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#### A note from the Editor

I am Chris Schmidt and I am pleased to be the new Editor of Dental Anthropology. This issue of DA marks a new chapter for a journal that has served as a cornerstone for the dissemination of dental anthropological news and research since 1986. Long time Editor Ed Harris is now the Editor Emeritus, a status well-earned for his years of dedication to dental anthropology and to DA. It is my hope to continue the tradition of excellence that Ed established and to maintain the journal at the highest level of academic value and integrity possible. To do that, of course, requires a commitment from the entire Dental Anthropology Association to submit outstanding manuscripts intended to inform us all on the current advancements of our field. As a young scholar, I read many articles from the giants of dental anthropology, many of whom are household names for those of us so enamored with teeth. Today, many of those giants are still producing great works, but the burden is now squarely on the younger generations to keep the excellence going. Therefore, I encourage scholars at all stages of their careers to consider publishing in DA. No other source so directly supports the research of dental anthropologists and, as we all know, dental anthropology's value to various subfields of anthropology only continues to grow. Morphology serves as the core of dental anthropology, but a number of additional avenues of dental study, ranging from pathology to wear, are important to our understanding of the dentition and research in these areas should find a home in DA. Finally, know that the giants of dental anthropology are still out there and that nothing would make them happier than to see the journal they started nearly three decades ago making new giants to join them in their Pantheon. And remember, "Teeth Rule". If you don't believe me, check out my license plate!



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### Brief Communication: Premolar Enamel Formation: Completion of Figures for Aging LEH Defects in Permanent Dentition

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#### Key Words: Cross striations; Striae of Retzius; Enamel formation; Tooth growth; Dental development

*ABSTRACT* Variation in enamel formation has become increasingly important in comparative studies of dental development. Previously published work on the development of human enamel in groups from southern Africa and northern Europe has allowed for more accurate estimation of formation timing of linear enamel hypoplasias. Currently, although data for all

Variation in enamel formation provides many avenues of inquiry for those interested in comparisons of developmental life histories and aging enamel defects. For example, assessing the age of formation of linear enamel hypoplasias provides one line of evidence to estimate rates of childhood morbidity (e.g., Goodman and Song, 1998). Due to the nonlinear nature of enamel growth, linear regression formulas utilizing total crown height cannot be used to accurately estimate age of LEH formation (Reid and Dean, 2000; Martin et al., 2008; Ritzman et al., 2008). Data on population specific variation in crown formation times based on percentages of total growth are therefore of great use for accurately estimating LEH formation times.

The Reid and Dean (2006) methodology for histological growth assessment reflects the variability of enamel growth rates along the length of the crown and incorporates standard deviations reflecting inter-individual variation, avoiding the problem of applying linear statistical models to a nonlinear growth pattern. Studies completed using other methods, including dental radiographs of living children, suggested high levels of variation in crown formation time among human populations (e.g., Tompkins, 1996). The implication is that new research on LEH formation times conducted without the histological growth assessment method should draw on crown formation timing data specific to the population being studied (Reid and Dean, 2000; Reid et al., 2006; Martin et al., 2008). However, recent work in

tooth types has been published, charts of enamel growth by decile useful in this type of estimation have been limited to molars and anterior teeth. This paper completes this series with a table and figure of mean formation times of human premolars for each decile of crown development using previously published histological data of daily enamel growth.

which incremental growth is assessed histologically has shown the anterior teeth and molars to be less variable between populations than previously reported (Reid and Dean, 2006).

Reid and Dean first published research comparing enamel formation times in populations from northern Europe (2000, 2006) and southern Africa (2006) that included tables presenting age in days for enamel formation at the completion of each decile of crown height for anterior teeth and molars (2006: 334-35). Figures depicting these data provided a visual guide for estimating LEH (2006: 343-44). These figures include an estimated age at mineralization based on previous histological studies (Reid et al., 1998; Reid and Dean, 2000; Antoine, 2001; Dean and Reid, 2001) but the authors note these initiation times are highly variable, as much as a full year in the M3 (Reid and Dean, 2006). A follow-up study (Reid et al., 2008) presented premolar data from these same populations, but did not provide charts of growth by decile of crown height or figures including initiation estimations.

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	LP3 n=33	LP4 n=28	UP3 n=45	UP4 n=41
Initiation	675	967	675	967
Cusp completion	902 +/- 27	1234 +/- 32	910 +/- 36	1211 +/- 42
10% complete	943 +/- 29	1270 +/- 34	949 +/- 36	1250 +/- 42
20% complete	995 +/- 33	1310 +/- 38	993 +/- 34	1292 +/- 42
30% complete	1053 +/- 37	1356 +/- 48	1039 +/- 34	1335 +/- 42
40% complete	1116 +/- 48	1408 +/- 53	1092 +/- 37	1382 +/- 46
50% complete	1187 +/- 61	1472 +/- 60	1157 +/- 44	1444 +/- 46
60%complete	1266 +/- 75	1548 +/- 74	1239 +/- 52	1526 +/- 50
70% complete	1356 +/- 92	1638 +/- 92	1336 +/- 55	1627 +/- 58
80% complete	1452 +/- 109	1741 +/- 110	1443 +/- 62	1737 +/- 70
90% complete	1558 +/- 124	1869 +/- 117	1570 +/- 71	1854 +/- 77
Crown completion	1665 +/- 141	1986 +/- 124	1703 +/- 76	1974 +/- 82
	Northern Europear	n Crown Formatio	n Times	
	LP3 n=33	LP4 n=22	UP3 n=34	UP4 n=44
Initiation	675	967	675	967
Cusp completion	891 +/-44	1231 +/-36	952 +/-65	1225 +/-63
10% complete	952 +/-54	1276 +/-41	1010 +/-61	1284 +/-52
20% complete	1018 +/-68	1328 +/-43	1068 +/-52	1338 +/-48
30% complete	1088 +/-80	1381 +/-52	1132 +/-50	1390 +/-44
40% complete	1164 +/-96	1441 +/-65	1203 +/-49	1448 +/-48
50% complete	1256 +/-113	1509 +/-76	1285 +/-53	1521 +/-52
60%complete	1359 +/-135	1594 +/-84	1389 +/-60	1613 +/-62
70% complete	1481 +/-164	1697 +/-98	1518 +/-66	1724 +/-71
80% complete	1614 +/-194	1817 +/-122	1663 +/-78	1856 +/-85
90% complete	1766 +/-224	1948 +/-143	1838 +/-84	1998 +/-98

