A HISTOLOGICAL EXAMINATION OF ENAMEL MICROEVOLUTION OVER 2000 YEARS OF HUMAN HISTORY USING BRITISH POPULATIONS

Christopher Aris

Skeletal Biology Research Centre

School of Anthropology and Conservation

University of Kent

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Abstract

The study of permanent enamel thickness and secretion rates of ameloblasts have yielded insights into the evolution of enamel when compared between hominin species. However, the study of modern human permanent tooth enamel has received far less attention, particularly regarding daily secretion rates (DSRs) and changes over time between populations. Moreover, enamel thickness and DSRs have rarely been analysed in conjunction for human samples. The research here uses dental histological thin sections to examine inner, mid, and outer DSRs for lateral and cuspal enamel, and the average (AET), relative (RET), cuspal (CT), and lateral (LT) thickness, for permanent first molar, upper canine, and upper first incisor crowns, from five British populations spanning the last 2000 years of history. The populations cover the Roman period (1-4AD) to modern day. A total of 265 teeth were analysed: first molar (n=115), upper canine (n=69), and upper first incisor (n=81). Results display consistent and significant trends towards decreasing DSRs from the ancient to modern populations. This was observed in all first molar cuspal regions (inner p<0.001, mid p<0.001, and outer p<0.001) and the inner and outer lateral regions (inner p=0.01 and mid p<0.001), in all upper first incisor cuspal and lateral regions (all p<0.001), and in all upper canine cuspal and lateral regions (all p<0.001). Enamel thickness features revealed less inter-population variation in first molars. The Early Anglo-Saxon RET was significantly larger than that of the Late Anglo-Saxon (p<0.001) and Medieval (p<0.001) populations. The Modern-day population LT was significantly thicker than the Roman populations (p=0.04). These differences allude to potential differences in diet between these populations. Alongside the differences seen between populations, the data provides the first major evidence for changes in the daily rate of enamel growth in human permanent dentition between populations. That similar variation was not observed in enamel thickness suggests thickness and growth can vary significantly and independently between human population.

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School of Anthropology and Conservation

University of Kent

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CHAPTER 1: INTRODUCTION, THESIS STRUCTURE, AND RESEARCH QUESTIONS

1.1 Introduction

Within dental anthropology there is a large volume of research regarding enamel. Studies concerning human samples most commonly analyse enamel within an intra-population context (e.g. Dean and Scandrett, 1995; Reid et al., 1998; Mahoney, 2010, 2013), or as a comparative basis for hominoid dental analysis (e.g. Bromage, 1991; Dean et al., 1993a, 1993b; Reid and Ferrell, 2006). The studies looking at differences in human enamel features between populations are fewer in number, and have primarily investigated the variation between populations of different geographic origin (Kajiyama, 1965; Fitzgerald, 1995; Reid and Dean, 2006; Reid and Ferrell, 2006; Smith et al., 2006, 2007a). Additionally, there has been very little research conducted into whether features of human enamel could vary over time within a temporal context. Those that have been conducted used archaeological samples to investigate for variation in enamel thickness (Smith et al., 2007a, 2010; Le Luyer et al., 2014; Le Luyer and Bayle, 2017). Until now, no such research has addressed modern clinical dental samples or studied enamel growth patterns. Research presented here will provide such analysis, by presenting two peer reviewed published manuscripts involving analyses using dentition from British populations spanning the last 2000 years, and additional discussion as to how the data gathered from the British populations compares to previously researched populations. The aims are to expand upon our understanding of the evolutionary plasticity of modern human permanent enamel. These aims will be met by investigating differences in enamel thickness and growth rates through daily secretion rates (DSRs) between populations within a temporal transect, across the tooth row, and between biological sexes.

Chapter 2 provides a detailed overview of general dental anatomy, compromised of an detailed description of enamel anatomy and composition, and descriptors of each stage of enamel growth and formation (amelogenesis). Chapter 3 consist of a review of existing literature will be conducted regarding all relevant dental histological research, all intraspecific studies that have been conducted on human dentition, and a brief assessment of the relevant literature regarding non-human primates and hominins. Chapter 4 will further review the enamel feature data collection methods associated with the novel research presented later. This literature review will serve to contextualise the subsequent novel research, and highlight why temporal analysis of a large human dental data set is crucial to ongoing research in anthropology and bioarcheology. After reviewing the relevant literature, the histological methods employed to analyse the dental samples, and gather data from the produced sections, will be detailed in Chapter 5. This methods section will also include

a profile analysis of each dental collection utilised, including analysis of their geographic region within the United-Kingdom, their date of origin, and relevant contextual information.

After the review and methods chapters, the published manuscripts will be presented. These chapters represent a streamlined version of the published articles with sections already covered in the literature review and materials sections removed. The full, unedited, and formatted articles can be found in Appendix C. Please note both the streamlined and full versions include edits made following the peer review process. Chapter 6 (Aris et al., 2020b) addresses temporal variation in the two-dimensional thickness and DSRs of permanent first molars. Chapter 7 (Aris et al., 2020a) addresses the daily growth patterns, but will focus on the anterior dentition (specifically first upper first incisors and upper canine) of the same British populations from the last 2000 years. Following the two novel pieces of research, a discussion (Chapter 8) and final conclusions (Chapter 9) will be presented. Chapter 8 will further bring together the theories and conclusions of each of the research project chapters, thus allowing for any observed changes in first molar dentition to be cross-examined with those from the upper anterior tooth analyses, and vice versa. Chapter 9 will present findings regarding: the inter- and intra-population variations observed in enamel thickness measures and regional DSRs, the exact relationship therein between enamel thickness and DSR variation, the similarities and/or dissimilarities of variations between tooth types, and the future research required to follow on from the discoveries presented here.

In regards to ethical considerations, all sampling procedures followed the guidelines for destructive sampling of human remains outlined by Mays and colleagues (2013). This primarily involved only analysing one tooth per individual set of remains, alongside the ethical treatment and storage of all human remains throughout the research process. The use of only one tooth per individual was also done on request from the institutions which curated the material. These steps were followed for the research of both the archaeological and modern-day clinical samples. In addition, in order to sample the modern-day samples ethical approval for histology research was obtained from the United Kingdom National Health Service research ethics committee (REC reference: 16/SC/0166; project ID: 203541).

1.2 Research questions

Using the aforementioned approach and structure, this research project will aim to answer the following research questions:

1. **Have enamel growth rates of permanent first molars and upper anterior teeth varied between populations over time?**
2. **Have enamel growth rates of upper anterior teeth varied between biological male and female groups between and within populations?**
3. **How have enamel growth rates varied between tooth types?**
4. **Has enamel thickness of permanent first molars varied between populations over time?**
5. **Do any variations in enamel growth and thickness correlate?**

CHAPTER 2: BACKGROUND – DENTAL ANATOMY AND AMELOGENESIS

2.1 Enamel anatomy

2.1.1 Enamel structures and components

Enamel is a composite mineralised tissue, formed of mineral and organic components, which cover the primary dentine of a tooth, creating the outer portion of the tooth (see Fig. A.2; e.g. Bowes and Marray, 1935; Helmcke, 1967; Weatherell et al., 1974; Boyde, 1997; Berkovitz et al., 2002; Antoine et al., 2018). While the shape and function of enamel varies between tooth types, in general it forms thickest at the cusps of a tooth and thinnest at the cervix and serves to protect the tooth throughout life, aided by its organisational structure which creates a hard and wear resistant surface (e.g. Helmcke, 1967; Bodye, 1997; Berkovitz et al., 2002:102).

Calcium hydroxyapatite ($Ca_{10}(PO_4)_6(OH)_2$) mineral accounts for between 90-92% of enamel volume and around 96% by weight (e.g. Bowes and Marray, 1935; Helmcke, 1967; Beynon et al., 1998; Berkovitz et al., 2002; Nanci, 2003; Antoine and Hillson, 2016; Antoine et al., 2018). The concentration of hydroxyapatite increases moving apically away from the enamel dentine junction (EDJ). Enamel hydroxyapatite forms as crystallites 68-70nm in width and 25-26nm thick (Berkovitz et al., 2002; Nanci, 2003). As a result of being highly mineralised, enamel is further able to withstand both shearing and impact forces whilst wearing down very slowly. This resistance is vital as enamel cannot be repaired or maintained in any way by the body (e.g. Bowes and Marray, 1935; Boyde, 1997; Berkovitz et al., 2002).

Of the remaining 8-10% of enamel volume 6-8% consists of water. The distribution of water within enamel is linked to the relative concentration of hydroxyapatite (Bowes and Marray, 1935; Helmcke, 1967; Berkovitz et al., 2002; Antoine et al., 2018). Near the EDJ there are a lower number of organised crystallites and higher porosity (Nanci, 2003). This results in water filling the spaces between crystals surrounding the organic components of the enamel as well as forming a hydration layer coating the hydroxyapatite crystallites (Helmcke, 1967; Nanci, 2003). Near the occlusal surface, water volumes are lower as crystal concentration and organisation increases (e.g. Boyde, 1997; Helmcke, 1967; Berkovitz et al., 2002; Nanci, 2003). Here the water is either trapped in the defects of the crystalline matrix, or coating the crystallites (Weatherell et al., 1974; Berkovitz et al., 2002; Nanci, 2003). The last 2-4% of enamel is an organic matrix. The majority of this organic mixture is a combination of amino acids, peptides, and other small molecules which in total make up between

50-90% of the matrix's volume (Helmcke, 1967; Berkovitz et al., 2002; Nanci, 2003). The amino acids are primarily glycine, alanine, and glutamic acid (Berkovitz et al., 2002:104).

The proteins present within enamel are amelogenins and non-amelogenins. Amelogenins compromise around 90-95% of all enamel proteins which spread across the whole of the enamel, forming a gel like substance from which ions can rapidly spread to produce the crystalline prism structure of enamel (see Fig 2.1; Weatherell et al., 1974; Boyde, 1989; 1997; Eisenmann, 1998; Berkovitz et al., 2002; Nanci, 2003; Robinson et al., 2003; Smith and Nanci, 2003; Fukumoto et al., 2014). Amelogenins are spread across enamel with the concentration of non-amelogenin proteins highest at the EDJ, specifically within enamel tufts (Berkovitz et al., 2002; Nanci, 2003; Boyde, 1997). These tuft regions are linear structural formations which run the length of the inner third of enamel, originating at the EDJ. Non-amelogenins (or enamelins) are far less in abundance, comprised by ameloblastin and tuftelin, making up the remaining 5-10% of enamel proteins (Berkovitz et al., 2002; Nanci, 2003).

2.1.2 Enamel prisms

Mature hydroxyapatite prisms make up the majority of structure enamel at a cellular level. Each prism is made up of approximately 10,000 near-parallel and regulated hydroxyapatite crystallites compacted into a thin rod with a diameter of 5-6µm and a length of 2.5mm (e.g. Boyde, 1997; Berkovitz et al., 2002; Nanci, 2003; Antoine and Hillson, 2016). The grouping of the crystallites into prisms accounts for the relative flexibility of individual crystals. Due to their micro-porosity and surrounding organic components single crystals possess too much bendability alone, grouping thereby strengthens and stiffens them and thus also strengthens the overall structure of enamel (Boyde, 1997). These grouped prism rods run apically from the EDJ to the occlusal surface (Helmcke, 1967; Boyde, 1989; Nanci, 2003; Robinson et al., 2003) and have been tracked using both 2D and 3D methodologies (Smith and Tafforeau, 2008). Prisms can vary at the angle at which they reach their apex, but typically reach the outer enamel surface at an angle around 120^{o} (Hillson, 1996; Antoine and Hillson, 2016). Prisms also tend to orientate in near-parallel groups, and always originate perpendicular to the EDJ (Helmcke, 1967; Boyde, 1997; Berkovitz et al., 2002). The centre of the prism is highly organised with little angular deviation, whereas the boundaries of the crystallites can deviate from anywhere between 40^{0} and 60^{0} in angularity (Helmcke, 1967; Eisenmann, 1998; Berkovitz et al., 2002). This creates an optical effect which can be observed using microscopic analysis (Helmcke, 1967; Boyde, 1997; Berkovitz et al., 2002). Further optical effects can be caused

by the variation in the pathways of enamel prisms. While they all run from the EDJ to the outer enamel surface they will regularly undulate around their long plane in groups around other groups of prisms, this is known as prism decussation (e.g. Boyde, 1989; Antoine et al., 2018). While decussation is reduced with distance from the EDJ and in lateral regions of enamel, it can make following single prisms across their length difficult (Boyde, 1989; Antoine et al., 2018).

2.1.3 Aprismatic enamel

Aprismatic enamel (also known as non-prismatic enamel; e.g. Boyde, 1997) is a small layer of prism-less enamel which can be observed at the outer surface of all unworn dentition (Helmcke, 1967; Mukherjee et al., 2018). Aprismatic crystallites are aligned perpendicular to the occlusal surface of the tooth and at strict right angles to each other. In permanent teeth the thickness of aprismatic enamel ranges from 20-70µm (Berkovitz et al., 2002; Mukherjee et al., 2018). The degree of thickness is dependent on the specific part of the tooth the enamel is formed at, being thicker at the cusps and thinner at the cervical border (Berkovitz et al., 2002). Even though it is not clustered into prisms, aprismatic enamel is still formed primarily of hydroxyapatite crystallites and has a low concentration of organic material due to the loss of boundaries between prisms and Hunter-Schreger bands (Boyde, 1997; Berkovitz et al., 2002; Nanci, 2003). The formation of this type of enamel is a result of ameloblasts losing their Tomes' processes during the final stages of enamel formation (see *2.2 Amelogenesis* and Fig 2.5b).

2.1.4 Incremental enamel lines

The development and growth of enamel involves periods of high activity which alternate with periods of dormancy (Helmcke, 1967; Berkovitz et al., 2002). As a result of this rhythmic process enamel develops a series of incremental lines which appear under histological and radiographic analysis (Berkovitz et al., 2002). The main types of lines which can be observed are short period lines (cross striations) and long period lines (lines of Retzius or enamel striae).

<u>*Cross Striations*</u> – These form at right angles to enamel prisms running across their transverse long axis, appearing as brighter or darker bands compared to the majority of the prism volume (Helmcke, 1967; Boyde, 1997; Berkovitz et al., 2002; Antoine and Hillson, 2016; Antoine et al., 2018). These lines form due to the diurnal circadian rhythms of ameloblasts, as the daily pattern of enamel secretion causes the formation of lines between periods of activation (see *2.2 Amelogenesis*)

(Helmcke, 1967; Bromage, 1991a; Dean, 1995; Dean and Scandrett, 1996; FitzGerald, 1998; Reid et al., 1998; Mahoney, 2008; Lacruz et al., 2012; Zheng et al., 2013; Antoine et al., 2018). Cross striations are observable as being brighter or darker under light microscopy due to the variation in the carbon dioxide available for incorporation into the enamel mineral (Boyde, 1997; Antoine et al., 2018). Variation in carbon dioxide variability is believed to correlate with variation in metabolic activity (e.g. Boyde, 1989; Antoine et al., 2018). The resulting differences in chemical composition between cross striations, in comparison to enamel matrix propagated during the secretion of new matrix, results in different refractive indexes and thus difference in appearance through light microscopy (Boyde, 1989; 1997; Antoine et al., 2018).

Past research has shown that, due to the pattern of cross striation development, they can be utilised to reconstruct an accurate chronology of enamel growth (Antoine and Hillson, 2016; Antoine et al., 2018). Cross striations are relatively consistent in their organisation, observed to form in 2.5-6μm intervals, with increasing distance occurring with proximity to the outer enamel surface as enamel matrix secretion speed increases (Berkovitz et al., 2002; Antoine and Hillson, 2016; Antoine et al., 2018; Aris et al., 2020a; 2020b). This difference is wider and more equal between spaces in the main body of the enamel at around 4μm, with lines forming more closely together near the EDJ (as close as 2μm) (Berkovitz et al., 2002). See Figure 2.1A.

Retzius Lines – Retzius lines, first described by Retzius (1837), form obliquely across the hydroxyapatite enamel prisms along the front of secreting ameloblasts. Like cross striations, Retzius lines develop from near the EDJ and span across the enamel to the occlusal surface of the tooth (e.g. Helmcke, 1967; Bromage, 1991c; Dean and Scandrett, 1996; Reid et al., 1998). These lines are less abundant than cross striations and form as a single structure across the entire circumference of enamel (Helmcke, 1967; Bromage, 1991c; Berkovitz et al., 2002; Antoine et al., 2018). In humans an average of 6-10 (mode = 8) cross striations form between the formation of two Retzius lines, resulting in an average 25-30μm gap between them (Bromage, 1991c; Dean and Scandrett, 1996; Reid et al., 1998; Schwartz et al., 2001; Reid and Ferrell, 2006; Mahoney 2008; Antoine and Hillson, 2016; Antoine et al., 2018). This can vary at the dental cervix where less enamel is secreted slower meaning the space between Retzius lines can be as small as 15-20μm (Berkovitz et al., 2002; Guatelli-Steinberg and Reid, 2010).

Whilst the distance between Retzius lines and the time between their formations is observed and predicted, their actual size as individual structures can vary with accentuated lines forming during periods of disturbance to the mineralisation of enamel (Helmcke, 1967; Risnes, 1986, 1990). Where Retzius lines appear accentuated and enlarged it is theorised to be the result of an

interruption to the secretory stage of amelogenesis (Helmcke, 1967; Berkovitz et al., 2002). In these cases the lines are referred to as Wilson Bands (Wilson and Schroff, 1970; Rose et al., 1978; Berkovitz et al., 2002). The exact etiology for Wilson Bands is unknown although there has been some evidence to suggest that these are the internal structures associated with linear enamel hypoplasia lines, with the accentuated form being the result of a period of stress (Gustafon, 1959; Rose et al., 1978; Hillson and Bond, 1997; Mahoney, 2008:133). Figure 2.1B.

