory mammals found at the site. Their foraging and scavenging behaviour is likely to sample an area or diet more representative of human subsistence behaviour, and here we use badger and dog enamel to define the local range of strontium isotopes (broken lines on Fig. 3) which gives a broader range than the human dentine. This range neatly brackets all the 5-point running mean isotopic values for the three human teeth. So notwithstanding movement to the site from an identical strontium geology later in life, our most parsimonious explanation is that the individual SP1 is local to the area of his burial. It would not have been surprising, however, if the individual had come from outside the region since Punta di Zambrone is a port with clear connections to elsewhere in the Mediterranean, for example, the pottery link to Achaea, Greece. However, the water samples (AC1–3, Fig. 3) indicate that he did not grow up in this area. The remains were also highly fragmented (a portion of a disarticulated mandible) and lacked a formal grave which may suggest an outsider and/or someone connected to the destruction of the site. However, given the Sr isotope result, and without further evidence, we must conclude at present that the individual was local and the remains represent either a grave disturbed during the Recent Bronze Age (with its contents partially re-deposited in the fortification ditch), or a burial rite that resulted in highly dispersed remains.



Fig. 4 Results of the Sr isotope analysis of pigs (graphics: A. W. G. Pike)

Animal Herding and Procurement

The fauna show a different pattern with some Sr values falling outside the local range, and matching to water samples taken further afield. Three of the five pigs match the local range and were probably raised within a few km of the site (Fig. 4). The other two match water samples from the Poro high plain rising to the south and southeast of Punta di Zambrone. None of them shows significant variation in Sr isotopes within the enamel, indicating that they were not moved to a different geology during enamel formation. This is not surprising as pigs can consume a wide variety of diets including human food-waste which means the provision of winter fodder or seasonal movement to fresh resources is less of an issue than for cattle.

The sheep and one bovine (SP17) show a similar pattern to the pigs (Fig. 5) with some of them consistent with a local origin, and others from the high plain. However, the horse (SP3) and the other bovine (SP12) show variation in Sr isotopes along the growth axis of the enamel indicating movement during the period when the enamel was mineralising. Both have at least one period where the isotopes are consistent with the local range for the site, though both spent the majority of the time away from the site. For the horse tooth, during its stage of mineralisation, the isotopes suggest movement between the high plain (water sample 1, Fig. 1), the local catchment (water samples WP 4, 5, 6, 8), and a third unknown location with low 87 Sr.

The bovine (SP12) moves from the high plain (represented by water sample 3) to the lowland near the site, returns to the high plain, moves again to the lowland and returns again to the high plain, all within the period of mineralisation of its maxillary P3. These results ought to be interpreted in the light of comparative morphology. While carnivores and pigs are characterised by brachydont (short-crowned) teeth with rather short stages of mineralisation (less than a year), herbivores like cattle and horse, at least in parts of their dentition, show proto-hypsodont (high-crowned) teeth. In horses the duration of the mineralisation process varies between 1.5 and 3 years; for the mandibular M1 an average time of 2 years is reported, starting at the age of one month.²⁴ For cattle, unfortunately, no reliable data on the duration of enamel formation in

²⁴ Hoppe et al. 2004.



Fig. 5 Results of the Sr isotope analysis of various fauna (graphics: A. W. G. Pike)

P3 is available,²⁵ but the mineralisation of a maxillary P3 should not be estimated at less than 2 years.

Cattle movement can be explained in several ways. In areas of seasonal climate, transhumance is a strategy of moving cattle (and sheep) to fresh pastures, usually on a seasonal basis. This can be vertically over short distances (e.g. to Alpine pastures), or horizontal transhumance, usually over longer distances.²⁶ In the case of SP12 the enamel formation process presumably started at an age of 1.5 years, and lasted after its eruption at the age of 2–2.5 years for another 0.5–1 year. The discernible changes of ⁸⁷Sr/⁸⁶Sr most likely reflect seasonal movements in the context of transhumant herding.

The horse SP3 apparently spent the first 6–8 months of its life at the high plain, then moved to the lowland for a similar span of time and, after a short excursion somewhere else, returned to the high plain at least until the end of its 2nd year of life, which again might be related to the need to find pasture away from the main settlement.

Figure 6 shows a summary of the origins of the fauna based on the best match to the water samples. The results show that while about half of the animals have Sr isotopic ratios consistent with an origin near to Punta di Zambrone, the inland plain is also an important source of domestic fauna found at the site in both the Early and Recent Bronze Age. Without further comparative sites it is not possible to determine if this pattern was the norm for this period, or whether the specialist nature of Punta di Zambrone as a port reduced its capacity to raise animals locally so it relied on farming communities in the hinterland to provide food, either through trade, or through remote land ownership. Interestingly, however, given that Punta di Zambrone is a port, we find no evidence of animals imported from further afield. While we can never rule out distant origins that have similar Sr values, all the animals have values consistent with an origin within about 15km of the site, and the port settlement appears self-sufficient, at a regional level, in domestic animals at least. This is an important result hinting at the economic capacity of a settlement system that

²⁵ Brown et al. 1960, erroneously also cited by Zazzo et al. 2005, report exclusively on the mineralisation of bovine incisors.

²⁶ Davies 1941; Bartosiewicz – Greenfield 1999.



Fig. 6 Sr catchment of the fauna. The Sr isotopes of water samples 4, 5, 6 and 8 (circled) are indistinguishable from the local Sr isotopic range of the site and are considered a single Sr catchment. Water sample 7 has been excluded from the interpretation because it contradicts results from the dentine and non-migratory fauna (see text) (graphics: A. Buhlke and A. W. G. Pike)

extended from the coast up to the Poro high plain, both in EBA 2 and in the later RBA 2. It does indeed seem that settlements had complementary roles depending on their topographical position in the three different geographical zones of this system.²⁷

Conclusions

This is the first high-resolution intra-tooth strontium isotope study on Italian archaeological material, and the first in the country to look at domestic animal herding and procurement strategies. Analyses of faunal Sr isotopes reveal a pattern of procurement of animals that includes local (<6km) sources as well as revealing the importance of the inland high plain (6–15km) as a region where animals were reared. Intra-tooth variation in Sr in a horse and bovid suggest movement of animals, possibly as part of a transhumance regime.

A comparison between the strontium isotopes of local non-migratory fauna (a badger and a dog) and dentine samples with the three molars from SP1 suggest the individual spent his/her childhood on similar strontium geology to that local to his burial. The simplest explanation is that he/she grew up close to the location of his/her burial, though we cannot rule out an origin with similar geology elsewhere.

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²⁷ See above with n. 21.

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