 TABLE 1. Age (in days) for enamel formation at each decile of crown height for molar teeth in each sample, +/-1

 standard deviation

 Southern African premolar tooth crown formation times

Currently, therefore, although enamel growth patterns of these populations have been established, the series of charts and figures providing a visual guide for estimating population specific LEH formation times has lacked the published information on premolars. This communication completes the publication of this series of figures by presenting premolar enamel growth by decile for populations from southern Africa and northern Europe (Table 1, Fig. 1).

#### MATERIALS AND METHODS

Data from 147 premolars collected from two populations, southern Africa and Newcastle, England, (northern Europe), were used to create tables of enamel formation (Reid et al., 2008). The premolars were originally collected after extraction during oral surgery and histological thin sections were prepared for polarized light microscopy (Reid and Dean, 2006). Individual periodicity for each tooth was established by counting daily cross-striations in enamel, and formation times for each decile of crown height was then recorded using measurements of long-period striations corresponding to the perikymata on the external crown surface. Initiation ages of crown mineralization for both samples were estimated from a third, French sample (Reid et al., 1998). By adding these decile data to age at initiation of crown mineralization, formation times for both cuspal and lateral enamel formation in days was determined (see Reid et al., 2008 and Reid and Dean, 1998 for full discussion of methodology).



**Fig. 1.** Mean estimates for the chronological ages of enamel formation in premolars for each decile of crown length rounded up or down to 0.1 year for the southern African sample vs. the northern European sample. Both initiation and cuspal enamel formation are included in these estimates.

Although extremely preliminary research by one of the authors on mineralization initiation times specific to these populations supports the current expectation that mineralization timing will vary, conclusive data from large scale studies remains unavailable. Given the absence of data, the significance of future publications in this area on the chart presented here would be speculative, however, the authors intend to update the available charts as new data are available.

#### DISCUSSION

Although total crown formation time was found to be significantly different between the populations (p<0.00) (see Reid et al., 2008 for full discussion of statistical methods), as with the anterior and molar teeth, premolar formation time between populations is more similar than radiographic studies once suggested (e.g., Tompkins, 1996). While the southern African sample crowns formed consistently more quickly, the means of both samples range only from 0.3 years difference in the lower P4 to 0.8 years difference in the upper P3. Thus, the small amount of variation may not be particularly meaningful in comparisons of human populations (Reid and Dean, 2006; Martin

et al., 2008).

Because this method presents formation time by decile, the estimation of age during LEH formation can be independent from any variation in length of the crown. This more accurate method has already been widely used for the anterior teeth and molars (e.g., Reid and Dean, 2000; 2006). The addition of premolar data will allow for comparisons of LEH within the dentition of individuals, specifically to match LEH manifestations of a single stress event across premolars and other teeth within an individual. Despite the fact that enamel growth is not linear, Martin et al. (2008) found no difference between a linear and nonlinear interpolation for ages that fall between the established deciles. They conclude that a linear interpolation is sufficient to age defects that fall within a decile. While we await further histological data from other populations, the charts presented here (Table 1, Fig. 1) make it possible to provide estimates of LEH formation times that utilize the population specific information currently available from two geographically distant populations.

#### CONCLUSION

Recent studies applying histological methods to establish enamel formation timing have shown less variation in modern human premolar formation between populations than previous methodologies; however, statistically significant differences have been documented between a large sample of northern European and southern African individuals (Reid and Dean, 1998; Reid et al., 2008). Previously published data on histological timing of each decile of premolar tooth crowns can be used to estimate timing of LEH formation without destructive histological sectioning. This paper presents a summary of the previously published data on crown formation variation and presents a graphic diagram of the premolar formation times by decile drawn from histological analysis (Table 1, Fig. 1). In addition to providing previously unpublished data on the mineralization initiation estimates used to create these decile formation charts, it is the hope of the authors that including a visual representation of premolar crown formation by decile to the existing charts for other tooth types will allow for more practical application of the known formation timing to analysis of the external enamel of premolars.

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#### Hypotrophic Roots of the Upper Central Incisors – a Proposed New Discrete Dental Trait

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**Key words:** Dental morphology, Upper Central Incisors, Discrete Dental Trait, Prehistory, Iberian Peninsula.

*ABSTRACT* This paper describes a newly defined nonmetric trait in the human dentition, i.e., Hypotrophic Roots of the Upper Central Incisors (HRUCI). Teeth presenting HRUCI are characterized by abnormally short roots whose crowns

In the mid 20th century, A. A. Dahlberg devised a set of standardized plaster casts to be used as scoring aids in recording the expression of discrete dental traits (Turner et al., 1991). Subsequently, publications in the 1970s through 2000s by Turner, Scott and others at Arizona State University helped to expand, systematize, and broadly disseminate the methodology commonly used today to score traits: the Arizona State University Dental Anthropology System (ASUDAS) (Turner et al., 1991; Scott and Turner, 1997). From a genetic standpoint these features are "discontinuous or quasi-continuous traits that are either present or absent or present in various degrees of expression" (Larsen, 2002:137-138). In living populations, first degree relatives have frequencies of some traits six times higher than in the general population (Mays, 1998). Statistical tests applied to the observation of discrete dental traits have helped answer questions on genetic proximity/distance within groups (Gorsky et al., 1998) and between groups (Matsumura, 2007) and questions of ancestry among contemporary groups (Larsen, 2002) as well as archaeological populations (Irish, 2006; Jackes et al., 1997; Jackes and Lubell, 1999). Such traits have been used as a basis for inferences on the peopling of geographic regions (Larsen, 2002; Silva, 2002) or for the understanding of population dynamics in times of crisis, or population, cultural and/or technologi

exhibit no apparent morphological alterations. The trait was observed in six samples from collective funerary sites in the Iberian Peninsula dated from the Late Neolithic to the Chalcolithic period.

cal shifts (Silva, 2002; Vargiu et al., 2009).