Retzius lines have morphological manifestations observable on the external surface enamel. The presence of these lines on the enamel surface produces smooth groove-like structures which are separated by clear ridges called perikymata (Helmcke, 1967; Bromage and Dean, 1985; Dean, 1987; Risnes, 1990; Dean and Scandrett, 1996; Reid et al., 1998; Shellis, 1998; Reid and Ferrell, 2006; Mahoney, 2008; Guatelli-Steinberg and Reid, 2010; Guatelli-Steinberg et al., 2012; Antoine et al., 2018). The distance between these grooves is the same as that of the interior Retizus lines which define them.

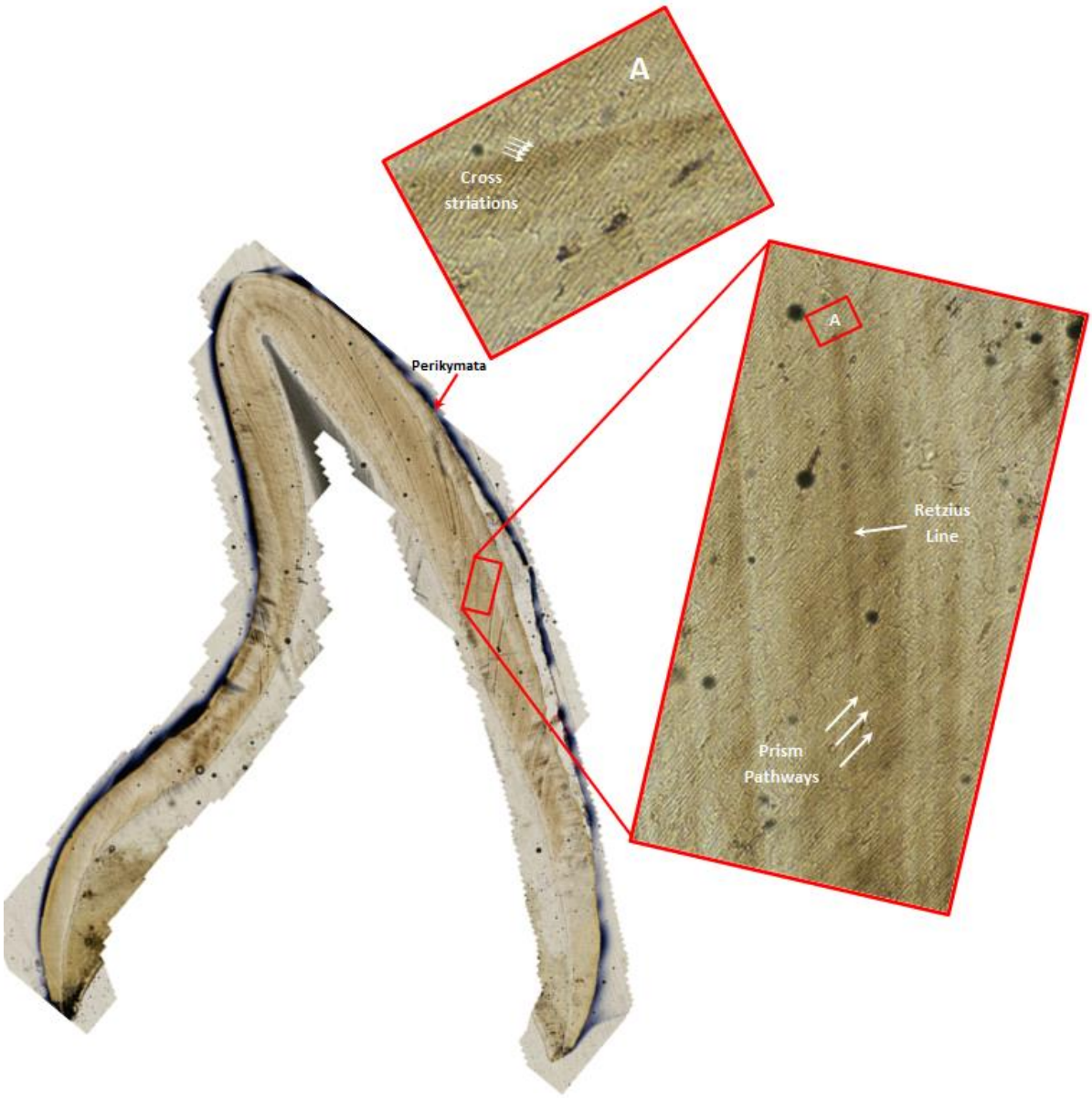

Fig. 2.1. *Cross-section of a Roman upper first incisor displaying the appearance of interior enamel formations and prism pathways under microscopic observation. The two superimpositions highlight the cross striations and the prism pathways they follow.*

2.1.5 Other enamel structures

Neonatal Lines – the largest Retzius line present in a tooth which forms at the point of birth, marking the boundary between pre-natal and post-natal enamel growth (Rushton, 1933; Schour, 1936; Christensen and Kraus, 1965; Kraus and Jordan, 1965; Beynon et al., 1991; Schwartz et al., 2005; Birch and Dean, 2014; Antoine and Hillson, 2016). Neonatal lines share the same function and

developmental process as other Retzius lines, but are marked by thicker morphology and darker appearance (Helmcke, 1967; Berkovitz et al., 2002).

Enamel Spindles – round structures with a similar shape to enamel tufts but far narrower in size. Enamel spindles develop to around 8μm in diameter and extend into the enamel for roughly 25μm originating at the EDJ (Berkovitz et al., 2002). Unlike enamel tufts, enamel spindles do not align with enamel prisms and are believed to be a formation defect caused by ondotoblast processes becoming compacted between ameloblasts during early dental development (Berkovitz et al., 2002; Nanci, 2003).

Enamel Lamellae – sheet-like formations which form along the diameter of enamel originating either at the EDJ or the outer portions of primary dentine (Bodecker, 1953; Berkovitz et al., 2002). Like enamel spindles they are narrower than tufts, but are longer and more expansive than both (Berkovitz et al., 2002). Enamel lamellae represent regions of hypo-mineralisation, a defect which appears as a crack-like structure in the enamel when overserved in a longitudinal histology cross section (Berkovitz et al., 2002). Differentiation can be made between enamel lamellae and actual cracks to the enamel, as during demineralisation cracks will break down whereas the lamellae will retain their form. Enamel lamellae are the result of prisms not completing maturation, resulting in a higher level of porosity and organic material (Berkovitz et al., 2002; Farooq et al., 2021).

Enamel Cracks – narrow fissure-like structures which appear on the surface of the enamel and are the external manifestation of enamel lamellae (Berkovitz et al., 2002; Bajaj and Arola, 2009). They range in distance along the surface of the enamel and can reach the occlusal and/or incisal edge of the tooth; this length is usually dictated by the enamel lamellae they represent (Berkovitz et al., 2002). It should be noted that there is a difference between enamel cracks and cracks in the enamel. Enamel cracks are an external, structural morphology, whereas cracks in the enamel are the result of trauma and can be both internal and/or external in their location (Berkovitz et al., 2002; Bajaj and Arola, 2009).

Tomes' Process – a morphological feature developing on the surface of ameloblasts parallel to the EDJ during the early stages of enamel secretion (Boyde, 1997; Berkovitz et al., 2002). These pronged pyramidal cellular organs act to guide the deposition of enamel matrix and form walls of interprismatic regions, and are ultimately responsible for the final prismatic structure of mineralised enamel (Berkovitz et al. 2002).

2.1.6 Enamel defects

A number of defects can affect the healthy growth of enamel. Only teeth deemed healthy were used in the novel research presented here. Below are the common enamel features that, when present, were used to classify teeth as unhealthy and thereby no suitable for use in this research.

Enamel Hypoplasia – a linear formation similar in appearance to a perikymata groove, forming as a part of the external surface of enamel (e.g. El-Najjar et al., 1978; Skinner and Hung, 1989; Berkovitz et al., 2002). Hypoplasia lines are much more exaggerated and less organised than perikymata grooves (Berkovitz et al., 2002). These lines form as a result of interference to the active section of enamel matrix during amelogenesis, and while it can be difficult to identify the exact cause of individual cases of enamel hypoplasia formation, the interference is required to influence the chemical composition of the matrix secreted at the time (e.g. El-Najjar et al., 1978; Berkovitz et al., 2002). Such interferences have been linked to external stressors, including psychological events and physiological events associated with malnutrition, disease, and/or drug use (e.g. El-Najjar et al., 1978; Rose et al., 1978; Boldsen, 2007; Guatelli-Steinberg et al., 2012; Miszkiewicz, 2015; Wright et al., 2015). Should the event occur during a stage of development of multiple teeth, then a hypoplasia line can form across multiple teeth denoting a single event. The impacts of stress, as observable on dentition, has been linked to slower crown formation (e.g. Guatelli-Steinberg and Lukacs, 1999; Fitzgerald and Saunders, 2005; Reid and Dean, 2006; Aris and Street, 2021). As a result, while direct comparison between the presence of enamel hypoplasia and internal growth of the same tooth have yet to be made, the potential as alluded to from past research warrants not analysing teeth with hypoplasia lines in this project.

Enamel Hypocalcification (enamel/amelogenesis imperfecta) – a morphological malformation of enamel where a lack of calcium and overabundance of protein in enamel results in imperfect mineralisation (Berkovitz et al., 2002). Enamel hypo-calcification occurs when there is interference during the enamel maturation, causing the prism structures to form incompletely (Robinson et al., 2003; Simmer et al., 2014; Wright et al., 2015). This results in both the increased concentration of organic material due to the loss of an organised structure, as well as pitted legions and reduced form of surface enamel. (Mangum et al., 2010; Simmer at al., 2010; Wright et al., 2015).

2.2 Amelogenesis

Amelogenesis refers to the formation of enamel through the actions of ameloblasts cells. Amelogenesis is spilt into five descriptive stages, each defined by a different period of an ameloblast's life cycle:

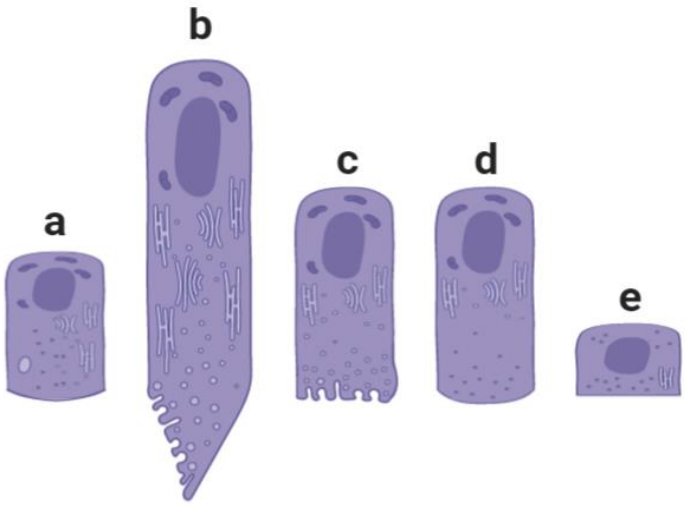

Fig. 2.2. *Figure displaying the life cycle of an ameloblast throughout the stages of amelogenesis:* ***a.*** *presecretory stage,* ***b.*** *secretory stage, where the cell possesses a triangular Tomes' process (seen on the bottom edge)* ***c.*** *transition stage where the cell's Tomes' process has begun to degrade,* ***d.*** *maturation stage, and* ***e.*** *post-maturation stage. Image produced using the BioRender tool set (*https://biorender.com/*).*

2.2.1 Presecretory stage

This stage is all ameloblast activity prior to the secretion of any enamel matrix (Berokvitz et al., 2002). The early presecretory stage involves the differentiation of mesenchymal cells into pre-ameloblasts within the internal enamel epithelium (Boyde, 1967; 1989; 1990; Berkovitz et al., 2002; Jiang et al., 2014; Antoine and Hillson, 2016). This occurs during the earliest stage of dental development. The first cells to differentiate are those positioned at what will later become the dentine horn apex, and continues down the enamel dentine junction (EDJ) towards to the dental cervix (Boyde, 1989; Berokvitz et al., 2002; Nanci, 2003; Smith and Nanci, 2003). These cells begin cuboid in shape before becoming polarised with their large and ovoid nuclei moving to the end of

the cell. This morphological change displaces all other organelles to the other end of the cell adjacent to the future EDJ (Boyde, 1967; 1989; 1990; Berkovitz et al., 2002; Jiang et al., 2014).

Once differentiated the pre-ameloblasts become a sheet-like structure which covers the EDJ (e.g. Boyde, 1997; Berkovitz et al., 2002 Antoine et al., 2018). This sheet of cells then begin secreting primary enamel matrix as they move away from the dentine in a perpendicular trajectory (Boyde, 1997; Antoine et al., 2018). This initial matrix is unorganised and is formed of small sheet of crystals rather than organised prisms (Boyde, 1997; Berkovitz et al., 2002; Nanci, 2003). After this early secretion begins it is typically between eight to ten days before regular cross-striations are formed (Boyde, 1997). This early secreted matrix serves to anchor the enamel layer to the primary dentine at the EDJ. Once the crystals are secreted the pre-ameloblasts undergo morphological changes, becoming more rectangular in shape and developing a pole-like structure which will later form a pyramidal Tomes' process (see Fig 2.5a-b) (Boyde 1997; Berkovitz et al., 2002). This transition marks the ending of the pre-secretory stage of amelogenesis.

2.2.2 Secretory stage

At the onset of the secretory stage, pre-ameloblasts have fully matured into ameloblasts capable of secreting true enamel matrix (Berkovitz et al., 2002). The ameloblasts are now identified by large strands of rough endoplasmic reticulum (RER) which form parallel to the long axes of the cells (Boyde 1997; Berkovitz et al., 2002; Jiang et al., 2014). These RER begin to produce enamel proteins which will later be secreted to form prismatic enamel matrix (Boyde, 1989; Berkovitz et al., 2002). The ameloblasts also develop a large number of Golgi apparatus at this stage, form along the long axis of the cell. These structures act to package the enamel proteins into secretory granules which then migrate to the newly formed Tomes' process (see Fig. 2.5b) (Berkovitz et al., 2002).

As the secretory stage begins, a thin layer of enamel matrix is secreted by the ameloblasts as they move apically away from the EDJ and towards the future occlusal surface of the tooth (Boyde, 1997; Berkovitz et al., 2002; Jiang et al., 2014; Antoine et al., 2018). As this initial secretion completes the rod of the cell proximal to the EDJ takes on its pyramid-like appearance, becoming a true Tomes' process (Skobe, 1976; Boyde, 1997; Berkovitz et al., 2002; Jiang et al., 2014). The Tomes' process possesses a set of prongs which sit on a single side of the pyramid. These prongs secrete the initial enamel matrix which form the walls of interprismatic regions (e.g. Skobe, 1976; Boyde, 1997; Berkovitz et al., 2002; Antoine and Hillson, 2016; Antoine et al., 2018). By creating these walls, the prongs create hexagonal pits which allow the Tomes' process to secrete enamel matrix from its

apex. This matrix fills the linear pits formed by the walls as the ameloblasts move away from the EDJ, thereby forming enamel prisms (Boyde, 1989; Berkovitz et al., 2002). The walls defining the shape of each prism also creates space, known as discontinuities, between different prisms. This means that the spaces between each prism possess different structural compositions and thereby different refractive indexes, meaning individual enamel prisms to be identified under light microscopy (Antoine and Hillson, 2016; Antoine et al., 2018).

During their movement away from the dentine and the EDJ, the ameloblasts are held in their linear structure by the cellular web formed during the pre-secretory stage (Berkovitz et al., 2002). This web is attached to the end distal to the EDJ. The movement of the cells away from the EDJ during this stage is highly regular, determined by a daily rhythm of matrix secretion (Robinson et al., 2003; Zheng et al., 2013). It is this rhythmic movement that produces the incremental layers which compromise enamel and produce the cross striation and Retzius lines described earlier in this chapter. After it is secreted, but before it matrix is mineralised, the enamel matrix is soft (Boyde, 1997). Soon after this initial secretion however, mineralisation of the enamel matrix begins, and enamel becomes tough as required by its masticatory function (Boyde, 1997; Berkovitz et al., 2002; Robinson et al., 2003).

2.2.3 Transition stage

The transition stage begins as enamel reaches its full thickness and is the shortest of all stages of amelogenesis (Berkovitz et al., 2002). During this stage ameloblasts undergo morphological changes including the loss of height and volume, the destruction of their Tomes' process, and a significant decrease in the organelle content (see Fig 2.5c) (Skobe, 1976; Boyde, 1989; 1997; Berkovitz et al., 2002). The Golgi apparatus and RER see especially large reductions, as the surviving cells undergo autophagocytosis (destruction via self-digestion) (Berkovitz et al., 2002). Such changes to ameloblasts are necessary in order for the maturation of enamel to occur. At the end of the transition stage around 25% of the ameloblasts undergo apoptosis (programmed cell death) (Berkovitz et al., 2002; Nanci, 2003).

Whilst fully formed in terms of size and morphology, at this stage the enamel of the tooth is still immature with high levels of water and protein, alongside a relatively low mineral content (Berkovitz et al., 2002; Nanci, 2003). During the transition stage enamel can possess as high as 65% water content, 20% other organic material, and as low as 15% hydroxyapatite crystals by weight (Berkovitz et al., 2002; Nanci, 2003). The transition stage ends as the surviving cells, now positioned

at the outer surface of the tooth surrounding the immature enamel, form a basal lamina which attaches the cells to the enamel via hemidesmosomes (stud-like connection structures) (Berkovitz et al., 2002).