Despite the considerable research and number of publications produced, the ASUDAS is continually 'under construction' (Scott, 2008). New contributions and changes are ongoing. New casts have been created and scoring grades for previously non-scored traits were recently published (Wu and Turner, 1993; Burnett et al., 2010). Moreover, new traits have been described and further discoveries are expected in under-studied regions and/or populations (Weets, 2009).

The study of dental remains from several human skeletal collections dated from the Late Neolithic to the Chalcolithic period allowed the observation of an unusual root feature. European collections are characterized by simple morphology (Scott and Turner, 1988). However, individuals from the collections included in the present study exhibit various atypical characters – including UI1 presenting unusually short roots. The aim of this paper is to describe this 'new' discrete trait and discuss its frequency in six Late Prehis-

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**Fig. 1.** (1) Cadaval Cave; (2) Tholos of Paimogo I; (3) Hypogeum of São Paulo II; (4) Monument of Cerro de las Baterías; (5) Perdigões ; (6) Pit burial.

toric population samples from different regions of the Iberian Peninsula (Fig. 1).

#### CLINICAL APPROACH TO SHORT ROOTS

Most of the dental literature deals with the occurrence of short roots in the human dentition from a clinical perspective in Modern populations. Specifically, the focus is related to orthodontic treatment. Hölta et al. (2004) suggest there are two general reasons for short roots: disturbances during root development, and resorption of originally well developed roots. A genetic etiology of short roots, particularly for UI1, is proposed by some (Lind, 1972; Jakobsson and Lind, 1973; Apajalahti et al., 1999; Apajalahti, 2004; Edgcomb, et al., 2011). However, the phenomenon is usually dealt with as an 'anomaly' or morphological alteration, i.e., resorption of the roots. Resorption is thought to be triggered by one or more external factors, including: (1) trauma from orthodontic treatments, surgical procedures, and occlusal pressure; (2) anticancer therapy with chemotherapeutic drugs and radiation, and/or (3) industrialized toxins such as dioxins (Ando et.al, 1967; Hylander, 1977; Brezniak and Wasserstein, 2002a, b; Apajalahti, 2004).

Population (Ando et al., 1967; Jakobsson and Lind, 1973; Thongudomporn and Freer, 1998), family (Apajalahti et al., 1999) and longitudinal studies (Ando et.al, 1967; Hölta et al., 2004) indicate that short roots in UI1 is a genetically controlled feature (Brezniak and Wasserstein (2002a) state that root resorption is "an unavoidable consequence of orthodontic tooth movement."). It is more frequent in females than males at a ratio of 2.7:1 (Jakobsson and Lind, 1973; Thongudomporn and Freer, 1998); moreover, there are differences in frequencies among populations (Table 1), and it may have an autosomal dominant pattern of transmission (Apajalahti et al., 1999). In cases where resorption is not considered the cause, shortened roots are bilaterally expressed (Jakobsson and Lind, 1973; Margues et al., 2010). However, as far as we know, this trait has not



**Fig. 2.** UI1 presenting average sized root (left) and another displaying HRUCI (right), both from Cerro de las Baterías, Spain.

Ethnic Group/Origin	n	Prevalence	Reference
Mongoloid (Japanese)	300	10%	Ando et.al, 1967.
Caucasian (Swedish)	1038	2.4%	Jakobsson and Lind, 1973.
Caucasian (?) (Australia)	111	23.4%	Thongudomporn and Freer, 1998.

TABLE 1. Prevalence of Bilateral Short Roots in UII

been employed as a normal epigenetic variant of the human dentition.

#### MATERIALS AND METHODS

Trait Description: Lind (1972) developed a metric method to record the phenomenon by determining the ratio between root length and crown height (R/C value) for the UI1; the method was later applied to a sample of 1,038 white children of both sexes (Jakobsson and Lind, 1973). The latter study reported a mean R/C value of UI1 with fully developed roots of 1.63 for males



**Fig. 3.** Distances measured in order to obtain the length of roots and crowns of incisors included in the study.

and 1.55 for females. Dimorphic differences were not significant, and when the sexes were pooled the general mean R/C value was 1.6.

The trait that we observed can be described as the occurrence of substantially shorter roots for UI1 that are either equal to, or shorter than the incisor crown height. For this trait, the crowns themselves are normal in appearance, and no other teeth exhibit root diminution. We propose that this trait, defined by a root:crown ratio of <1.5:1, be named Hypotrophic Roots of the Upper Central Incisors (HRUCI) (Fig. 2). Further studies on samples both within and outside Portugal are planned to assess the trait's presence in other local and world regions; it also will be determined if simple presence/absence scoring, like that in ASUDAS, is warranted for this trait.

Both crowns and roots of UI1 in all samples were measured using a Mitutoyo Digimatic caliper with an accuracy of 0.01 mm. Measures were taken in labial view. Root length was measured between its apex and the cement-enamel junction in the sagittal plane of the tooth (D1 in Fig. 3). The crown length was measured between the cement-enamel junction, again in the mid-sagittal plane, to the most occlusal point of the incisal edge (D2 in Fig. 3). Teeth exhibiting roots of equal length or shorter than the crown were considered to present HRUCI.

Teeth with incisal wear of grade 3 and above [scale proposed by Smith (1984), as modified by Silva (1996)] were excluded from the study for two reasons: first, to avoid shortening of roots resulting from occlusal trauma caused by pressure loading; and second, because severe tooth wear would interfere in the crown-root ratio. For the same reason fragmented incisors and others in early stages of development were not considered. Only teeth with an apex almost or completely closed were measured, i.e. individuals older than 8 yrs of age (Smith, 1991).

Presence/absence dichotomy of the trait was registered to obtain the frequency of HRUCI in the collections.

#### Samples

Cadaval (CDV) is one of the burial caves in the limestone region of Canteirões in the Nabão river valley – a sub-tributary of the Tagus river, Portugal. Radiocarbon dates from the human skeletal material indicate that CDV was in use between the third quarter of the 5<sup>th</sup> millennium and middle of the 4<sup>th</sup> millennium BC (Tomé, 2011).

Cerro de las Baterías is a funerary monument located in the Province of Badajoz, Spain (Fig. 1) near the town of La Albuera in the Guadiana River Basin. Although <sup>14</sup>C dating was impossible due to taphonomic factors, the funerary assemblage accompanying the human remains presents clear parallels with local archaeological contexts dating to the 3<sup>rd</sup> millennium BC for this region of the Iberian Peninsula (Márquez Gallardo, 2008).

The vaulted chamber tomb (*tholos*) of Paimogo 1 (PMI) and the hypogeum of São Paulo II (SPII)

are located in the same geomorphological region in the Southwest coast of the Iberian Peninsula. Radiocarbon dating places the use of PMI between the end of the 4<sup>th</sup> millennium and the mid 3<sup>rd</sup> millennium BC. Radiocarbon dating on human remains produced dates in the 3<sup>rd</sup> millennium BC for SPII (for detailed osteological data on these samples see Silva, 2002; 2003).

The tholos-like structures of tombs 1 and 2 of Perdigões (PDG) have absolute dates pointing to the 3<sup>rd</sup> millennium BC. The site is located in Reguengos de Monsaraz, Southeast Portugal (Map1) (Valera and Godinho, 2009).

The pit burial of Monte das Covas 3 (MCOV3) is a collective funerary monument for which there are no absolute dates. Typologically this type of inhumation was in use from the Late Neolithic to Bronze Age. The site is located near Beja, Portugal (Miguel and Brazuna, 2008).

In all sites except Monte das Covas 3, skeletons were disarticulated. Although all teeth in this study come from fragmented maxillae, the skulls from Monte das Covas 3 were removed en bloc from the field and excavated in the labora-



**Fig. 4.** A pair of compatible UI1 from BT07 displaying HRUCI (below) and a right presenting UI1 regular root length (above).

tory; this approach allowed the identification of individuals in addition to isolated teeth from undetermined individuals.

Due to the nature of the trait described in this article, identification can only be made in loose teeth or via medical imaging (e.g., radiographs, CT/CAT scans). Cases described here were observed in loose, unassociated teeth, although in the BT07 samples compatibility of antimeres was observed (Fig. 4), and in MCOV3 the loose teeth could be assigned to individual skulls.

The Tagus and Guadiana rivers, along with their tributaries, form major routes for population movement in the southern inland of the Iberian Peninsula; thus, a specific pattern of genetic exchange is likely. Cerro de las Baterías and Perdigões are inland sites near the Guadiana river, while Cadaval is located in the Tagus Basin. The other sites, Paimogo I and São Paulo II, are geographically more prone to coastal influences. The populations from Paimogo I and São Paulo II might have had other genetic influences from groups living on and moving along the Atlantic coast.

#### **RESULTS AND DISCUSSION**

Table 2 presents the total number of UI1 in the samples, the number of specimens considered for the study, and the frequencies of the trait. Several other teeth excluded from the study appeared to exhibit HRUCI, but could not be measured and included in the total numbers due to missing portions of their anatomy.

The frequency of HRUCI differs significantly between samples from the inland sites of CDV, BT07, PDG and MCOV3 and the coastal sites of PMI and SPII. Coastal sites present a frequency under 16% while inland sites have frequencies above 20%. This variation may suggest a different composition of the coastal populations, different patterns of genetic exchange, and/or high endogamy in the inland sites. Further study of other coeval coastal and inland populations is necessary to support any of these hypotheses.

Concerning possible causes that lead to the shortened roots, one common reason, suggested in archaeological, ethnographic (Hylander, 1977) and clinical publications (Ando et al., 1967; Apajalahti, 2004; Silva Filho et al., 2007), is heavy occlusal pressure. This factor is not likely attributable to HRUCI in the present samples. Load/bite

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	Total of coll	UI1 in the ection	Measu	red UI1	UI1 Presen	ting HRUCI
Site	Left	Right	Left	Right	Left	Right
CDV	10	11	3	6	1 (33.3%)	3 (50.0%)
PDG	50	31	25	11	5 (20.0%)	3 (27.3%)
BT07	80	87	37	28	15 (40.5%)	10 (35.8%)
PMI	119	103	108	90	17 (15.7%)	11 (12.2%)
SPII	89	76	66	59	10 (15.2%)	8 (13.6%)
MCOV3	10	13	5	7	2 (40.0%)	3 (42.9%)

TABLE 2. Frequencies of HRUCI in the sites of Cadaval (CDV), Perdigões (PDG), Cerro de las Baterías (BT07),Paimogo I (PMI), São Paulo II (SPII) and Monte das Covas 3 (MCOV3) per side of jaw.

force resulting in resorption or premature closure of the UI1 radicular apex would have been distributed along the other anterior teeth, particularly in lower incisors to cause shortening of their roots as well. In the sample of Cerro de las Baterías, 158 loose lower incisors (LI1 and LI2) presenting closed apex and intact roots were observed to determine if a similar frequency of shortened roots matched that for UI1. Only three teeth (1.89%) presented apparently short roots, and none of the LI1 and LI2 presented roots



**Fig. 5.** Smooth root surface on a HRUCI UI1 (right) and irregular surface of a blunt root in a UI2 (left) presenting evidence of hypercementosis.

equal or shorter than crowns. Hypercementosis, another result of heavy occusal trauma, is absent in UI1 from the samples.