2.2.4 Maturation stage

During the maturation stage immature enamel is mineralised into its final mature state. This involves the removal of water and amelogenin-rich surrounding matrix by ameloblasts, and the subsequent expansion of pre-existing enamel crystals to reduce the inter-crystalline space (Rönnholm, 1962; Boyde; 1998; Beynon et al., 1998; Berkovitz et al., 2002; Nanci, 2003; Smith and Nanci, 2003; Antoine and Hillson, 2016; Antoine et al., 2018).

The maturation of enamel is the direct result of the actions of the ameloblasts. At the start of this process approximately 25% more ameloblasts undergo apoptosis leaving only 50% of the original cells (Berkovitz et al., 2002). Those surviving ameloblasts undergo more morphological changes as the remaining organelles move and collate at the end of the cell adjacent to the Tomes' process (Skobe, 1976; Boyde, 1997; Berkovitz et al., 2002). The Tomes' process itself becomes a striated and ruffled border which allows the ameloblasts to actively control the release of accumulated calcium and phosphate ions into the enamel through active transport calcium pumps (see Fig 2.5c) (Wöltgens et al., 1995; Simmer et al., 2014). This release causes the final transition from immature, highly porous enamel, to mature, highly mineralised enamel as the calcium moves in an occluso-cervical direction (Beynon et al., 1998). During this process hydroxyapatite crystals enlarge, causing the crystallites to change from an initial size of 1.5nm thick up to 25nm in thickness (Wöltgens, 1971; Boyde, 1997; Berkovitz et al., 2002; Antoine et al., 2018). The expanding crystals cause a significant reduction in inter-crystalline space and organic composition (Hiller et al., 1975; Robinson et al., 2003). Once fully mature enamel prisms can never be treated by ameloblasts as these are lost during the post-maturation stage, meaning then cannot undergo further development or repair (see section *2.2.5*). As a result mature enamel prisms are consequently a permanent record of ameloblast secretory history and enamel growth (Dean, 1989; Antoine et al., 2018).

In order to remove excess water, the ameloblasts will transition between ruffled and smooth stages between five to seven times during the maturation stage (Berkovtiz et al., 2002; Wright et al., 2015). The smooth stage produces junctions between the cells, which allow for the escape of water and proteins which are squeezed out by expanding crystallites (Berkovtiz et al., 2002; Wright et al., 2015).

2.2.5 Post-maturation stage

The post-maturation stage begins at the climax of enamel mineralisation where the mineral to organic material ratios are as described at the beginning of this chapter. At this stage the remaining ameloblasts become flattened, losing their columnar shape (see Fig. 2.5d) (Berkovtiz et al., 2002). Here the ameloblasts are separated from the enamel as a 1µm thick layer of amorphous protein forms between them (Berkovtiz et al., 2002; Skobe et al., 2017). Once separate the ameloblasts are joined together by a basal lamina on their ends proximal to the enamel before being absorbed by the surrounding dental follicle around the time of crown eruption (Boyde, 1997; Berkovtiz et al., 2002; Lacruz et al., 2013; Skobe et al., 2017; Antoine et al., 2018). This lamina continues to surround the mature enamel and acts as a protective barrier during the eruption of the tooth, only fully disintegrating when the tooth has erupted from within the oral cavity of the alveolar bone (Boyde, 1997; Berkovitz et al., 2002; Antoine et al., 2018).

CHAPTER 3: LITERATURE REVIEW

LITERATURE REVIEW I – HISTORY OF ENAMEL HISTOLOGY AND VARIATION IN HUMAN ENAMEL GROWTH AND DEVELOPMENT

3.1 Importance of enamel growth and earliest history of enamel histology

Enamel growth of human and non-human primates is plastic with both external and internal structures developing, or developing differently, according to different events and personal life histories which coincide within the periods of enamel formation. Such events include stress (e.g. Guatelli-Steinberg et al., 2012), accessory enamel growth (e.g. Aris and Street, 2021), nutritional deficit (e.g. Wright et al., 2015), and weaning practices (e.g. Dirks et al., 2010). In addition, enamel thickness is a commonly researched area of anthropology, especially within dental- and paleo-anthropology (e.g. Skinner et al., 2015). Comprehension of the timing and pattern of enamel growth can only provide useful context to data on enamel thickness of the same teeth, especially when such data may be used to differentiate groups on a population or even species level. Both the events which can influence enamel growth, and features of enamel thickness are a clear and common area of anthropological research. Understanding the process and variation of enamel crown growth is therefore vital to such research. As a result the study of enamel crown growth has been ongoing for over a century, often through the analysis of incremental lines.

The study of dental incremental lines was first recognised during the mid-19th century (Retzius, 1837; Owen, 1845). Following this, research found dentition developed as the result of biological systems and cellular secretion (Halberg et al., 1959; Needham, 1964; Neville, 1967; Scheving and Pauly, 1974). The earliest examples of enamel histology date from the turn of the century, with studies by von Asper (1916) and Gysi (1931) – they were the first to claim cross striations were an incremental representation of daily growth. Subsequent studies provided further evidence for the incremental growth of dentition (Schour and Pincher, 1937; Okada, 1943; Massler and Schour, 1946). These studies thus showed how enamel histology could be used to investigate enamel daily secretion patterns (Asper, 1916; Gysi, 1931; Schour and Pincher, 1937; Okada, 1943; Massler and Schour, 1946). It is only by the discovery of the incremental growth of enamel, and the development of enamel histology as a method, that the research presented here can be conducted.

3.1.1 Early study of incremental lines

Studies of enamel histology involve the analysis of human and non-human dentition. Mimura (1939) used a system of labelling developing teeth in dogs, rabbits, and pigs by injecting lead acetate and sodium fluoride. This acted to stain the teeth at intervals while they were forming, and were observed as distinct striae within the enamel. It was further noted that the lines which formed between stained intervals corresponded to the number of days between injections (Mimura, 1939), supporting the hypothesis of daily lines forming incrementally during enamel formation. Histological and scanning electron microscopy (SEM) studies published in the mid-20th century, using mammalian and human archaeological material, showed that cross striations can be both viewed and analysed as a daily forming structure (Gustafon, 1959; Boyde, 1963, 1967, 1979; Kajiyama, 1965; Tagiguchi, 1966; Helmcke, 1967). It is the discovery of the human-based studies in particular (Gustafon, 1959; Kajiyama, 1965; Tagiguchi, 1966; Boyde, 1967, 1979), that enamel develops under a daily pattern, that allows the published manuscripts (Chapters 6 and 7) presented later to examine enamel growth rate differences between populations.

In contrast, Weber and Glick (1975) theorised that incremental lines within enamel could be the product of bisecting enamel prisms. However, enamel histology routinely involves the vertical sectioning of teeth which produces dental sections along the plane which prisms form, deviations of enamel prisms can on occasion be viewed on the same slide as cross striations. Warhawsky and Bai (1983) suggested that striation-like markings could be the products of knife chatter when making ultra-thin slides. This claim has since been discredited as cross striations have been observed via both SEM analysis and through thicker ground sections of enamel.

3.1.2 Early human histology

Compared to the fossil and primate research, the frequency of early studies conducting histology on human teeth is low. Early studies utilised small sample sizes (Gustafon, 1959; Boyde, 1963; Shellis, 1984; Risnes, 1986 Dean and Beynon, 1991; Dean et al., 1991; Beynon, 1992; Dean and Scandrett, 1995; Reid et al., 1998), highlighting the low number of available human sections which existed at the time. The material used was mostly archaeological, which shows that the use of clinical dentition is only now becoming more widely used. The first research using histological techniques tested whether those methods pioneered on non-human primates could accurately be replicated on archaeological material (Gustafon, 1959; Boyde, 1963; Shellis, 1984). Subsequent research worked to identify the common growth patterns of human enamel, and made preliminary comparisons to

known patterns of extinct hominin enamel formation rates and enamel thicknesses (Wilson and Beynon, 1989; Dean and Beynon, 1991; Beynon 1992). The identification of common human enamel thickness and growth patterns is of particular use to the enamel research presented later, as it can be used for comparison to the data collected here.

3.2 Modern history of enamel histology

3.2.1 inter-species and population analyses

Since the start of the 21st century, the frequency at which studies have been published on the subject of enamel histology has increased. These newer studies discussed how histological enamel data can be used to estimate age of death and the life history of human and non-human primates (Dean, 1998; Dirks, 1998; Reid et al., 1998; Antoine, 2000; Dean and Reid, 2001; Schwartz et al., 2001; Dirks et al., 2002; Dean and Schrenk, 2003; Reid and Dean, 2006; Smith et al., 2007a; 2007b). In addition, a number of studies created models of enamel characteristics unique to each hominin taxa (Beynon et al., 1998; Reid and Ferrell, 2006; Ramirez-Rozzi, 1998; Dean and Smith, 2009). More recent studies have continued to investigate from a more inter-specific perspective. These studies built upon earlier research (Wilson and Beynon, 1989; Dean and Beynon, 1991; Beynon 1992) by comparing findings of one hominin species to modern humans, rather than making isolating conclusions to a single taxa (Reid and Dean, 2000; 2006; Schwartz et al., 2001; Smith et al., 2007a; 2007b; 2009; 2010; Guatelli-Steinberg et al., 2005; 2007; 2012b; Guatelli-Steinberg and Reid, 2005; Reid et al., 2008; Dean, 2009).

One assumption made in many of these histological studies is that enamel thickness and growth patterns are consistent within hominoid species, allowing for the use of single populations are representative samples of a species. More recent research by Guatelli-Steinberg and colleagues (2012b) compared data gathered from modern human and extinct fossil hominin histological samples. They found a significant cross over between extension rates and crown formation times (CFTs), across both the enamel of multiple tooth types and species (Guatelli-Steinberg et al., 2012b). They concluded that small data sets were unreliable in assessing enamel features of entire species, which had previously been the norm (Shellis, 1984; Guatelli-Steinberg et al., 2012b). Within the literature these is additional appreciation that small samples sizes may not be representative of populations both for human and non-human species (Schwartz et al., 2002; 2003; Smith et al., 2003; 2004; Lacruz, 2006; Lacruz and Bromage, 2006; Mahoney et al., 2007) and caution should be taken in

their interpretation. As a result a study investigating for intraspecific variation in enamel formation between populations within a species is pertinent.

3.2.2 Amelogenesis biomechanics

Recent enamel research focuses on how the mechanisms of amelogenesis can result in enamel growth variation between enamel regions and tooth types. This can be seen in studies by Mahoney (Mahoney, 2008; 2010; 2011; 2012; 2013; 2015; Mahoney et al., 2016; 2017) and Guatelli Steinberg et al (2012b). These most commonly include influencers of prenatal enamel growth (Mahoney, 2008; 2011; 2012; 2015; Mahoney et al., 2017), eruption and cuspal sequencing (Mahoney, 2008; 2012; 2015), and evolutionary morphological adaptations (Smith et al., 2006).

The specific study of deciduous enamel and the comparison of deciduous to permanent enamel development and formation have also become more frequent in recent literature. Research by Grine (2005), Birch (2009; 2012), and Mahoney (2011; 2012; 2013; 2015; Mahoney et al., 2016; 2017) have further developed the field of histology by focussing their discussion on how enamel develops differently according to its region and tooth type, and concluding with how future research should continue to investigate this (Mahoney et al., 2017). Recent research concerning dental and skeletal biorhythms have followed such suggestions by investigating possible causes for the development of cross striations and Retzius lines (Lacruz and Bromage, 2006; Antoine et al., 2009; Bromage et al., 2012; Lacruz et al., 2012; Zheng et al., 2013; Mahoney et al., 2016). These articles collectively have been successful in identifying variations in enamel growth patterns within modern humans, and thereby show the potential of using histological methods to identify and interpret other existing intraspecific variations in human enamel.

3.3 Intraspecific study of human enamel histology

The earliest research published conducting intraspecific analysis of human enamel identified the ranges of human enamel periodicity (Weber and Glick, 1975; Boyde, 1979; Weber and Ashrafi, 1979; Risnes, 1986; Beynon and Reid, 1987; Sunderland et al., 1987; Beynon, 1992; Dean et al., 1993; Fitzgerald, 1998; Reid et al, 1998a; Risnes, 1998; Li and Risnes, 2004), age estimation (Boyde, 1963; Moorrees et al., 1963; Christensen and Kraus, 1965; Kajiyama, 1965; Gustafon and Koch, 1974; Huda and Bowman, 1995; Liversidge, 1995; Fitzgerald, 1998; Reid et al., 1998a; Antoine, 2000; Fitzgerald and Saunders, 2005; Antoine et al., 2009), and crown formation times (Kajiyama, 1965; Tagiguchi,

1966; Dean and Beynon, 1991; Dean et al., 1993; Liversidge, 1995; Fitzgerald, 1995; Beynon et al., 1998). These studies present a reliable pattern of enamel formation for all human tooth types, with enamel secretion rates found to continuously increase from the dental cervix to the occlusal surface of enamel, and with relatively slow CFTs compared to other primates. This research laid the foundations for intraspecific studies of human enamel, however few addressed inter-population variation (Fitzgerald, 1995; Reid and Dean, 2006; 2008; Reid and Ferrell, 2006; Smith et al., 2006), and even fewer temporal variation (Smith et al., 2010; Le Luyer et al., 2014; Le Luyer and Bayle, 2017). It is therefore pertinent that subsequent research be conducted which further examines inter-population differences in human enamel thickness and growth across a temporal transect.

3.3.1 Intraspecific permanent and deciduous patterns

Research by Harris (1998), Grine (Grine et al., 2001; Grine, 2002, 2005), and Gantt and colleagues (2001) present preliminary investigations of variation in enamel thickness of permanent and deciduous teeth. They found that enamel thickness followed a trend of increased thickness moving distally within the maxilla and mandible, with no variation between the sexes. However, the most influential finding was the significant difference between the relative enamel thickness (RET) of deciduous and permanent molars (Gantt, 2001; Grine, 2002, 2005). Subsequent research by De Menezes Oliveira and colleagues (2010) and Mahoney (2010, 2013) provided further evidence for morphological variation, finding mean thickness (De Menzes Oliveira et al., 2010) as well as average enamel thickness (AET) (Mahoney, 2010, 2013) of the deciduous teeth to be significantly reduced compared to that of equivalent permanent teeth. In addition, De Menzes Oliveira and colleagues (2010) investigated the shape of enamel prisms and mineral composition of the same teeth. Their results showed that while deciduous teeth contained a higher percentage of organic material and denser enamel prisms, the morphology of deciduous enamel prisms was identical to that of permanent dentition (De Menezes Oliveira et al., 2010). The combination of the similarities and variations between deciduous and permanent human enamel features show how tooth types can develop according to varying patterns of amelogenesis. It is therefore vital to widen research of human dental growth to multiple tooth types, even with the permanent dental arcade.

In response to the studies on enamel thickness, Birch and Dean (2009, 2014) researched deciduous teeth, comparing growth rates in correlation with thickness of deciduous human teeth to known permanent ranges (see Reid et al., 1998). While their preliminary deciduous sample was compromised of only 20 individuals from an unspecified population(s), they found that deciduous

enamel followed the same general DSR pattern, with volume secretion increasing with distance from the EDJ. In contrast, deciduous enamel DSRs were found to increase at a slower rate from inner to outer regions compared to permanent teeth (Birch and Dean, 2009, 2014). These findings can be replicated through an analysis of data published by Fitzgerald and Hillson (2009), gathered from an ancient Greek juvenile cemetery, when compared to permanent enamel DSRs published by Reid and colleagues (1998).

These conclusions have subsequently been supported in studies by Mahoney (2011, 2012), which provided in depth analyses of formation patterns involved in the development of deciduous mandibular first and second molars (Mahoney, 2011), and mandibular and maxillary canines and first and second incisors (Mahoney, 2012). They found that deciduous DSRs were faster within the inner regions of enamel but slower in the outer regions when compared to permanent dentition. In addition, the DSRs of all deciduous enamel regions were found to remain consistent within the tooth row, similarly to what has been observed in permanent dentition (Beynon et al., 1991b). This showed again how deciduous teeth develop more consistently and do not undergo the exponential increase in DSRs regularly seen in permanent teeth (Mahoney, 2011, 2012). Overall, the consistent findings of variation between deciduous and permanent enamel growth highlight the differences that can exist between different human teeth. It is important therefore to expand this research, including comparisons of DSRs between different permanent tooth types.

3.3.2 Intraspecific patterns of permanent enamel growth and thickness

The first studies to investigate variation within permanent human dentition looked into differences between the teeth of the same function. Macho and Berner (1993) and Macho (1994) found a significant reduction in crown diameters when moving distally along the tooth row. Within the maxilla, first molars had significantly thinner enamel than second and third molars and overlapping ranges in RET (Macho and Berner, 1993; Macho, 1994). Macho (1994) suggested that these findings indicated that a species-specific range of enamel traits that could not be accurately formulated and presented without large scale in-depth interspecific studies.

Grine (2002, 2005) investigated variation between human molar types. They found that RET progressively increased from first to third molars, and for cuspal enamel thickness of molars to be more variable than lateral thickness. However, this was thought to be the result of decreasing dentine volume in more distal molars, than the result of adaptations in enamel (Grine, 2002, 2005). Smith and colleagues (2006) studied the maxillary and mandibular teeth, finding AET to also increase

from between molars moving distally along the tooth rows. Some enamel thickness features also displayed sex differences, with thicker AET in females, and greater dentine area, EDJ length, and bi-cervical diameter in males (Smith et al., 2006).