In cases of severe root resorption from repetitive or continuous trauma (e.g., occlusal trauma, tooth movement, etc.), shortening appears to be the natural response of the organism to repair the affected area. In such cases, dentin layers beneath the cementum are affected. The repair process generally begins two weeks after forces that caused the trauma end, and involve active cementum deposition (Brezniak and Wasserstein, 2002a). In archaeological material the most obvious result of this repair process is an irregular layer of cementum altering the physiologic limits of the root – hypercementosis (Fig. 5). Although there are cases of hypercementosis in the present study, none affected UI1.

Other factors listed for shortening of roots in contemporary populations (i.e., clinical treatments, drug use, contamination by chemical agents) do not fit the profile for the Prehistoric samples. Therefore, a genetic etiology for the phenomenon seems to be the most likely explanation. Clinical literature suggests that at least part of the cases studied in Contemporary populations appear to have a genetic pattern of inheritance and family prevalence (Lind, 1972; Jakobsson and Lind, 1973; Apajalahti et al., 1999; Edgcomb et al., 2011). Observation of the sample from MCOV3 and cases of compatible antimeres in BT07 suggest that HRUCI is often bilateral in expression. At present, sample sizes are too small to assess trait independence. Inter-trait associations also will be addressed in future research.

Further research on this trait is necessary to verify its genetic nature. Other collections need to be assessed to confirm the differences of frequencies between inland and coastal areas in the Late Prehistory of the Iberian Peninsula. Comparative studies with other archaeological samples are needed to verify if this trait is of local expression or is present in other regions. These studies will be important tools in the assessment of biodistances for populations recovered from prehistoric contexts in the Iberian Peninsula.

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### A unique case of mandibular osteomyelitis arising from tooth germ infection in a 7,000-year-old infant from Siberia

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**Keywords**: Paleopathology, Tooth Calcification, Bone Infection, Oral Microbiology, Osteomyelitis, Periostitis, Central Asia

*ABSTRACT* Excavations from a 7,000-year-old mortuary site in the Lake Baikal region of Siberia, Russian Federation, revealed an infant with osteomyelitis of the mandible. The lesion exhibits deformation of the anterior mandibular base, an extra-oral cloaca, and new periosteal bone layers on the corpus. Entry of oral microorganisms was likely via the deciduous left canine, with infection

Knowledge of diseases is furthered by their evidence in ancient peoples. Osteomyelitis of the mandible arising from a tooth germ infection is relatively rare in modern populations (Baltersperger and Eyrich, 2009) and is not widely reported in paleopathology literature. This case report presents what is argued to be a 7,000-year-old case of mandibular osteomyelitis with accompanying new periosteal bone in an infant from a mortuary site named Shamanka II in south-central Siberia, Russian Federation. The lesion's hypervascular appearance is rather distinctive, with a small cloaca and extensive, irregular periosteal bone deposits on the mandibular corpus. Osteomyelitis of the jaws differs from that of the long bones because of the former's unique tooth bearing function and connection to the oral cavity and periodontal membrane. Thus, local immunological and microbiological factors are important in the disease's etiology and pathogenesis (Baltensperger and Eyrich, 2009; Slootweg, 2010). Immature teeth are especially susceptible to infection because of incomplete root formation with open apices and large canal sizes (Huang, 2009). Further, because common nonpathogenic oral microorganisms have the potential to cause infection (Willet et al., 1991), osteomyelitis is possible even in the absence of carious or periodontal lesions, as is argued for this case. When considering paleopathology of the jaws it should be kept in mind that oral microbes have great antiquity and thus the potential for an endodontic infection route has been present for many millennia. Therefore, this case report is useful for dental anthropologists and paleopatholothen concentrated around the forming permanent canine tooth germ. This infection route is not widely documented in paleopathology and the pathogenicity of oral microorganisms is discussed. This unique case is one of the oldest examples of infant osteomyelitis of the jaws, adding to our understanding of the antiquity and development of infectious diseases in humankind.

gists for several reasons: a) in presenting an ancient case of mandibular osteomyelitis of unique appearance, b) in demonstrating the presence of the tooth germ infection route, even in the absence of carious and periodontal infection, and c) in illustrating the importance of considering the pathogenicity of otherwise normal oral microorganisms.

#### MATERIALS

The infant, numbered 66-2, was excavated from an Early Neolithic mortuary site located at the southern end of Lake Baikal, Siberia, Russian Federation (Fig. 1). Preservation and completeness are very good, with the only bones missing being the scapulae, ischium, and pubis. Shamanka II has been radiocarbon dated to 7,000-6,100 calibrated years before present (calBP), a period when these hunter-fisher-gatherers began establishing large formal burial areas for their dead (Bazaliiskii, 2010; Weber et al., 2010). Diets were predominately composed of lake fish (Katzenberg et al., 2010). Individual 66-2 was interred beside a young adult female, numbered 66 -1, both in an extended supine position with no evidence of post-depositional disturbance (Fig. 2). Individuals 66-1 and 66-2 are radiocarbon dated to 6931+/-39 and 6890+/-40 calBP respectively, suggesting that they were buried at the same time (Weber et al., 2010). Future ancient DNA analysis may determine the infant's sex and relation to the adult female.

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The following is a description of the mandibular lesion in individual 66-2 (Figs. 3, 4 and 5): The anterior aspect of the mandibular corpus, bracketed by the canines, and extending from the alveolar margin to the inferior margin of the body, has new periosteal bone laid down in roughly horizontal bands, typical of hypervascular lesions (Fig. 3). Both fiber (woven) and lamellar bone are evident. There is some minor, largely resorbed pitting in this area. All this suggests that an episode (or episodes) of healing occurred, but that the lesion was active at the time of death. Slightly left of the mandibular apex, along the inferior border, a small opening is visible, that on radiograph is confirmed to be a cloaca originating at the inferior margin of the forming permanent canine (in the cap stage of development) socket (Fig. 5). The inferior border of the mandible is uneven from the right central incisor to just past the left deciduous canine socket. On the corpus, on either side of the mental eminence, there is increased porosity without much new bone formation. The pores appear to have relatively smooth margins and bone in this area is generally even.

The individual had no other pathological lesions on the elements that were recovered, most notably the maxilla. Note that there is minor post -mortem damage to the anterior alveolar margins of the canine sockets in the form of a small amount of breakage and flaking of the thin bony plates. Otherwise, there are no indications of taphonomic changes to the mandible - no bone distortion, discolouration, fracture, or cortical erosion. The anterior aspect of the mandible is very well-preserved and a considerable amount of morphological detail can be observed. This, combined with the fact that within the lesion new bony deposits, a cloaca, and areas of healing are clearly evident, confirm that the lesion is not the result of taphonomic processes.

#### **METHODS**

The age-at-death of individual 66-2, based on tooth formation and eruption standards, is estimated at 21+/-6 months (1.75 years) (Moorrees et al., 1963a and b; Liversidge and Molleson, 2004). This age range should encapsulate any age difference resulting from the use of standards derived from populations of different ancestry (i.e. European vs. Asian ancestry; Liversidge, 2003). None of the teeth in individual 66-2 are carious (Fig. 4),

and there is no loss of alveolar margin height associated with periodontal disease in the mandible or maxillae. None of the observable deciduous teeth, nor the forming permanent first molars, have hypoplastic enamel defects associated with periods of non-specific stress such as malnutrition or disease.

A number of classification systems have been established for the distinct clinical entity of osteomyelitis of the jaws (i.e. Cierny et al., 1985) of which the Zurich system, based primarily on clinical appearance and radiological features, will be followed herein (see Baltensperger and Eyrich, 2009 for a thorough review). Radiographs were taken using the NOMAD Pro handheld X-ray system.

#### RESULTS

#### **Differential Diagnosis**

There is a huge variety of pathological lesions that occur in the mandible that can overlap considerably in morphological, radiological, and histological appearance, often making diagnosis difficult even in clinical settings (Slootweg, 2010). In this case the differential diagnosis is heavily based upon evidence of endodontic infection, via the cloaca originating from the left deciduous and permanent mandibular canines, as well as a lack of evidence for trauma. The route of infection would have been the left mandibular deciduous canine, which is linked to the forming permanent canine via the gubernacular canal which connects the shared tooth sac, facilitating the transfer of infectious microorganisms (Rodriguez Cordeiro and de Carvalho Rocha, 2005).

Due to differences in the macroscopic and radiologic appearance of the following conditions they have been ruled out of the differential diagnosis: osteosarcoma, Ewing's sarcoma, fibrous dysplasia, and periapical osseous dysplasia. In short, there is no localized tumour-like permeative lytic lesion with a radiological appearance that is 'moth-eaten' or 'sun-burst' as is typical of neoplastic diseases, the radiograph shows no evidence of the presence of fibrous tissue as in the extremely rare fibrous dysplasia, and there is no involvement of the tooth roots or masses of cementum resulting in multiple, circumscribed, non-corticated radiolucencies as in periapical osseous dysplasia, which also differs in that its mean age of onset is after 30 years.



Fig. 1. Lake Baikal, Siberia, and the location of the mortuary site of Shamanka II.

Four additional diseases, described below, are considered more likely options in the differential diagnosis: tuberculosis, noma (gangrene of the face), infantile cortical hyperostosis (Caffey's disease), and periostitis ossificans.

Tuberculosis (TB) is caused by the Myobacterium tuberculosis complex, which is estimated to have appeared 15,000 to 20,000 years ago (Kapur et al., 1994), with an early example from a 9,000village agricultural year-old in Israel (Hershkovitz et al., 2008). Its prevalence in humans increased with animal domestication, eventually becoming endemic in large, sedentary populations across the world. TB osteomyelitic lesions are most common in the spine, hip and knees, with bacilli localized especially in areas of (Aufderheide hematopoietic marrow and Rodriquez-Martin, 1998; Roberts and Manchester, 2010). TB rarely causes cranial lesions, but in those rare cases where it does, it is often in a child (Thijn and Steensma, 1990). However, lesions usually occur in the cranial vault (because of bacilli localization in the diploe), sometimes with hypervascular lesions on the endocranial surface (Mukherjee et al., 2002; Pálfi et al., 2012), and children are more likely to have lesions in several bones (Messner, 1987; Gupta and Singh, 2007), both features lacking in this case. Gadgil et al. (2012) estimate that less than two percent of the roughly five to seven percent of tuberculosis cases that result in skeletal lesions are located in the mandible (<0.10 to 0.14%) (also see Erasmus et al., 1998). Ortner (2003) notes that TB lesions in the mandibulae of children are located near the angle, again dissimilar to the lesion in question. Given the low population density, lack of animal domestication, and lack of evidence for TB in any other Cis-Baikal skeletons from this time period, it is very unlikely the lesion was a result of TB. TB is also marked by more of a destructive (lytic) than proliferative process (i.e., see example in Lewis 2011: 18; Roberts and Manchester, 2010) which does not fit the appearance of the lesion in individual 66-2 with its extensive periostitis and single, small cloaca.