Mahoney (2008) investigated a wider number of growth pattern traits within the context of specific teeth and found that within first molar Retzius periodicity (RP), DSRs, and CFTs could vary within both a population and a tooth while following the normal trend associated within human enamel development. Dean (2009) and Guatelli-Steinberg and colleagues (2012b) found enamel extension rates (EERs) varied alongside CFT and variation in human EERs both within and between premolars and molars. Guatelli-Steinberg and colleagues (2012b) further showed that EERs declined in modern humans, and were strongly related to EDJ length. These results first provide further evidence for variation existing within humans, and the shape of the tooth may be the cause of variation. While the combination of this research represents an appreciation of different enamel growth patterns existing within humans, these studies focused on specific populations. Future research would benefit from conducting similar research comparing across multiple human populations.

3.3.3 Intraspecific geographic variation between populations

Fitzgerald (1995) first published analysis of different populations, using histological samples of modern human molars. Their statistical analysis revealed significant differences in the growth rates, particularly in the CFTs, of Japanese and African samples (Fitzgerald, 1995). While this research did not identify a pattern to geographic variation, it did show evidence for significant intraspecific variation existing in modern human enamel.

Reid and Dean (2006) and Reid and Ferrell (2006) conducted inter-population research into human enamel growth. They studied 678 samples covering all tooth types, spread across five different populations from four different continents. The CFTs varied sporadically between African and European populations especially, with both presenting significantly higher and lower rates than the other depending on the tooth types analysed (Reid and Dean, 2006; Reid and Ferrell, 2006). Smith and colleagues (2006) conducted a similar study using three of the four histological collections used by Reid and Dean (2006). They presented more supporting evidence for variation in enamel thickness between posterior teeth; they also found further differences in the EDJ length and thickness between the first molars of the American and European populations (Smith et al., 2006). While these studies displayed further evidence for variation in enamel growth patterns between

human populations they do not consider whether the difference in time period between the populations may have influenced inter-population differences in enamel features.

Reid and colleagues (2008) found significant differences in all upper premolars and first lower premolars between the European and African populations. The differences between European and African human CFTs were found to vary according to tooth and cusp, but with a general trend to longer CFTs in European individuals. These results further highlighted the variation between populations in modern human teeth and encourage the further expansion of similar research. These studies presented crucial insight into enamel variation in humans. However, whilst location may be the causation for varying dental development patterns, it must be appreciated that all the above research used the same clinical sections from southern Africa and medieval sections from the Netherlands/Denmark. As a result, the cause of varying enamel formation could lie within the temporal distinctions differentiating the populations, instead of or in addition to their geographic differences.

3.3.4 Temporal studies of modern human enamel

Despite the breadth of intraspecific human studies, the volume of those making direct conclusions from temporal analysis is low. Smith and colleagues (2007a) present the first study directly commenting on temporal variation in modern human enamel. They showed CFTs of modern humans from Europe and South Africa to a 160,000 year old sample (*Homo neanderthalensis)* from Morocco. Their initial analysis found the same sporadic variations between the African and European incisors and molars as Reid and Dean (2006). Moreover, Smith et al (2007a) found the Moroccan individual presented longer CFTs (up to ~800 days) than modern humans. Following this, Smith and colleagues (2010) continued investigating temporal changes in enamel formation, by calculating EERs of a modern human pre-historic population and of fossil human deciduous teeth. While the fossil sample was small, the EERs they presented were near evenly split between falling within, and faster than, the ranges calculated from the modern humans (Smith et al., 2010). These analyses did not involve sufficient pre-history human specimens to make any final assessment as to how much variation exists within human enamel formation patterns. However, they do provide the first evidence of significant temporal variation in human enamel growth features.

Le Luyer and colleagues (2014) and Le Luyer and Bayle (2017) investigated temporal variation in modern human enamel thickness. Le Luyer and colleagues (2014) research focussed on upper molars from populations of Upper Palaeolithic, Mesolithic, and Neolithic periods, and involved

comparing the RET and a series of linear thickness measures between these populations. Using histological evidence and micro-scans of the molars they found a significant reduction in the RET and metacone expression of the Mesolithic molars compared to the Neolithic dentition (Le Luyer et al., 2014). Le Luyer and Bayle (2017) further concluded that the transition from hunter-gatherers to agriculture resulted in the significant changes seen in enamel morphology over the short time span. This research further highlighted how human enamel is able to rapidly change between populations in a temporal context.

Overall, earlier research provided strong evidence for both human enamel growth rates and thickness being highly plastic, and able to respond to evolutionary pressure over a short period of time. This research is still in its infancy, and more research is required to further expand our understanding of temporal variation in human enamel.

3.4 Concluding remarks I

To conclude, the literature regarding histological analyses of human dentition shows enamel to be highly researched. Our understanding of how human enamel thickness can vary according to tooth types, and how enamel growth rates vary between regions of the dental crown and between tooth types, is particularly well studied. Subsequent analyses of human enamel variation regarding enamel thickness and growth rates, have been mostly limited to studies between populations of different geographic location (Fitzgerald, 1995; Reid and Dean, 2006; Reid and Ferrell, 2006; Smith et al., 2006; Reid et al., 2008). Less attention has been given to possible temporal variation and where this has been examined it is limited to the study of enamel thickness (Le Luyer et al., 2014; Le Luyer and Bayle, 2017) and CFT analyses (Smith et al., 2007a). It can be seen that enamel DSRs have not been analysed for variation between populations overtime, or alongside similar analyses of enamel thickness. Our understanding of how these features of enamel development vary between human populations is therefore limited. As a result it is pertinent for research to analyse human populations of similar geographic origin, dating from different time periods, for inter-population variations in enamel growth rate and thickness measures.

LITERATURE REVIEW II – MEASURES OF PERMANENT ENAMEL

3.5 Growth measures

3.5.1 Daily enamel secretion rate (DSR)

Daily secretion rates (μm/day) represent a quantitative aspect of dental development which can be used to assess the speed at which a tooth's enamel formed (e.g. Beynon et al., 1991a; Schwartz et al., 2001; Reid and Dean, 2006; Mahoney, 2008, 2011; Smith, 2008). They are calculated by measuring the length (μm) between a number of (typically six) adjacent daily formed cross striations which develop as enamel matrix is secreted during amelogenesis (see superimposition of Fig 4.1; e.g. Schwartz et al., 2001; Mahoney, 2008).

By measuring the length of cross striations through the length of the enamel before comparing them to the Retizius lines, early research found regular and consistent relation relations between the two (Risnes, 1986; Dean et al., 1993a; Dean and Scandrett, 1996). These findings showed that measuring cross striations could accurately estimate the speed of enamel growth. More recent studies have compared known age at death to direct counts of cross striations, showing beyond reasonable doubt that short period lines are directly associated with a daily rhythm of enamel matrix secretion (Antoine, 2000; Antoine et al., 2009). Research has also investigated the most accurate method for calculating DSRs. When both radiographic and histological methods were compared, Beynon and colleagues (1998a) found histology to be significantly more accurate.

Enamel growth speeds vary during different periods of dental development, therefore DSRs can be highly variable according to when in the developmental cycle of the tooth the enamel was undergoing secretion (e.g. Beynon et al., 1991a; Schwartz et al., 2001; Mahoney, 2008). As a result DSRs are calculated for specific time periods by analysing isolated regions of the tooth (see Fig 3.1). For instance, measuring the average DSR of inner cuspal enamel will indicate the speed at which the tooth was developing during the initial stages of growth, whereas measuring outer cervical enamel DSRs show the speed of the tooth's final period of development (Berkovitz et al., 2002).

To increase accuracy, DSR measures are taken multiple times within any given region of enamel, before calculating a mean of the results to give an average DSR (e.g. Beynon et al., 1991a; Schwartz et al., 2001; Mahoney, 2008). Daily Secretion rates vary greatly within a tooth, with DSRs increasing with distance from the EDJ, and regional cuspal DSRs being faster than lateral DSRs of

equivalent regions (e.g. Reid and Dean, 2006; Smith, 2008; Mahoney, 2011). Daily Secretion rates have been found to remain consistent within an individual's dental arcade (Beynon et al., 1991a) and between the same enamel areas of different cusps within the same tooth (Mahoney, 2008).

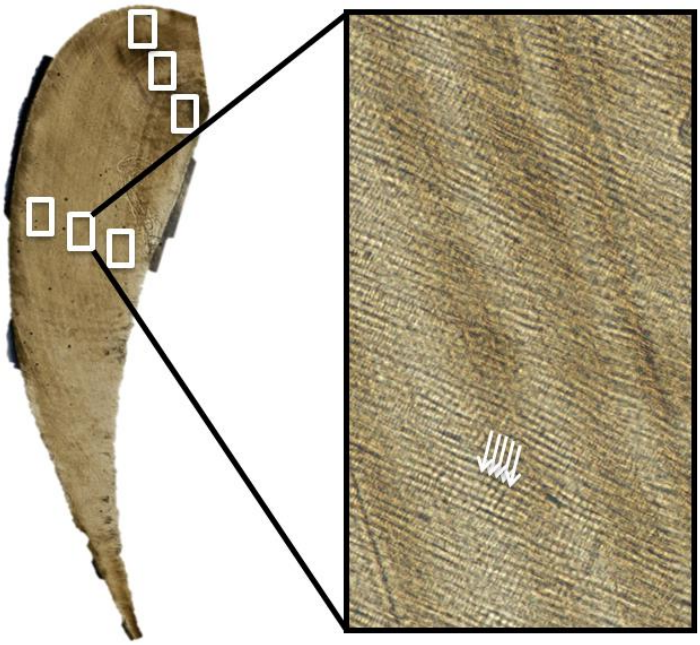

Fig. 3.1. *Digital image of a paracone (the mesial-buccal located cusp) from one individual dating to the Roman period. White squares show the areas of the cuspal and lateral enamel subdivided into inner, mid, and outer regions. The black square shows a 40x magnified superimposition of the mid lateral enamel. White arrows indicate individual cross striations.*

3.6 Thickness measures

Average enamel thickness and relative enamel thickness are a standardised measure of enamel first described by Martin (1983; 1985), and later updated by Shellis and colleagues (1998). While similar, due to the nature of comparing enamel between modern humans, non-human primates and extinct hominins, they are implemented independently according to what is appropriate to the associated study.

3.6.1 Average enamel thickness (AET)

Average enamel thickness (mm) is a quantitative measure which gives an average enamel thickness accounting for all regions (cervical, lateral, and cuspal) of a tooth's enamel (Martin, 1983). Even

though this measurement cannot be used to represent any isolated part of the tooth, it is a useful figure with which to compare the overall thickness of enamel between individuals or populations (e.g. Smith et al., 2006a; Olenjniczak et al., 2008). However, AET is only accurate for use when it is compared between teeth of the same type and position within the dental arcade, and type between individuals of the same species.

Within anthropological literature, AET measures have been presented routinely for unworn permanent dentition, including first molars (Smith et al., 2006a; Olejniczak et al., 2008), canines (Schwartz and Dean, 2005, 2006; Feeney et al., 2010; Buti et al., 2017), and incisors (Reid and Dean, 2006; Smith et al., 2008; Benazzi et al., 2014). In some cases AET has been calculated from worn teeth, but only where enamel wear was minimal or moderate (Smith et al., 2006a). Traditionally a single controlled plane of cross section is used to analyse enamel thickness of human dentition. In molars this is typically the buccal-lingual plane (Schwartz and Dean, 2005; Olejniczak et al., 2005) and in anterior teeth the labial-lingual plane (Reid and Dean, 2006; Smith et al., 2008; Benazzi et al., 2014), both passing through associated cusp and dentine horn apices. In cases where multiple planes are available for thickness analyses, RET is used (see section *3.6.2*) in place of AET to account for the impact of obliquity on enamel thickness calculations (Smith et al., 2004; Smith et al., 2006a). To further account for obliquity of cross sections, AET is only calculated where the associated dentine horn presents as a sharp V-shape (Smith et al., 2006a). These studies provide data from a broad range of populations with drastically different geographic origins and time period dates, as well as providing guidance for using methods for calculating enamel thickness from cross sections. However, despite the range of human populations covered, little research has conducted direct inter-population analyses for enamel AET.

3.6.2 Relative enamel thickness (RET)

Relative enamel thickness is a free scale derivative of AET which accounts for researchers aiming to compare the average enamel thickness across varying tooth types, and dentition of different species (e.g. Martin, 1983; Olenjniczak et al., 2008). Relative enamel thickness is a quantitative figure which represents the overall thickness across the entire area of dental enamel that accommodates for the volume of dentine encapsulated by the enamel crown (Martin, 1983). Relative enamel thickness allows for more accurate comparison of different tooth types and morphologies as it removes the influence of dentine shape and has also been used to reduce the impact of obliquity on enamel thickness calculations where multiple cross section planes are available (Smith et al., 2006a).

The data published for human permanent RET concerns first molars (Smith et al., 2006a; Olejniczak et al., 2008), canines (Schwartz and Dean, 2005, 2006; Saunders et al., 2007; Feeney et al., 2010; But et al., 2017), and incisors (Reid and Dean, 2006; Smith et al., 2008; Benazzi et al., 2014). These provide a significant volume of data and allow for a strong understanding of the ranges existing within human permanent RET. However, no direct analysis for how human RET can vary between populations within the context of changes over time has been conducted.

3.6.3 Linear enamel thicknesses: cuspal (CT) and lateral (LT) thickness

Linear enamel measurements can be taken from two dimensional cross sections of permanent dental crowns. These measures represent the maximum distance that can be measured from the EDJ to the outer enamel surface, with the measured line drawn perpendicular from the EDJ region in question (molars: Macho and Berner, 2005; anterior tooth types: Reid and Dean, 2006). Similarly to AET, linear measures of enamel are only collected from dental cross sections with low obliquity (Smith et al., 2004; Olejniczak et al., 2005), where the dentine horn of associated sections possesses a sharp V-shape (Smith et al., 2006a). Linear enamel thickness measurements are also routinely collected and compared between controlled planes of cross sections, to account for potential differences in measures produced when teeth are sectioned at different angles (Schwartz and Dean, 2005). These are again typically the buccal-lingual plane for molars (Schwartz and Dean, 2005; Olejniczak et al., 2005) and the labial-lingual plane (Reid and Dean, 2006; Smith et al., 2008) in anterior teeth.

As a result of the method for their collection, these features only represent isolated regions of enamel, and are less frequently analysed than AET and RET particularly for anterior tooth types. However, they represent an important measure for showing how enamel volume can vary in its distribution across the enamel cap, where AET and RET only display the overall thickness of enamel which encompasses dentine. Both CT and LT are therefore valuable measures to take in conjunction with analyses of AET and RET.

Linear measures of permanent human first molars are well represented in the literature for both CT (e.g. Schwartz, 2000a; Suwa and Kono, 2005; Reid and Dean, 2006; Mahoney, 2010) and LT (e.g. Macho and Berner, 1993; Suwa and Kono, 2005; Mahoney, 2010) measurements taken from sections across the buccal-lingual plane. Despite the volume of data for human molar linear thickness such measures have yet to be analysed for inter-population variation within the context of changes over time. Anterior tooth type CT has only been published from labial-lingual plane sections

produced from Southern African and European populations and considered for its sexual dimorphism (Reid and Dean, 2006). Anterior tooth type LT has yet to be presented for human populations. As a result, it is imperative to investigate both CT and LT measures for inter-population variation in order to appreciate the level of variation which exists within the human species.

3.7 Concluding remarks II

Both thicknesses and DSRs of human enamel are highly researched. In particular, AET and RET have been researched for variation in molars (Smith et al., 2006a; Olejniczak et al., 2008), canines (Schwartz and Dean, 2005, 2006; Saunders et al., 2007; Feeney et al., 2010; Buti et al., 2017), and incisors (Reid and Dean, 2006; Smith et al., 2008; Benazzi et al., 2014), but these analyses have not investigated for inter-population differences. Linear measures of human enamel thickness are also widely researched for molar teeth (Macho and Berner, 1993; Schwartz, 2000a; Suwa and Kono, 2005; Reid and Dean, 2006; Mahoney, 2010). Anterior tooth linear enamel thickness has been less widely researched, with only CT having been previously studied (Reid and Dean, 2006). In addition to LT having never been published for human populations, CT has never been investigated for inter-population variation. Human daily secretion rates are similarly well researched compared to enamel thickness. While DSR studies have been investigated for variations between regions of the enamel crown (e.g. Reid and Dean, 2006; Smith, 2008; Mahoney, 2011), across individual dental arcades (Beynon et al., 1991a), and between the cusps of individual teeth (Mahoney, 2008), no study has yet investigated for inter-population differences. It is therefore pertinent to examine both enamel thickness measures, and regional DSRs, for multiple tooth types, for differences between human populations. Such analysis will also allow for comments to be as to the correlations, or lack thereof, between the inter-population difference of enamel growth and thickness.

CHAPTER 4: MATERIALS AND METHODS

4.1 Materials – dental samples

Two hundred and eighty five teeth over five different time periods: Roman (70-400AD), Early Anglo-Saxon (500-600AD), Late Anglo-Saxon (800-1200AD), Medieval (1000-1600), and modern-day clinical material (extracted within the last 20 years) (Table 4.1). All archaeological samples came from British populations and modern-day samples compromised teeth extracted from England and Southern Scotland. Unworn teeth were selected where possible. When worn, only teeth with approximately 80% of their crown height remaining were selected based on criteria outlined by Guatelli-Steinberg and colleagues (2005), and when wear was present no data relating to the cuspal region of the enamel cap was collected (see Fig. 4.1). One tooth was taken from each individual during the sampling process, following the guidelines for destructive sampling of human remains guideline outlined by Mays and colleagues (2013) and on request from the institutions which curated the dental material utilised. Left teeth were utilised wherever possible, with the right tooth only being used in instances where the left was missing, poorly persevered, or heavily damaged. Selection preference was also given to individuals presenting an antimere to the tooth being selected for sectioning, but in a select few cases of the Roman collections this condition could not be met in order to attain suitable sample sizes for tooth types within each population.