Noma (also known as cancrum oris) results from gangrenous sores in the gingiva caused by a range of fusospirochetal bacteria species (Enwonwu et al., 2000; Baratti-Meyer et al., 2003). It frequently occurs adjacent to carious or periodontal lesions, in malnourished or ill children,



**Fig. 2.** Grave 66, Shamanka II. Individual 66-1 is a 25 to 35-year-old female; Individual 66-2 is a 1.75-year-old infant.

most often between the ages of two to six years, resulting in severe disfigurement with a high mortality rate (Enwonwu et al., 2000). It is unlikely the lesion in individual 66-2 is the result of noma because noma spreads quickly, causing extensive jaw and and facial destruction, and lacks a bone-producing aspect. In addition, osseous noma lesions most commonly originate in the molar area, and sequestrum almost always result, which do not fit the appearance of the lesion in question.

Infantile cortical hyperostosis (Caffey's disease) is a rare inflammatory disease characterized by soft-tissue swelling usually with accompanying periosteal hyperostosis, most often occurring in the mandible, but also in the clavicles, ribs, scapulae and/or long bones (Caffey and Silverman, 1945). It is most often found in infants less than six months of age, after which it frequently spontaneously resolves with complete recovery by two to three years of age (MacLachlan et al., 1984). While age of occurrence is not that dissimilar, lesions usually occur at the angle or ramus of the mandible and are characterized by new lamellae layers with an onionskin appearance (see Lewis and Gowland (2009) for archaeological examples), which is different from the lesion in individual 66-2. As well, because infantile cortical hyperostosis does not have a purulent aspect (which would cause a cloaca), it is not a good diagnostic fit for this lesion.

Finally, periostitis ossificans (also called Garre's osteomyelitis) is a rare non-purulent form of osteomyelitis with intense proliferation of the periosteum resulting in new bone formation, often with new lamellae layers giving an onionskin appearance (Felsburg et al., 1990; Belli et al., 2002) that can affect an extensive part of the jaw. It usually affects individuals before the age of 25, primarily children and adolescents (Felsburg et al., 1990; Belli et al., 2002). The lateral, toothbearing aspects of the mandible (usually below first molar) are most affected, but important for this diagnosis is that it rarely crosses midline. It is similar to infantile cortical hyperostosis in that purulent discharge is rare, meaning cloacae are unlikely (although see Gonclaves et al., 2002), making this diagnosis a poor fit to the lesion morphology of individual 66-2.

Thus, none of the aforementioned diseases are a good match for the appearance of the lesion in individual 66-2. Rather, the disease classification that fits most closely is osteomyelitis with proliferative periostitis. Osteomyelitis is an infection of the bone by purulent microorganisms (Resnick, 2002) beginning in the medullary cavity and Haversian systems and extending to involve the periosteum. The Zurich system classifies it into three major categories: 1) acute osteomyelitis, 2) secondary chronic osteomyelitis, and 3) primary chronic osteomyelitis. Categories 1 and 2, acute and chronic, are the same disease separated by an arbitrary time limit of four weeks after disease onset, as recommended by Marx (1991) and Mercuri (1991), and subsequently widely adopted by clinicians (i.e. Lew and Waldvogel, 2004). The third category, primary chronic osteomyelitis refers to a rare, nonsuppurative, chronic inflammation of the jaws of unknown cause and thus does not apply to this case. The extent of the lesion in individual 66-2 suggests the disease had



Fig. 3. Anterior view of the mandible of Individual 66-2, Shamanka II, showing pathological lesion.



**Fig. 4.** Superior view of the mandible of Individual 66-2, Shamanka II, showing status of dental eruption and the lack of carious or periodontal lesions.

entered a chronic phase (category 2). Prior to antibiotic treatment, mandibular osteomyelitis usually presented in the secondary chronic phase (Wilensky, 1932). Compromised local blood supply is an important factor in the establishment of osteomyelitis, since immune cells and oxygen cannot reach the infected area facilitating the growth and spread of microorganisms, especially anaerobes (Bruder et al., 2009). The acute to chronic phase is often due to microorganism biofilm colonization of necrotic bone leading to inflammation and a suppurative response (Bruder et al., 2009). Age does not have a major role on the incidence of maxillary osteomyelitis (Baltensperger et al., 2004).

Chronic osteomyelitis of the skull is found most often in the mandible, even in comparison to the maxilla (see Baranoff, 1934). The lesion is usually focused in one anatomical site (Baltensperger et al. 2004; Lew and Waldvogel, 2004), a point which differs to a certain extent from the appearance of the lesion in individual 66-2, although in this case it is mostly the periosteal reactive bone that is non-localized with the deformation of the inferior border of the mandible located mostly on the left side nearest the cloaca. The partially bilaterally symmetrical appearance of the lesion in individual 66-2 is suggested to have occurred because the location of the cloaca terminus is close to the midline (its direction is angled from the left canine socket to the area under the left central incisor) and exudate discharge would have resulted in substantial periosteal elevation (see below) that allowed the spread of microorganisms. As to why the infection spread across the midline and became secondarily situated around the right incisor and canine sockets (as opposed to spreading posteriorly towards the left molars), possibly the active eruption of the deciduous canines caused antecedent loosening of the periosteum that presented little barrier against infiltrate spread. It is only speculative, but the periosteum around the left first molar may have been more firmly attached to the bone thereby presenting a more effective barrier because that area underwent tooth eruption at an earlier age. Radiologically there is no evidence of a sequestrum, which may be due to the individual's young age precluding development, however Ortner (2003) notes that osteomyelitis of the mandible rarely results in the formation of a sequestrum. Thus, these factors are congruent with a diagnosis of osteomyelitis for individual 66-2.

The presence of new periosteal bone is also consistent with a diagnosis of osteomyelitis, as it occurs relatively commonly as a result of perio-



**Fig. 5.** Radiograph of mandible of Individual 66-2, posterior to anterior plane. Box demarcates the cloaca running from the left permanent canine bud to the inferior mandibular border.

steal elevation prompting chronic inflammatory cells and proliferating fibroblasts to form reactive bone (Resnick, 2002). Mandibular bone is known for exuberant peripheral reactive bone formation, a feature that is rare in the maxilla (Betts et al. 1996). The appearance of periosteal reaction is determined by the intensity, aggressiveness, and duration of the underlying insult (Rana et al., 2009). The periosteum in children is more active and less adherent to the cortex than in adults. Thus, periosteal reaction can occur earlier and appear more aggressive in children than in adults (Rana et al., 2009). In individual 66-2 the reactive periosteal bone exhibits a laminated and separated pattern, suggesting alternating periods of reaction that were more or less aggressive. In sum, for infant number 66-2 from the site of Shamanka II, the macroscopic and radiological appearance of the mandibular lesion is most consistent with a tooth-germ related purulent osteomyelitic infection with concomitant new periosteal bone formation.

#### DISCUSSION

#### Etiology

While not frequently noted, osteomyelitis of the mandible has been documented in a range of archaeological skeletal samples (Roney, 1966; Gregg and Gregg, 1987; Khudaverdyan, 2011). At Shamanka II osteomyelitic lesions are rare (Lieverse, 2010); no other individuals from this site, nor from others in the same area and timeperiod, had osteomyelitis of the jaws. Today osteomyelitis of the jaw is common enough to warrant categorization as a distinct clinical entity, however, the appearance of the lesion in individual 66-2, and the proposed source and route of infection are rare (Baltersperger and Eyrich, 2009). Most osteomyelitic infections of the jaws arise from carious or periodontal infections, with dental biofilm recognized as a possible but less likely option (Brady et al., 2006). Osteomyelitis can arise secondarily, spread to the jaws hematogeneously, but this is unlikely in this case as the cloaca pinpoints the infection location and none of the other bones have lytic lesions. The location of the cloaca, arising off the forming left permanent mandibular canine, and the lack of caries or vertical or horizontal alveolar bone loss, suggest that oral microorganisms were responsible for the infection.

Clearly, the microorganisms responsible for infection in this individual will not be preserved so it is not possible to isolate and identify specific bacteria. However, oral bacterial genera that have been implicated in osteomyelitis in the jaws of children are: Staphylococcus (especially S. aureus), Streptococcus, Actinomyces, and Enterococcus (Zbinden, 2009). Anaerobic bacteria include Gram-positive and negative cocci and Gramnegative rods (Zbinden, 2009). Normal healthy oral flora also have the potential for pathological effects (Willet et al., 1991); approximately 700 bacterial species have been identified in the human oral cavity (Aas et al., 2006), although a healthy individual will have a limited number in their oral cavity at any one time (Zbinden, 2009).

While it is well documented that huntergatherer and agricultural populations experience a different suite of infections from each other because of differences in population size and density (Armelagos and McArdle, 1975; Dobson and Carper, 1996), infections resulting from dental biofilm may be similar as they are likely not affected by these population parameters. Furthermore, oral microorganisms have great antiquity (Schultz, 1956; Caufield et al., 2007). However, it is not well known what effect diet has on the types, prevalence, and degree of pathogenicity of oral microorganisms that cause osteomyelitis. Analysis of a recent hunter-gatherer group from

Central Africa found high diversity in oral microbial communities and many previously unreported genera (Nasidze et al., 2011) suggesting the human oral cavity can harbour population specific microbiomes of substantial diversity. As well, fish-based diets have been shown to expose humans to a unique range of pathogens (Novotny, 2004). Thus, it is possible the hunterfisher-gatherer people of the Lake Baikal area had a rather distinctive oral environment. Although, given the rarity of osteomyelitis of the jaws, it is not expected that they experienced an abnormal level of pathogenicity. Future research should investigate if markers of oral bacteria in dental calculus can be identified in Cis-Baikal huntergatherers, as has been done with some success in ancient and modern hominins (Moorer et al., 1993; Arensburg, 1996). Furthermore, research that explores the range of variation in oral bacteria between groups with different subsistence patterns could be very useful in improving our understanding of the causes of dental disease.

It is also important to consider that breastfeeding can inhibit Staphylococcus and Streptococcus infection (Welsh and May, 1979). Stable nitrogen isotope ( $\delta^{15}N$ ) values of bone collagen samples are used to determine the breastfeeding status of subadults. The  $\delta^{15}N$  value of individual 66-2 is elevated suggesting breast-milk was still a major source of protein. This is in keeping with a mean age of complete weaning of three to four years in the Shamanka II sample (Waters-Rist et al., 2011). The fact that the infant did not succumb to acute infection, but rather survived into the period of chronic infection whereby a distinctive bony lesion was formed, may have been partially the result of the immune boosting effects of breastfeeding. Ultimately however, the osteomyelitic infection was likely the cause of death in this infant. Osteomyelitis of the mandible is frequently associated with bacteraemia, which can be lethal (Aufderheide and Rodriguez-Martin, 1998).

#### CONCLUSION

An infant from a 7,000-year-old archaeological site in Siberia presented with a mandibular lesion most consistent with a diagnosis of osteomyelitis with periosteal new bone, arising from infection of the canine tooth germ possibly from normally non-pathogenic oral bacteria. This case study extends the time depth of our knowledge of osteoosteomyelitis of the jaw and highlights a rather rare infection source and route.

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