Fig. 4.1. *Cross sections displaying examples of worn and unworn teeth analysed. The left cross section, a Medieval upper first incisor, displays no occlusal wear. The right cross section, a Roman upper first incisor, displays occlusal wear and thus no data requiring the cuspal region was collected from it.*

Ethnicity was unknown for all individuals across all populations from which samples were taken. In the case of the archaeological collections this was due to individual records not existing for any of the individuals of any of the populations studied. For the modern-day individuals, due to data laws the only information available was the biological sex and town/city location from which the individual had the tooth sampled extracted. Factoring in ethnicity to any analysis was thereby impossible, an unavoidable limitation of this project.

Table 4.1. *Descriptive information of dental samples.*

Population	Date	Location	Collection Names	Number of teeth sampled		
				Upper incisors	Upper Canines	First Molars
Roman	70-400AD	Cirencester, Gloucester	St James' Place/ Bath Gate	10	11	11
Early Anglo-Saxon	500-600AD	Ramsgate, Kent	Ozengell	22	20	20
Late Anglo-Saxon	800-1200AD	Newcastle-Upon-Tyne, Northumberland	Black Gate	10	10	21
Medieval	1100-1500AD	Canterbury, Kent	St Gregory's Priory	19	8	43
	1000-1600AD	York, North Yorkshire	Fishergate House	8	8	5
Modern-day	Extracted within the last 20 years	Newcastle-Upon-Tyne and Glasgow	UCL/Kent	12	12	15

4.1.1 Roman samples

The Roman samples were from two sites: St James' Place and Bath Gate cemeteries (see Fig 4.2). Both sites dated between 70-400AD (see Table 4.1), presented archaeological material consistent with Roman-British populations, and are thought to have been small urban populations with access to marine resources from the River Severn (McWhirr et al., 1982).

4.1.2 Early Anglo-Saxon samples

The Early Anglo-Saxon samples came from a site in Thanet, Ozengell (see Fig 4.2), dated to 500-600AD (see Table 4.1). The population is thought to have been small and coastal, with regular access to marine resources from the North Sea and/or the English Channel (Millard et at al., 1969). The Anglo-Saxon period in Thanet is associated with newly developing urban areas following Roman occupation (McKinley et al., 2015).

4.1.3 Late Anglo-Saxon samples

The late Anglo-Saxon sample came from the Black Gate Cemetery site (see Fig 4.2), dated to 800-1200AD (see Table 4.1). This was a large urban population with access to marine resources through proximity to the River Tyne (Swales, 2012).

4.1.4 Medieval samples

The Medieval samples come from two sites: St Gregory's Priory, Canterbury, and Fishergate House, York (see Fig 4.2). The sites were dated between 1100-1500AD (Hicks and Hicks, 2001) and 1000-1600AD (Holst, 2001) respectively (see Table 4.1). Both are documented to have been large urban populations (Hicks and Hicks, 2001; Holst, 2001).

4.1.5 Modern-day samples

The modern-day samples came from the UCL/Kent collection, a repository of teeth collected from dental surgeries in northern England and southern Scotland (see Fig 4.2). All samples were extracted within the last 20 years (see Table 4.1). Ethical approval for histology research on this collection of teeth was obtained from the United Kingdom National Health Service research ethics committee (REC reference: 16/SC/0166; project ID: 203541).

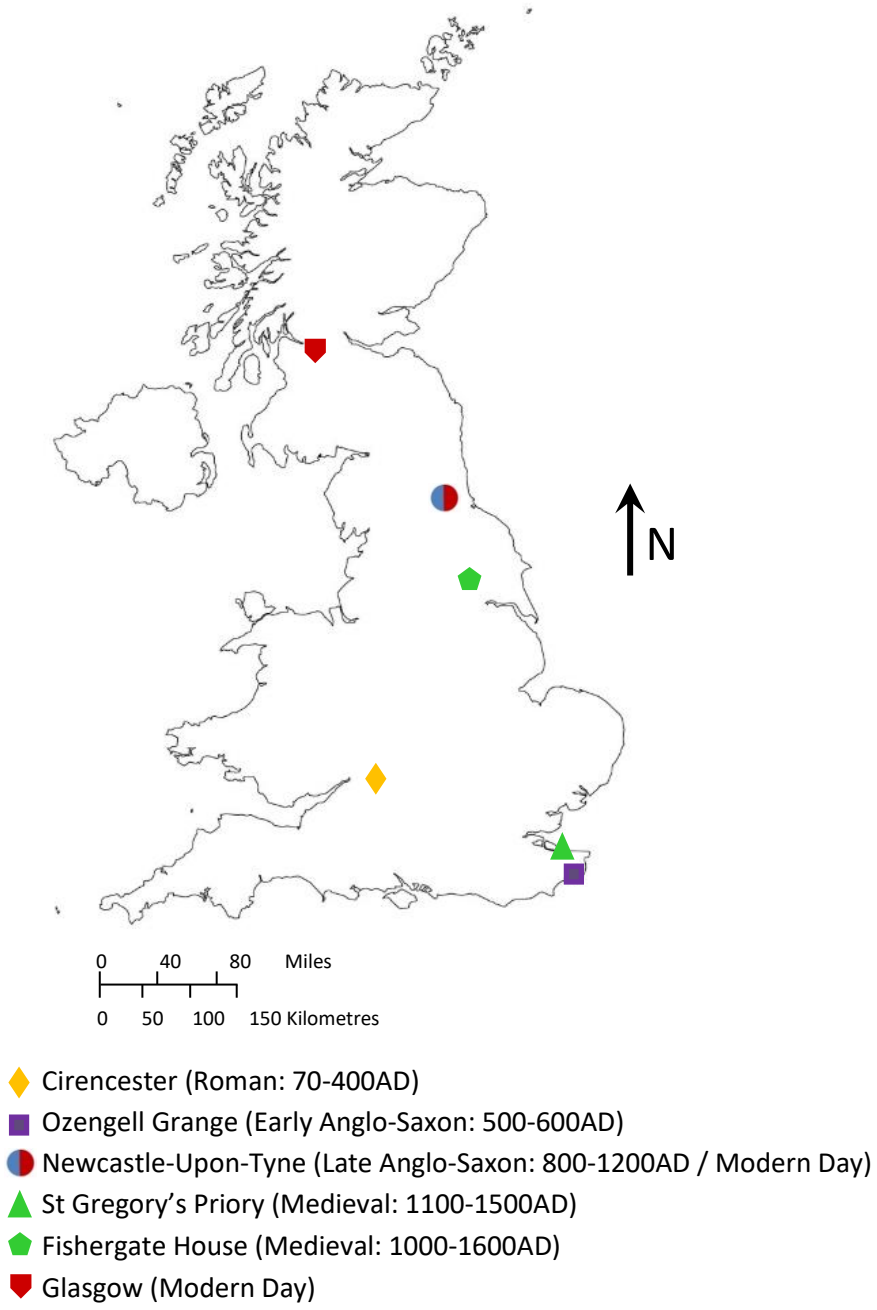

Fig. 4.2. *Map of the United-Kingdom and Northern Ireland displaying the geographic location where the archaeological samples were excavated/modern-day samples were extracted. Shapes denote the samples geographic origin, colour the time period they associated with (multi coloured shapes thereby meaning individuals from more than one time period originated from the same location): Roman = Orange, Early Anglo-Saxon = Purple, Late Anglo-Saxon = Blue, Medieval = Green, Modern-day = Red.*

4.2 Methods – sample preparation

4.2.1 Epoxy resin embedding

Methods for sampling human biological material using semithin histological techniques (Cathey, 1963; Dancey et al., 1976; Islam and Frischm, 1985) were used to produce all dental cross sections according to standard procedures used in dental anthropology research (e.g. Aris, 2020; Mahoney, 2008; Schwartz et al., 2003; Schwartz, 2000). On each tooth the cusp apex, cervical line, and root apex was marked with a fine point marker as to draw a straight line through cusp apex and dentine horn (see Fig. 4.3). For the first molars this line was along the buccal-lingual plane as to pass through: the paracone and protocone of the upper molars; the metaconid and protoconod for the lower molars. For the upper first incisors and canines the line was along the labial-lingual plane. On teeth with curved or broken roots no mark was made on the root apex. A 30mm sample cup (SamplKup) was lined with an epoxy mould releasing agent twice, leaving 30 seconds between each application. The tooth was then embedded in an epoxy resin mixture, with a 4:1 ratio mixture of epoxy resin (Epoxycure 1) and epoxy hardener (Epoxycure 2), for 24 hours. The resin mixture was produced in 20ml disposable scintillation vials (Beuhler). Once lined with a releasing agent the marked tooth was placed within a sample cup, ensuring that the apex of the tooth's enamel surface was in contact with the sample cup's edges, while the root and dental cervix was near to the centre of the cup. Positioning the tooth is this way meant that the first cut into the tooth itself could be aligned as required with the saw starting as near to the cusp as possible to reduce chance of making a misaligned cut (see section *4.2.2*). Once positioned the resin mixture was then poured onto the tooth ensuring it was fully submerged.

Fig. 4.3. *Diagram of how marks were made on upper first incisors, upper canines, and first molars (left to right respectively) before cutting to create a line through the cusp apex and dentine horn. The dashed red line displays the line created by the marks made at the blue crosses. Note the lack of blue cross on the unaligned root apex of the upper canine. The teeth displayed all came from the Fishergate House Medieval collection.*

4.2.2 Cross sectioning

Once the resin had hardened, the embedded tooth was removed from the sample cup. The resin mould was then reduced in size so the tooth could be sectioned immediately adjacent and parallel with the applied markings. Once the mould was reduced the tooth was positioned against a lubricated wafering blade, with the blade aligned with the guiding marks applied previously. No cuts were made into the tooth until it was certain that the blade and marks were aligned. Precise alignment produces a cross section which passes directly adjacent to a tooth's cusp and dentine horn, thus producing a cross section showing the length of the enamel spanning the dentine horn and enamel cusp apices. Whether this was successful can be seen in the shape of the dentine horn of a cross section, where a sharp point (with a V-shaped appearance; Smith et al., 2006a) at the apex denotes and precise cut, or a curved/rounded apex a misaligned oblique cut (Reid and Guatelli-Steinberg, 2017). Where oblique cuts were noted, and thus the precise distance between the dentine horn and cusp apexes could not be accurately measured, the associated sections were not used for analysis of enamel thickness. See Figure 4.4. To reduce the number of teeth not used because of misaligned oblique cuts, the entire method outlined here was practiced on disarticulated

archaeological teeth with no known provenance, provided by the University of Kent Human Osteology Laboratory. Only when suitable cuts could be made reliably were the samples analysed here prepared. Moreover, teeth from the population with larger sample sizes (Medieval and Early Anglo-Saxon) were sectioned first, ensuring that the population with the smallest sample sizes (Roman and modern-day) were sectioned when the most time had been the methodological skill was most refined.

A precision saw (Beuhler IsoMet 4000) and 152-178mm diamond-edge wafering blades (IsoMet) were used to section the tooth in two. To cut the tooth, the blade was spun between 100 to 300 rotations per minute. Slower speeds were maintained when cutting dental material to avoid causing the enamel to shatter. Particular caution was taken when sectioning the more fragile and less well preserved teeth, such as those from the early Anglo-Saxon Ozengell collection.

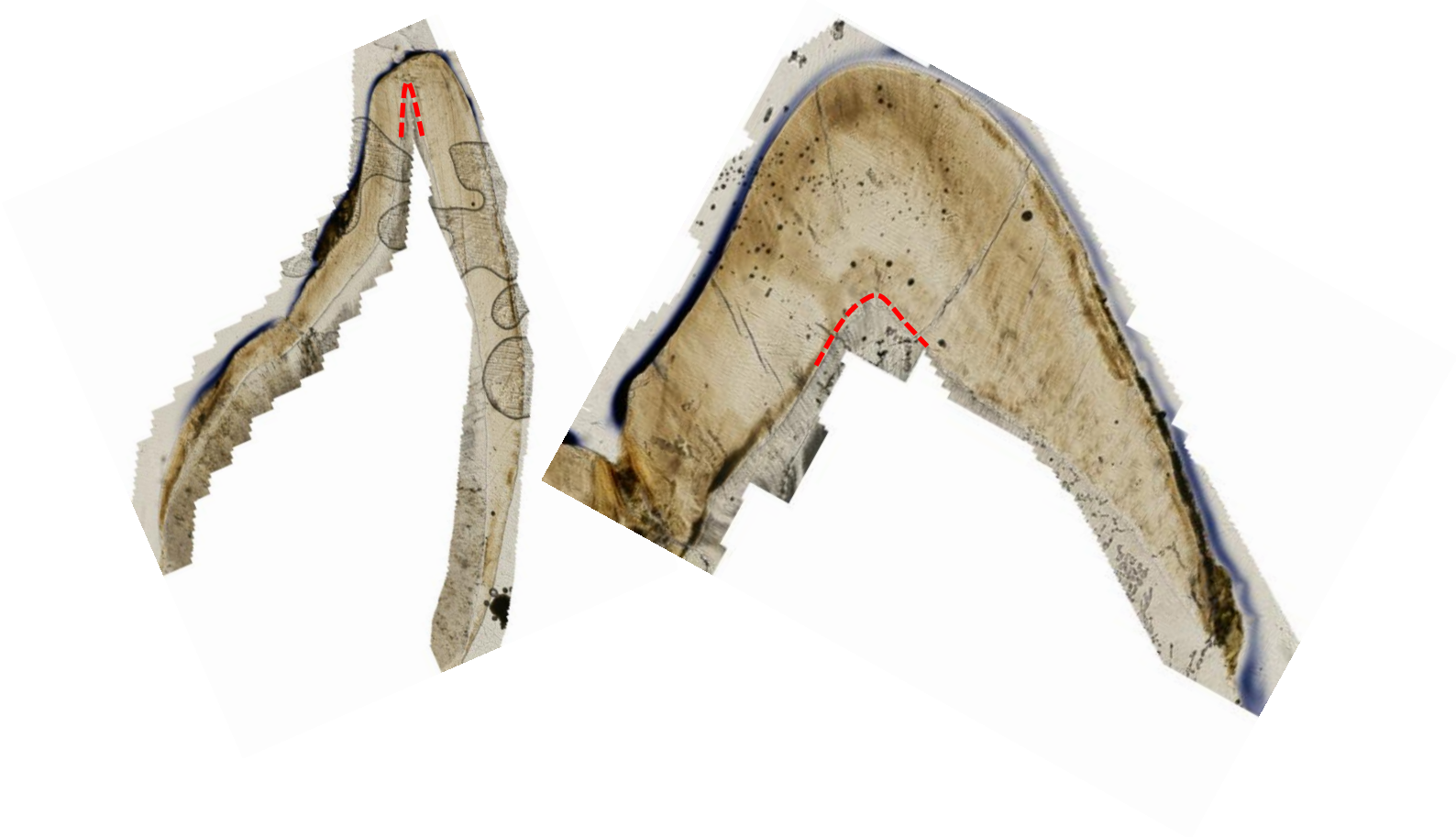

Fig. 4.4. *Cross sections displaying aligned and misaligned cuts, both observed through the shape of the dentine horn (highlighted with dashed red lines). The left cross section, a Medieval upper first incisor, displays an aligned cut with sharp pointed, V-shaped, dentine horn apex. The right cross section, a Roman first molar, displays a misaligned cut with a rounded dentine horn.*

4.2.3 Initial Slide Production

Once cut, the surface of the resin surrounding the sectioned tooth displaying the interior structures was ground down using sheets of 200mm grinding paper (Beuhler SiC). This process removed any resin fragmentation. The tooth was then adhered to standard histological glass slides (Fisherbrand) using the same epoxy resin mixture used to create the resin mould. The adhering resin was left 24 hours to dry. Once dried the mounted cross section was again cut using the wafering blade to reduce the cross section to around 1-2mm in thickness. Once cut, the cross section was cleaned of any remaining macroscopic debris using distilled water.

4.2.4 Grinding and polishing

To grind down the cross section it was placed in a handheld grinder (Behuler). The slide was then lapped against waterproof silicon carbide paper sheet (CarbiMet) of varying degrees of grit at a rate of 50 to 80 rotations per minute using a grinder-polisher (EcoMet 300). The first stage of grinding used a 300mm 600 grit waterproof silicon carbide paper sheet, moving then to 600 grit, and finally 1200 grit paper sheets (CarbiMet). Between using each paper sheet the slide was washed with distilled water and dried, before being analysed under a light microscope (Olympus BX53 Upright Microscope) to assess the clarity at which the enamel structures could be observed. Grinding was complete when slides were between 100-120μm thick (as recommended by: Antoine et al., 2018; Aris, 2020), when enamel areas were a moderate light-brown colour (as seen, for example, in Fig. 4.4), and any striae of Retzius were clear and defined under a 4x magnification.

The slide was then polished to remove scratches created through grinding. The slide was polished using the handheld grinder and a polishing plate applied with aluminium oxide powder (Buehler Micro-Polish II: 0.3μm). The plate was first saturated with water and then aluminium oxide powder was scattered evenly over its surface. The polishing plate was rotated, without further lubrication, at a rate of 80 rotations per minute. Polishing was complete when no scratches were observable on enamel areas when observed under a 20x magnification.

4.2.5 Cleaning and cover slipping

The slide was cleaned in order to remove any particulates, micro- and macroscopic debris, and water. First the slide was placed in a 50ml beaker containing distilled water. The beaker was then vibrated in an ultrasonic bath (Fisherbrand FB11002) for two minutes. While vibrating the polished surface of the tooth was sprayed with distilled water every 30 seconds. The slide was then dried under light. Drying was conducted as quickly as possible to avoid any air particulates accumulating on the surface of dental material.

Once dry the slide was submerged in a 95% ethanol solution for two minutes. At the end of the two-minute period the slide was dried under light. The slide was then submerged for two minutes in 100% ethanol. The slide was then again dried with heat. The slide was then twice dipped in a histological cleaning agent solution (Histoclear) and left to dry for two minutes.

Once dried the slide was given a 22x40mm cover glass slip (Fisherbrand). This was adhered to the slide using a xylene-based mounting medium (DPX). The DPX medium was spread over the

cross section surface of the dental material in order that it covered both the tooth and surrounding slide surface. The cover slip was applied quickly to avoid creating air bubbles between the glass and the cross section. The cover glass slip was allowed to dry for 24 hours. While drying, weights were placed to surround the slide in order to maintain a flat surface area.

4.2.6 Health and safety

Risk assessment forms were completed, and approved by the University of Kent, School of Anthropology and Conservation, ethics review board. Gloves were worn during all stages of sample preparation both for personal protection, and to prevent sample contamination. Particular care was taken during any stage involving the use of releasing agents as well as during the mixing and application of epoxy resins. In addition, when polishing the slides using the histoclear solution and when mounting cover glass slips using DPX medium, both gloves and a face filter mask were worn. Furthermore, at no point was the saw or grinder-polisher left unattended whilst active, and both machines were disassembled whenever they were out of use. Moreover, no chemical component was ever left unattended or exposed to the environment for any longer than necessary, regardless of its hazard level.

4.2.7 Preparation time

The time taken to produce the 285 dental sections is worth noting. Due to the precision required for the marking, cutting, and grinding/polishing stages, and the additional time needed for the embedding and cover slipping processes, the production of a histological slide took four days to complete. Within these four days, outside the time needed for drying, approximately two concentrated work hours were needed to attend each individual tooth over the entire sectioning process. Accounting for five teeth being prepared in groups approximately 72 days and 570 concentrated work hours, over a three month period, was required to produce the entire histological sample examined throughout this project. The time spent conducting histological methods as a part of this project was longer than this, owing to time taken to practice the sampling procedure, and some later samples produced not meeting the required quality not being included among the 285 teeth analysed throughout this project.

4.3 Measurements taken

4.3.1 Daily secretion rates

Daily secretion rates (μm/day) were calculated for the inner, middle, and outer regions of the cuspal and lateral areas of each tooth. These regions were determined by dividing the length of the associated enamel area into three equal-sized parts along the length of local enamel prisms. Standardised methods were utilised to calculate DSRs for the six enamel regions (see Fig 4.5; e.g. Beynon et al., 1991a; Schwartz et al., 2001; Mahoney, 2008; 2011; Bromage et al., 2012). First, the outlined enamel regions were determined by dividing the length of the associated enamel area along the longitudinal aspect of the enamel prisms, into three equal parts. Where the tooth was unworn the cuspal enamel regions were determined within the appositional enamel of each tooth. Lateral enamel regions were determined within the imbricational enamel at a point equidistant between the dentine horn and the dental cervix measured along the EDJ.

Within each enamel region an initial measurement was made along the long axis of an enamel prism corresponding to the length of five consecutive cross striations. This length was divided by five to give a single mean daily rate of secretion. This measurement and mean calculation was repeated five additional times, and the sum of the six means further averaged in order to give a grand mean and standard deviation. Both these procedures were repeated for all three determined regions of both cuspal and lateral enamel (Fig 4.5 superimposition). All measurements of cross striations were taken between 20x and 40x magnification using either single captured images or stitched composite images of whole enamel regions using Olympus cellSens software. Within the Olympus cellsens software magnification-calibrated linear measuring tools were used to take all measurements. Calibrations were checked each new time the software was utilised.

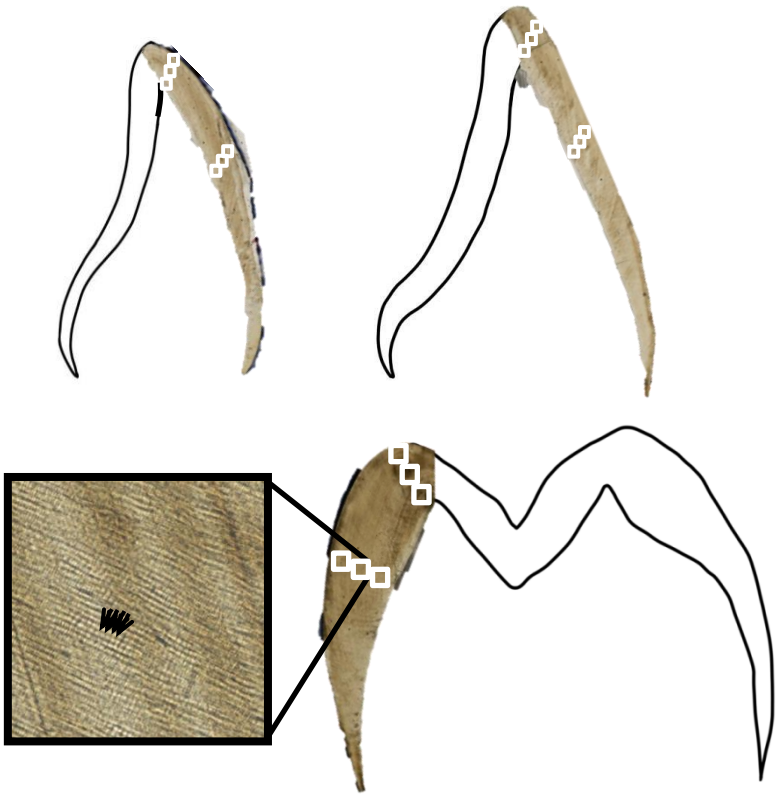

Fig. 4.5. *Digital image of an upper canine (top left), upper first incisor (top right) and a first molar paracone from individuals dating to the Roman period. White squares show the areas of the cuspal and lateral enamel subdivided into inner, mid, and outer regions. These regions were used for DSR calculations. The black square shows a 40x magnified superimposition of the mid lateral enamel. Black arrows indicate individual cross striations.*

4.3.2 Average enamel thickness

Average enamel thickness is calculated by dividing the area of the dental enamel cap, as viewed from a 2D plane after sectioning, by the length of the EDJ. For AET calculations the distance of the EDJ is measured following the path of the junction between the two most distant points of the dental cervix (e.g. Martin, 1983; Smith et al., 2006; Olenjniczak et al., 2008; Feeney et al., 2010). See Figure 4.6. Average enamel thickness was calculated by observing a dental crown under a high powered light microscope at 4x magnification using Olympus cellSens software. All measurements taken to calculate AET were also taken using Olympus cellSens software, using magnification-calibrated measuring tools. Calibrations were checked each new time the software was utilised. For all first molars AET was calculated using the mesial cusps (paracone and protocone).

4.3.3 Relative enamel thickness

Relative enamel thickness is calculated by dividing the AET, as calculated from a 2D plane after sectioning, of a tooth by the square root of the area of dentine. See Figure 4.6. The output of the division is then multiplied by 100 to give the tooth's RET. It is by using the square root of the dentine area that RET becomes a free scale figure. Within the context of this project RET will be used when equivalent teeth of different positions are compared (e.g. UM1s to LM1s). Relative enamel thickness was calculated by observing a dental crown under a high powered light microscope at 4x magnification using Olympus cellSens software All measurements taken to calculate RET were also taken using Olympus cellSens software, using magnification-calibrated measuring tools. Calibrations were checked each new time the software was utilised. For all first molars AET was calculated using the mesial cusps (paracone and protocone).

4.3.4 Cuspal thickness

Cuspal thickness (mm) is a linear measure and was taken from 2D cross sections of each tooth. Measurements were made from the apex of the sharp V-shaped dentine horn to the unworn occlusal surface tip of the enamel (e.g. Gantt, 1977; Beynon and Wood, 1986; Grine and Martin 1988). See Figure 4.6. As this measurement utilises the unworn occlusal surface of a tooth, it was not taken for any teeth presenting any degree of wear. Cuspal thickness was taken from the mesial-buccal cusp for each first molar. Using the mesial-buccal cusp allowed the measurement to be standardised. This was an important protocol to follow as thickness can vary significantly between different first molar cusps (e.g. Macho and Berner, 1993; Macho, 1994; Mahoney, 2010). Cuspal thickness measures were taken under a high powered light microscope at 10x magnification using Olympus cellSens software. Within the Olympus cellsens software magnification-calibrated linear measuring tools were used to take cuspal thickness measurements. Calibrations were checked each new time the software was utilised.

4.3.5 Lateral thickness

Lateral thickness (mm) is a linear measurement which was taken from the buccal-lateral regions of each first molar. For all teeth LT was calculated by measuring the distance between the EDJ and outer enamel surface. The point measured from on the EDJ was located approximately 1mm from the dentine horn, adjacent to the first Retzius line to make contact with enamel surface (See Fig. 4.6;

e.g. Beynon and Wood, 1986; Grine and Martin, 1988; Schwartz, 2000a; Grine, 2005; Mahoney, 2010; Le Luyer et al., 2014). The linear measurement was then made from this point of the EDJ, staying perpendicular to the EDJ, until it reached the outer enamel surface. It should be noted that, as LT does not follow an enamel prism, it is a purely morphological feature not directly reflecting enamel growth. See Figure 4.6. Lateral thickness was measured under a high powered microscope at 10x magnification using Olympus cellSens software. Within the Olympus cellsens software magnification-calibrated linear measuring tools were used to take lateral thickness measurements. Calibrations were checked each new time the software was utilised.

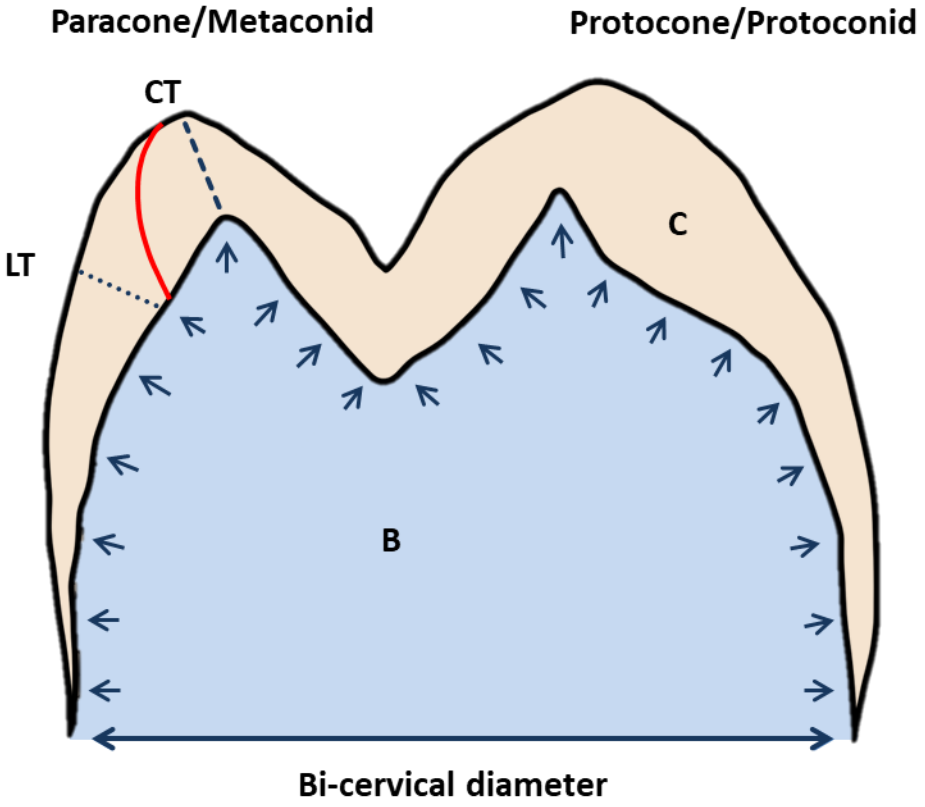

Fig. 4.6. *Cross-sectional diagram of 2D enamel thickness measures taken.* ***C****. the enamel cap area and* ***B****. the dentine encompassed by the enamel and bi-cervical diameter (double-headed arrow). The area of* ***C****. was divided by the length of the EDJ (marked by arrows) to give the average enamel thickness (AET) in mm. The AET is divided by the square root of the area of* ***B*** *and multiplied by 100 to give the relative enamel thickness (RET) (e.g. Martin, 1983), which is a dimensionless index. The first Retzius line that emerges on the outer enamel surface (solid red line) marks the border between cuspal and lateral enamel. The dashed line (CT) illustrates the cuspal enamel thickness measurement (e.g. Beynon and Wood, 1986). The dotted line (LT) illustrates the lateral enamel thickness measurement (e.g. Grine and Martin, 1988).*

4.3.6 Inter-observer error

For each measurement collected, including each of the six regional DSR measures, data collection was repeated a minimum of three times each on a different day. Once all repeats were conducted, if notable differences were seen across the three measures the associated data was deleted and collection restarted for the given measure. This process was repeated where needed until consistent data for each measurement taken was achieved. Note that this meant, for a tooth where all thickness and DSR measures could be collected, an absolute minimum of 126 measurements were taken (108 DSR measurements; Fig. 4.5 and 18 measurements associated with enamel thickness; Fig. 4.6).

CHAPTER 5: RESEARCH CHAPTER – ENAMEL THICKNESS AND GROWTH RATES IN MODERN HUMAN PERMANANENT FIRST MOLARS OVER A 2000 YEAR PERIOD IN BRITAIN

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Co-authored with: Dr Patrick Mahoney, Mackie O'Hara, and Dr Chris Deter

Abstract

Objectives

This study explores variation and trends in first molar enamel thickness and daily enamel secretion rates over a 2000 year period in Britain.

Methods

Permanent first molars (*n*=89) from the Roman, Anglo-Saxon, and Medieval periods, as well as modern-day Britain, were analysed using standard histological methods. Relative enamel thickness (RET) and linear measurements of cuspal and lateral thickness were calculated for mesial cusps. Daily secretion rates (DSRs) were calculated for inner, mid, and outer enamel regions in both cuspal and lateral enamel. Significant differences and trends were identified between samples using non-parametric statistical tests.

Results

Enamel thickness differed between some populations, but no temporal trends were identified. Early Anglo-Saxon first molars had significantly thinner RET than both Late Anglo-Saxon ($p<0.001$) and Medieval ($p<0.001$) first molars. Lateral enamel from the Roman first molars was significantly thinner than the modern-day sample ($p=0.04$). In contrast, a significant slowing trend in DSRs was observed across the more ancient to modern-day samples in every measured region except the mid-lateral enamel region.

Discussion

This study presents the first evidence for a gradual slowing in the daily rate that enamel is secreted in first molars over the past 2000 years in Britain. However, this trend was not matched by consistent or significant positive or negative shifts in enamel thickness. These findings suggest that modern human first molars of similar enamel thickness, from different modern and ancient populations, formed at different rates.

5.1 Introduction

This intraspecific study of modern human permanent molars investigates enamel thickness (e.g. Macho and Berner, 1993; Suwa and Kono, 2005; Reid and Dean, 2006; Smith et al., 2006a) and daily enamel secretion rates (DSRs) (e.g. Beynon et al., 1991b; Lacruz and Bromage, 2006; Mahoney, 2008) in modern and ancient British populations. Enamel thickness and DSRs will be compared across a 2000 year period, between populations from St James' Place and Bath Gate (70-400AD), Ozengell Grange (500-600AD), Black Gate (800-1200AD), St Gregory's Priory (1100-1500AD), Fishergate House (1000-1600AD), and a modern collection of clinical dental extractions. The objective is to identify variation in these two components of dental development when compared between these populations over this period of time.

Some components of enamel thickness, including average enamel thickness and the area of the enamel cap, can vary amongst modern humans when compared between populations, the sexes, and tooth types (e.g., Smith et al., 2006a). Variation in enamel thickness has also been identified amongst modern human populations from different time periods (e.g. Le Luyer et al., 2014; Le Luyer and Bayle, 2017). Thicker enamel has been observed in modern human molar crown regions where surface wear was expected to be greatest (Grine, 2005; Le Luyer et al., 2014; Le Luyer and Bayle, 2017). Subsequent research suggests that a shift in the distribution of enamel thickness upon a molar, when compared between Mesolithic and Neolithic groups from France, may represent a functional adaptation related to the transition to agriculture (Le Luyer and Bayle, 2017). One aim of this study is to further explore this finding in British samples, to determine if molar enamel thickness changed in Britain over the last 2000 years. During this time period there was substantial population movement into and out of Britain, as well as cultural, dietary, and social developments (e.g. Scull, 1993; Privat et al., 2002; Lightfoot et al., 2009). Given the potential association between these factors and enamel thickness, we could expect significant changes between British populations.

Enamel thickness of primate teeth is the product of several growth mechanisms including the daily rate at which ameloblasts secrete enamel (Grine and Martin, 1988; Macho, 1995; Mahoney et al., 2016). Daily secretion rates (DSRs) are typically highly variable when compared within or between primates (e.g. Boyde, 1989; Shellis, 1998; Schwartz et al., 2005; Smith et al., 2007a, 2007b, 2007c), and are typically slower in modern humans relative to earlier fossil hominins (Dean et al., 2001). Several studies have reported mean DSRs for modern humans (e.g. Beynon et al., 1991b; Lacruz and Bromage, 2006; Mahoney, 2008; Smith et al., 2007b). However, while a previous study calculated DSRs for both archaeological and modern-day human sample (e.g. Smith et al., 2007b),

the aim in that study was not focused on variation in DSRs through time. One aim in the present study is to determine if there is a temporal trend in DSRs amongst modern humans from Britain.

5.1.1 Amelogenesis and enamel secretion rates

As DSRs can vary within a tooth, comparisons of these rates between tooth types, or between populations, are usually undertaken for specific regions within a crown (e.g., Dean, 1998) where the molar crown is divided into cuspal, lateral, and cervical enamel, and then further subdivided into inner, mid and outer regions. However, DSRs are broadly similar when equivalent regions are compared between cusps within a molar (Mahoney, 2008).

Most modern human DSR studies use single human populations (Beynon et al., 1991b; Lacruz and Bromage, 2006; Mahoney, 2008). When multiple human populations are examined it is often part of a larger, interspecific study of a grouped single representative sample (e.g. Smith et al., 2007b). Recently, Nava and colleagues (2017) reported slower DSRs in deciduous incisors from the present day compared to equivalent samples from imperial Rome. While their analysis was not focused on a temporal transect, their results do suggest a temporal trend in the DSRs of these populations. No study has specifically compared DSRs from equivalent enamel region of permanent teeth, between modern human populations from different time periods. Thus, our understanding of how DSRs can vary between populations from different time periods is limited.

5.1.2 Enamel thickness

The thickness of enamel is established at the end of the secretory stage of amelogenesis (Nanci and Smith, 1992; Smith and Nanci, 1995). A number of linear measures, and summary measures calculated from them, associated with enamel thickness can be calculated from virtual and histological sections and have featured heavily in anthropological research, often in studies of diet in non-human primates (e.g. Martin, 1983; Molnar and Gantt, 1977; Dumont, 1995; Smith et al., 2007a), extinct hominins (e.g. Kay, 1981; Olejniczak et al., 2008; Skinner et al., 2015; Zanolli et al., 2016) and humans (e.g. Grine, 2005; Reid and Dean, 2006; Feeney et al., 2010; Le Luyer et al., 2014; Le Luyer and Bayle, 2017).

Hlusko and colleagues (2004) reported enamel thickness to be highly heritable in baboons over relatively short periods, in an evolutionary perspective. Examination of the human genome and the genetic components involved in the activation of the enamel matrix protein *enamelin*, have

revealed differences between humans and non-human primates consistent with the selective pressures of natural selection (Daubert et al., 2016; Horvath et al., 2014; Kelley and Swanson, 2008). These interspecific variations in enamelin appear to correlate with known variations in diet between species, strongly supporting the evolutionary link between dietary changes and enamel thickness (Kelley and Swanson, 2008; Horvath et al., 2014).

Within humans, single-nucleotide polymorphisms associated with enamel thickness have been identified between individuals of African and non-African ancestry, suggesting enamel thickness can differ significantly between modern human populations (Daubert et al., 2016). Human enamel thickness might therefore, respond to selective pressures and differ among modern human populations across time as well. This study will therefore focus on features on enamel thickness, including linear measures of lateral (e.g. Macho and Berner, 1993; Suwa and Kono, 2005; Mahoney, 2010) and cuspal molar thickness (e.g. Schwartz, 2000a; Suwa and Kono, 2005; Reid and Dean, 2006), and relative enamel thickness (Martin, 1983; Smith et al., 2006a; Olejniczak et al., 2008) of the molar enamel cap (Figure 6.1). The aim is to determine if enamel thickness, and its linear distribution over a molar crown, changes over a 2000 year time period when compared between British populations.

5.1.3 Inter-specific studies of permanent enamel thickness and diet

Inter-specific studies of primates suggest thicker enamel may relate to the increased consumption of abrasive foods (e.g. Martin et al., 2003).Thicker enamel may allow teeth to retain their ability to chew and process foods throughout an individual's lifetime, delaying exposure of dentine and protecting the pulp chamber while the enamel continues to wear (e.g. Molnar and Gantt, 1977; Janis and Fortelius, et al., 1988; Martin et al., 2003; Pampush et al., 2013). Within hominins for example, quartz dust in or on foods has been suggested to serve as an abrasive additive that may select for thicker enamel (Lucas et al., 2013).

Others have proposed a second model explaining how thicker enamel may relate to food hardness in some mammalian species (e.g., Kay, 1981). Dumont (1995) observed significantly greater molar relative enamel thickness (RET) values in species consuming a hard diet compared to a closely related non-durophagous species. Martin, Olejniczak, and Maas (2003) observed a similar relationship in extant primates, noting that primates with thickly enamelled molars regularly consumed hard diets. The relationship between the mechanical properties of food and enamel

thickness has been considered in *Pan troglodytes* and *Pongo pygmaeus* (Vogel et al., 2008). While both species display preferences for ripe fruit, *Pongo* molar enamel is relatively thicker and possesses occlusal crenulations that may facilitate increased fracture resistance in the face of consumption of a harder diet during fallback episodes. Additionally, genetic evidence indicates that enamelin, one of the genes associated with enamel production and thickness, has undergone evolutionary selection since our divergence from chimpanzees, potentially due to shifts in diet (Kelley and Swanson, 2008; Horvath et al., 2014).

Differences in molar enamel thickness have also been attributed to variation in dietary hardness and composition between hominoid species. Kay (1981) found Ramapithecine species to have thicker enamel that may have related to the consumption of hard-rind nuts or seeds. Studies of Neanderthal populations have suggested that thin enamel in the post-canine teeth of European Neanderthal populations, as compared similarly aged human samples, allude to variation in diet between the two groups (Olejniczak et al., 2008) with particular reference to softer meat-based diets in Neanderthals (Smith et al., 2012). This alternative model suggests that consuming softer foods relaxes selective pressures leading to thinner enamel between species. Skinner and colleagues (2015) analysed a wide range of hominins, finding enamel thickness to follow a trend towards thickening throughout the Pliocene, culminating in the thick enamelled robust *Australopithecus*, followed by a thinning trend throughout the genus *Homo* (Skinner et al., 2015). Evans and colleagues (2016) have further suggested the variation in enamel thickness between hominin species to be result of developmental inhibitory cascade mechanism, wherein decreases in mesenchymal activation and inhibition maintenance within dentition resulted in progressively thinning teeth in *Homo*. It is therefore plausible, as enamel thickness has decreased over time within the *Homo* genus, that a similar thinning of molar enamel to continue in modern human populations. However, while this developmental feature could potentially further explain inter-species hominin variation, it may not necessarily predict patterns within a species over a period as short as 2000 years.

5.1.4 Studies of enamel thickness within human permanent molars

Enamel thickness can vary between modern human populations. Cuspal enamel thickness varies between South African, North American, and European populations samples (Reid and Dean, 2006). Mean average enamel thickness (AET) of molars differed between Southern African, Northern English, Northern American, and medieval Danish samples (Smith et al., 2006a). More recently, Le Luyer and Bayle (2017) reported AET and RET for 40 maxillary second molars from individuals in

France representing a 7000 year period between the Upper Paleolithic, Mesolithic, and Neolithic periods. They found a significant difference between the RET of the Early Mesolithic and Early Neolithic molars, with Mesolithic dentition possessing the thinnest enamel. Within a crown, enamel thickness can also vary over the cusps (e.g., Kono and Suwa, 2000; Kono et al., 2002; Kono, 2004; Mahoney, 2010).

Molar enamel thickness may differ between males and females within populations. Macho and Berner (1993) found linear measures of maxillary third molars from females to have marginally thicker enamel than their male counterparts. Schwartz and Dean (2005) reported that female mandibular third molars had slightly thicker enamel (RET) on average compared to males. Others (Smith et al, 2006a; Saunders et al., 2007; Sorenti et al., 2019) have similarly found thicker RET in female teeth and greater dentine volumes in male molars. Several studies have related increases in enamel thickness along the molar row to increased bite forces along the molar row during chewing, and a change in the proportions of enamel or dentine within a molar (e.g. Macho and Berner, 1993; Schwartz, 2000; Grine, 2005; Mahoney, 2013).

All these findings highlight the actual and potential variability in human molar enamel thickness. They thereby justify conducting wider inter-population studies of modern human enamel thickness. In this particular study, it is therefore reasonable to expect to see significant variation in enamel thickness measures between the populations spanning the last 2000 years in Britain.

5.1.5 Relationship between enamel thickness and underlying growth mechanisms

The relationship between DSRs and enamel thickness within humans is poorly understood. Past research has alluded to links between DSRs and enamel thickness of equivalent teeth in modern humans (Smith et al., 2006a; Mahoney, 2015). When compared along the deciduous tooth row, incisors with thinner enamel had faster mean DSRs compared to molars with thicker enamel that had slower DSRs (Mahoney, 2015). However, permanent first molar cusps with thicker enamel do not necessarily present faster DSRs than first molar cusps with thinner enamel (Mahoney, 2008). The possible association between thicker enamel and faster or slower secretion rates requires further work in humans. Smith and colleagues (2006a) also presented molar DSRs alongside thickness data. However, it was not an aim of their study to explore relationships between this measure of enamel growth and thickness. Thus, whether DSRs vary alongside molar enamel thickness when compared between human populations from different time periods has not been examined previously.

5.2 Materials and methods

5.2.1 Dental sample

Permanent first molars (*n* = 89) were selected from the British populations representing five time periods. These populations date from the Roman period (*n* = 11), Early Anglo Saxon (*n* = 12), Late Anglo-Saxon (*n* = 19), Medieval (*n* = 32) and Modern-day (*n* = 15). Only non-pathological teeth and unworn first molars were selected. Right first molars were selected unless it was unavailable or the left was better preserved. Sex was not known for the majority of these samples as many of the first molars were separate from the rest of the skeleton. For details pertaining to the specific breakdown of each population sample by tooth position, the curated state of samples and the information regarding sex, see Table 5.1.

5.2.2 Enamel thickness

For each first molar, relative enamel thickness (RET), cuspal thickness (CT), and lateral thickness (LT) were measured. Each was measured and calculated on a composite image produced by stitching together 20x magnified images using cellSens digital software. The two linear measures of enamel thickness (CT and LT) were measured on the paracone of maxillary molars, and the metaconid of mandibular molars. These cusps experience reduced masticatory loads compared to the protocone and protoconid (Schwartz, 2000a). While cusps experiencing higher masticatory loads and abrasion levels have been found to be more responsive to external selective pressures, particularly in the in the maxillary molars (e.g. hypocone; Kono et al., 2002), the variable wear levels and preservative states (particularly cracks, chips, and chalk damage) of the archaeological population highly influenced the sample sizes for these cusps. The paracone and metaconid were less affected by wear, meaning the sample sizes for each population were more consistent. As a result, the paracone and metaconid was selected to explore temporal variation in enamel thickness.

5.2.3 Daily secretion rates of enamel

Daily secretion rates were calculated for the inner, mid, and outer regions of the lateral and cuspal enamel areas of each tooth using standard methods (e.g. Beynon et al., 1991a; Schwartz et al., 2001; Mahoney, 2008). The regions were determined by dividing the length of the associated enamel area into three equal parts along the length of the enamel prisms. All cross striations were measured at magnifications between 20x and 40x on the paracone and metaconid. See Figure 5.1.

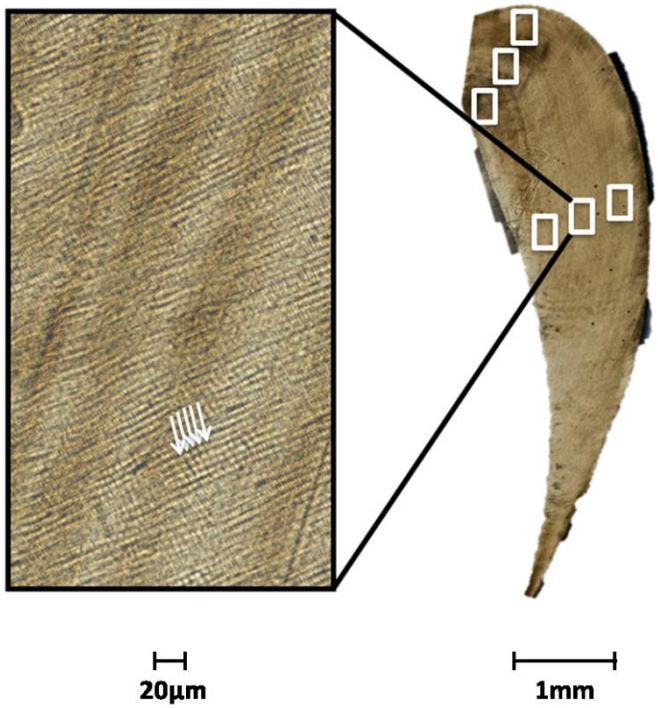

Fig. 5.1. *Digital image of a paracone from one individual dating to the Roman period. White squares show the areas of the cuspal and lateral enamel subdivided into inner, mid, and outer regions. These regions were used for DSR calculations. The black square shows a 40x magnified superimposition of the mid-lateral enamel. White arrows indicate individual cross striations.*

5.2.4 Statistical analysis

Differences in dentin area were tested for between populations using a Kruskal-Wallis test, as a proxy for tooth size. A subsequent Kruskal-Wallis test was then conducted to test for differences in dentine area between maxillary and mandibular teeth. Next, Mann Whitney-U tests were conducted to test for differences in enamel thickness (RET, LT, and CT) and DSRs (from each region), between the maxillary and mandibular M1s within each sample population. Following this, Kruskal-Wallis tests combined with Dunn-Bonferroni pairwise comparison was used to compare enamel thicknesses and DSRs, between archaeological time periods. Kruskal-Wallis tests first identify whether any significant inter-group differences are present, with pairwise comparisons subsequently identifying the exact differences. Jonckheere-Terpestra tests consider changes in means between groups over a defined order (Jonckheere, 1954), so were conducted to identify any significant trends in enamel feature changes with the populations ordered by their time period of origin to test for any trends in changes over time. Positive results from a Jonckheere-Terpstra test indicate increasing

trends, and negative results indicate decreasing trend. All statistical analyses were performed using SPSS 24.0.

5.3 Results

5.3.1 Tooth size and maxillary and mandibular molars

No significant differences were identified in the dentine areas between any two populations for either maxillary or mandibular teeth (Table 5.1). Thus, no scaling of the CT and LT measures was required in subsequent analyses. Dentine area varied significantly between maxillary and mandibular molars (p<0.001; see Table 5.2). This meant RET was analysed rather than AET for subsequent analyses so that the maxillary and mandibular molars could be combined when comparing populations. There were no significant differences between RET or linear measures of enamel thickness, or the DSRs, when compared between the maxillary and mandibular molars of any population (see Table 5.3 and 5.4 respectively). Therefore, the data for maxillary and mandibular molars of each population were combined to form time-period representative samples for subsequent statistical tests.

Table 5.1. *Results of the Kruskal-Wallis test for the comparison of first molar dentine areas between populations for maxillary and mandibular teeth.*

Population	N	Mean	S.D	Min	Max	Kruskal-Wallis Sig.
Maxillary molars						
Roman	4	34.77	5.69	29.27	39.55	0.12
Early Anglo-Saxon	4	27.46	5.74	19.16	32.13	
Late Anglo-Saxon	9	39.89	7.27	31.91	52.02	
Medieval	13	39.78	4.02	32.68	46.59	
Modern Day	2	28.84	3.42	26.42	31.26	
Mandibular molars						
Roman	3	36.17	3.18	32.61	38.74	0.10
Early Anglo-Saxon	3	25.62	3.38	21.78	28.18	
Late Anglo-Saxon	1	34.14	-	-	-	
Medieval	12	38.29	6.09	26.87	49.14	
Modern Day	6	36.61	3.26	31.36	41.09	

Table 5.2. *Kruskal-Wallis test with Monte Carlo resampling procedures comparing dentine areas between maxillary and mandibular first molars. Due to uneven distribution of sample sizes between populations and tooth positions, differences were tested using 19 (10 upper and 9 lower) randomly selected samples, four (two upper and two lower) from each population, except the Late Anglo-Saxons where only one mandibular tooth was available. Significant results are marked in bold.*

	Total sample					Analysed sample			
Molar Position	N	Mean	S.D	Min	Max	N	Mean	S.D	Sig.
Maxillary	32	34.90	6.28	21.78	47.85	10	32.90	4.15	**0.00***
Mandibular	25	39.35	5.52	28.18	52.02	9	40.04	7.21	

*$p<0.001$

Table 5.3. *Comparison of difference in the mean values for each population of each DSR region between the maxillary and mandibular teeth using Mann-Whitney U tests.*

Population	Feature	M1 Position	N	Mean	S.D	Sig. (2-tailed)
Roman	Inner Cuspal DSR	Maxillary	4	3.68	0.33	0.85
		Mandibular	3	3.78	0.21	
	Mid Cuspal DSR	Maxillary	4	4.15	0.13	0.34
		Mandibular	4	4.25	0.22	
	Outer Cuspal DSR	Maxillary	4	4.54	0.29	0.85
		Mandibular	3	4.59	0.13	
	Inner Lateral DSR	Maxillary	5	3.72	0.38	0.93
		Mandibular	6	3.62	0.19	
	Mid Lateral DSR	Maxillary	5	4.20	0.49	0.79
		Mandibular	6	4.19	0.16	
	Outer Lateral DSR	Maxillary	5	4.47	0.46	0.66
		Mandibular	6	4.65	0.20	
Early Anglo-Saxon	Inner Cuspal DSR	Maxillary	4	3.66	0.23	0.62
		Mandibular	3	3.74	0.16	
	Mid Cuspal DSR	Maxillary	4	3.98	0.27	1.00
		Mandibular	3	3.98	0.29	
	Outer Cuspal DSR	Maxillary	4	4.39	0.28	1.00
		Mandibular	2	4.51	0.41	
	Inner Lateral DSR	Maxillary	7	3.31	0.19	0.73
		Mandibular	5	3.58	0.27	
	Mid Lateral DSR	Maxillary	7	3.83	0.23	0.63
		Mandibular	5	3.90	0.16	
	Outer Lateral DSR	Maxillary	7	4.26	0.15	0.75
		Mandibular	5	4.22	0.30	
Late-Anglo-Saxon	Inner Cuspal DSR	Maxillary	14	3.31	0.27	0.30
		Mandibular	3	3.19	0.19	
	Mid Cuspal DSR	Maxillary	15	3.68	0.28	0.36
		Mandibular	3	3.48	0.10	
	Outer Cuspal DSR	Maxillary	15	3.99	0.35	0.73
		Mandibular	3	4.01	0.13	
	Inner Lateral DSR	Maxillary	16	3.40	0.25	0.10
		Mandibular	3	3.17	0.17	
	Mid Lateral DSR	Maxillary	16	3.65	0.34	0.48
		Mandibular	3	3.48	0.34	
	Outer Lateral DSR	Maxillary	16	3.89	0.39	0.87
		Mandibular	3	3.83	0.48	
Medieval	Inner Cuspal DSR	Maxillary	16	3.32	0.22	0.34
		Mandibular	7	3.44	0.29	
	Mid Cuspal DSR	Maxillary	18	3.58	0.27	0.27
		Mandibular	9	3.81	0.22	
	Outer Cuspal DSR	Maxillary	18	3.88	0.31	0.34
		Mandibular	9	3.96	0.21	
	Inner Lateral DSR	Maxillary	19	3.33	0.26	0.57
		Mandibular	10	3.42	0.25	
	Mid Lateral DSR	Maxillary	19	3.63	0.26	0.51
		Mandibular	10	3.69	0.35	
	Outer Lateral DSR	Maxillary	18	3.87	0.29	0.34
		Mandibular	9	3.93	0.18	
Modern Day	Inner Cuspal DSR	Maxillary	3	2.95	0.43	0.22
		Mandibular	11	3.27	0.35	
	Mid Cuspal DSR	Maxillary	4	3.31	0.41	0.17
		Mandibular	11	3.59	0.38	
	Outer Cuspal DSR	Maxillary	4	3.40	0.17	0.40
		Mandibular	11	3.88	0.34	
	Inner Lateral DSR	Maxillary	4	3.06	0.25	0.66
		Mandibular	11	3.16	0.20	
	Mid Lateral DSR	Maxillary	4	3.32	0.26	0.57
		Mandibular	11	3.45	0.34	
	Outer Lateral DSR	Maxillary	4	3.45	0.21	0.07
		Mandibular	11	3.81	0.42	

Table 5.4. *Comparison of difference in the mean values for each population, of each enamel thickness feature and supplementary thickness data, between the maxillary and mandibular teeth using Mann-Whitney U tests.*

Thickness features						
Population	Feature	M1 Position	N	Mean	S.D	Sig. (2-tailed)
Roman	RET	Maxillary	3	17.97	1.61	0.20
		Mandibular	3	16.08	1.51	
	LT	Maxillary	4	1.39	0.06	0.35
		Mandibular	6	1.31	0.14	
	CT	Maxillary	3	1.58	0.21	0.40
		Mandibular	3	1.34	0.11	
Early Anglo-Saxon	RET	Maxillary	4	20.53	1.84	0.22
		Mandibular	3	22.83	1.76	
	LT	Maxillary	7	1.39	0.18	0.78
		Mandibular	4	1.41	0.24	
	CT	Maxillary	5	1.08	0.22	0.57
		Mandibular	3	1.34	0.49	
Late Anglo-Saxon	RET	Maxillary	7	16.39	1.50	0.50
		Mandibular	1	14.97	-	
	LT	Maxillary	16	1.43	0.11	0.25
		Mandibular	3	1.52	0.09	
	CT	Maxillary	12	1.17	0.23	0.79
		Mandibular	2	1.15	0.33	
Medieval	RET	Maxillary	13	16.33	1.79	0.89
		Mandibular	9	16.27	02.45	
	LT	Maxillary	18	1.44	0.22	0.98
		Mandibular	9	1.43	0.18	
	CT	Maxillary	18	1.08	0.25	0.56
		Mandibular	9	1.03	0.15	
Modern Day	RET	Maxillary	2	15.43	2.87	0.08
		Mandibular	8	20.19	3.45	
	LT	Maxillary	4	1.77	0.45	0.66
		Mandibular	11	1.64	0.31	
	CT	Maxillary	4	1.23	0.71	1.00
		Mandibular	11	1.29	0.55	
RET component thickness data						
Roman	Enamel cap area	Maxillary	4	20.98	2.72	0.22
		Mandibular	3	18.49	2.17	
	EDJ length	Maxillary	4	18.58	1.78	0.62
		Mandibular	3	19.11	0.44	
	AET	Maxillary	4	1.12	0.13	0.22
		Mandibular	3	0.96	0.11	
Early Anglo-Saxon	Enamel cap area	Maxillary	4	21.83	2.64	0.11
		Mandibular	3	18.43	2.96	
	EDJ length	Maxillary	4	18.76	0.79	0.06
		Mandibular	3	16.74	0.82	
	AET	Maxillary	4	1.16	0.09	0.62
		Mandibular	3	1.10	0.13	
Late Anglo-Saxon	Enamel cap area	Maxillary	9	22.52	3.70	0.80
		Mandibular	1	22.2	-0.40	
	EDJ length	Maxillary	9	20.00	1.51	0.40
		Mandibular	1	22.34	-	
	AET	Maxillary	9	1.12	0.19	0.60
		Mandibular	1	0.99	-	
Medieval	Enamel cap area	Maxillary	13	21.02	2.70	0.47
		Mandibular	12	20.32	2.15	
	EDJ length	Maxillary	13	20.56	1.18	0.32
		Mandibular	12	19.71	1.93	
	AET	Maxillary	13	1.02	0.10	1.00
		Mandibular	12	1.03	0.15	
Modern Day	Enamel cap area	Maxillary	2	15.54	6.95	0.14
		Mandibular	6	21.87	1.41	
	EDJ length	Maxillary	2	16.58	1.63	0.14
		Mandibular	6	19.44	1.55	
	AET	Maxillary	2	0.92	0.32	0.42
		Mandibular	6	1.13	0.12	

5.3.2 Enamel thickness measures

Table 5.5 reports the mean RET, LT, and CT of each population along with the standard deviations and results of the Kruskul-Wallis tests. The Early Anglo-Saxon RET was significantly larger than both the Late Anglo-Saxon (p<0.001) and Medieval (p<0.001) populations (see Fig. 5.2). Cuspal enamel thickness (CT) did not differ significantly between any of the populations (see Fig. 5.3), but the lateral enamel thickness (LT) of the modern day population was significantly thicker than the Roman population (p = 0.04; see Fig. 5.3). No significant trends were found in RET ($p < 0.24$), CT ($p < 0.21$) ($p < 0.27$), or LT measurements across the 2000 years represented in this sample (see Table 5.5).

Table 5.5. *Results of the Kruskal-Wallis, post-hoc pairwise Dunn-Bonferroni, and Jonckheere-Terpstra tests for the comparison of RETs, LTs, and CTs between each population. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. Inclusion of - and + on Jonckheere-Terpstra results indicate a trend towards reduction or increase respectively. Significant results are marked in bold.*

Population	N	Mean	Min	Max	S.D	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.				Jonckheere-Terpstra Sig.
							Early Anglo-Saxon	Late Anglo-Saxon	Medieval	Modern day	
Relative enamel thickness											
Roman	6	17.02	14.70	19.16	1.74	**0.00***	0.13	1.00	1.00	1.00	+0.24
Early Anglo-Saxon	7	21.52	19.00	24.01	2.05			**0.00***	**0.00***	0.98	
Late Anglo-Saxon	8	16.21	13.79	18.20	1.48				1.00	0.39	
Medieval	22	16.30	12.56	21.04	2.03					0.14	
Modern day	10	19.24	13.40	27.24	3.77						
Lateral enamel thickness											
Roman	10	1.34	1.14	1.50	0.12	**0.04**	1.00	1.00	1.00	**0.04**	+0.21
Early Anglo-Saxon	11	1.39	1.09	1.77	0.19			1.00	1.00	0.20	
Late Anglo-Saxon	19	1.44	1.24	1.68	0.11				1.00	0.81	
Medieval	27	1.44	1.07	1.86	0.21					0.24	
Modern day	15	1.68	1.17	2.36	0.57						
Cuspal enamel thickness											
Roman	6	1.46	1.21	1.78	0.20	0.07					-0.27
Early Anglo-Saxon	8	1.18	0.80	1.77	0.34						
Late Anglo-Saxon	14	1.17	0.80	1.53	0.23						
Medieval	27	1.16	1.07	1.86	0.22						
Modern day	15	1.27	0.57	2.27	0.57						

*p<0.001

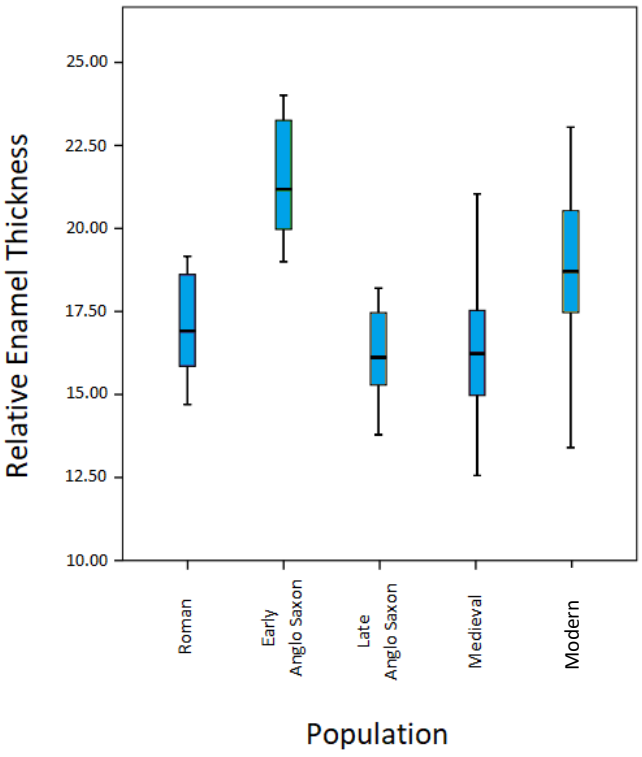

Fig. 5.2. *Plot of RET data distribution of each population. The central line displays the median thickness for the associated population.*

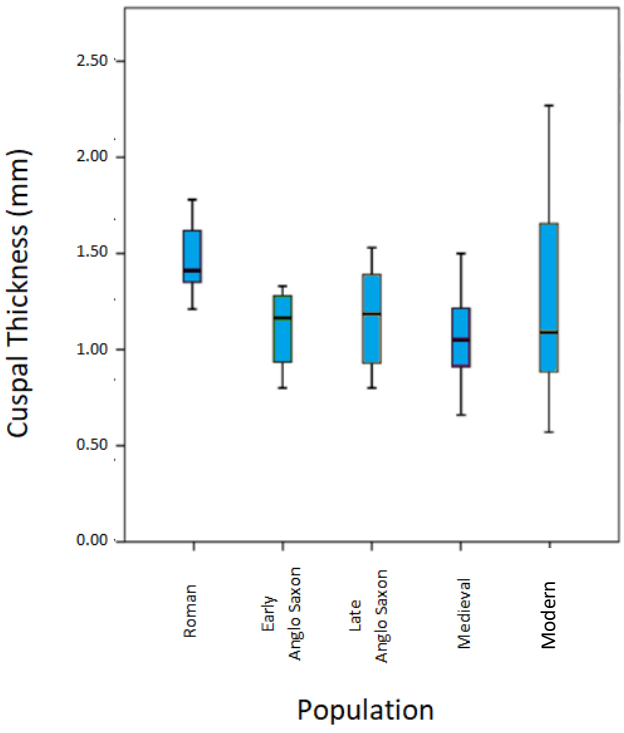

Fig. 5.3. *Plot of CT data distribution of each population. The central line displays the median thickness for the associated population.*

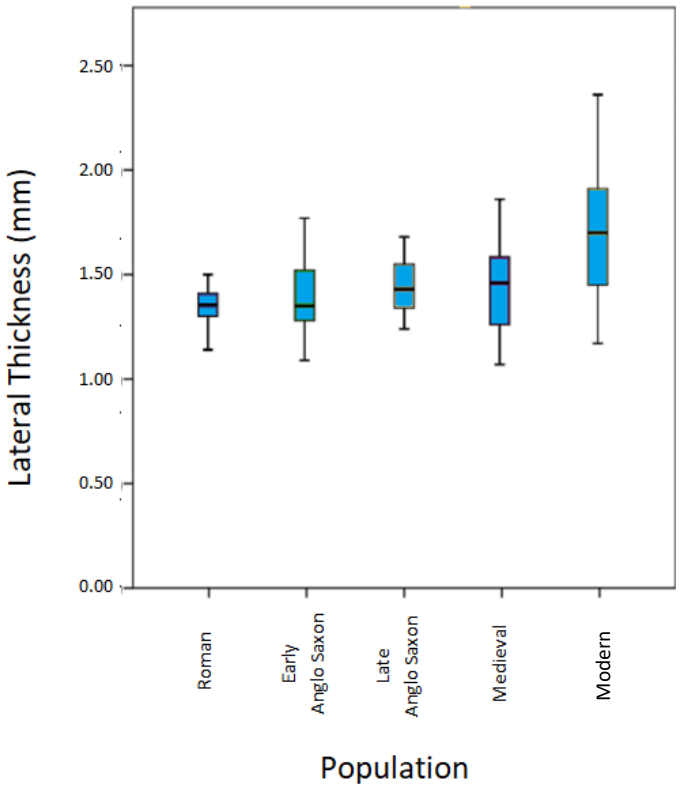

Fig. 5.4. *Plot of LT data distribution of each population. The central line displays the median thickness for the associated population.*

5.3.3 Cuspal daily secretion rates

Table 5.6 reports the mean inner, mid, and outer cuspal DSRs of each population along with the standard deviations and results of the Kruskul-Wallis, post-hoc Dunn-Bonferroni pairwise comparisons, and Jonckheere-Terpstra tests.

Inner cuspal DSRs show a significant slowing trend of ameloblast daily secretion rates across time (p<0.001). The modern day mean is 0.52µm/day slower than the Roman mean rate (p<0.001; see Fig. 5.5). The mean DSRs from the modern day sample were also significantly slower than both the Roman (p<0.001) and Early Anglo-Saxon (p<0.001) secretion rates. The Late Anglo-Saxon DSRs were significantly slower than the Roman (p=0.04) and Early Anglo-Saxon (p=0.04) secretion rates. Differences between the DSRs of inner enamel of the more ancient populations were less marked. Rates only increased slightly from the inner enamel of the Roman period to the Early Anglo-Saxon period (0.03µm/day mean difference), and only a 0.02µm/day difference between the Late Anglo-Saxon and Medieval population.

Mid cuspal DSR's show a similar significant trend of slowing (p<0.001); the modern day mean is 0.69µm/day slower than the Roman mean (Fig. 5.5). DSRs from this enamel region in the Roman sample were significantly faster compared to the Late Anglo-Saxon (p = 0.04), Medieval (p<0.001), and the modern day (p<0.001) populations. Rates did not differ significantly between the Late Anglo-Saxon and Medieval populations.

Outer cuspal DSRs also showed a significant slowing trend (p<0.001); the modern day mean is 0.81µm/day slower than the Roman mean, which represents the most significant decrease in cuspal DSR across this 2000 year period (see Fig. 5.5). Pairwise comparisons revealed significant differences between five pairs of populations. The mean DSRs were significantly faster in the Roman population compared to the Late Anglo-Saxon (p = 0.02), Medieval (p<0.001), and modern day (p<0.001). The Early Anglo-Saxon population also presented significantly faster secretion rates than the Medieval (p = 0.01) and the modern day (p<0.001) populations. Again, the Roman and Early Anglo-Saxon, and Late Anglo-Saxon and Medieval population pairs showed notably less reduction in secretion rates over time – 0.16µm/day and 0.09µm/day respectively.

Table 5.6. *Results of the Kruskal-Wallis, post-hoc pairwise Dunn-Bonferroni, and Jonckheere-Terpstra tests for the comparison of* ***cuspal*** *DSRs (µm/day) between each population. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. Inclusion of - on Jonckheere-Terpstra results indicate a trend towards reduction. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	Min	Max	S.D	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.				Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Late Anglo-Saxon	Medieval	Modern day	
Inner	Roman	7	3.72	3.23	4.02	0.26	**0.00***	1.00	**0.04**	0.09	**0.00***	**-0.00***
	Early Anglo-Saxon	7	3.69	3.48	4.01	0.19			**0.04**	0.90	**0.00***	
	Late Anglo-Saxon	17	3.34	3.02	4.20	0.27				1.00	1.00	
	Medieval	23	3.36	2.82	3.84	0.24					1.00	
	Modern day	14	3.20	2.54	3.99	0.38						
Mid	Roman	8	4.21	4.07	4.58	0.42	**0.00***	1.00	**0.00***	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	7	3.98	3.69	4.40	0.25			0.25	0.40	0.06	
	Late Anglo-Saxon	18	3.65	3.41	4.66	0.32				1.00	1.00	
	Medieval	27	3.65	2.91	4.13	0.28					1.00	
	Modern day	15	3.52	2.96	4.18	0.35						
Outer	Roman	7	4.56	4.12	4.76	0.22	**0.00***	1.00	**0.02**	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	6	4.43	4.20	4.81	0.29			0.10	**0.01**	**0.00***	
	Late Anglo-Saxon	18	3.99	3.41	4.66	0.32				1.00	1.00	
	Medieval	27	3.90	3.13	4.76	0.28					1.00	
	Modern day	15	3.75	3.15	4.58	0.37						

*$p < 0.001$

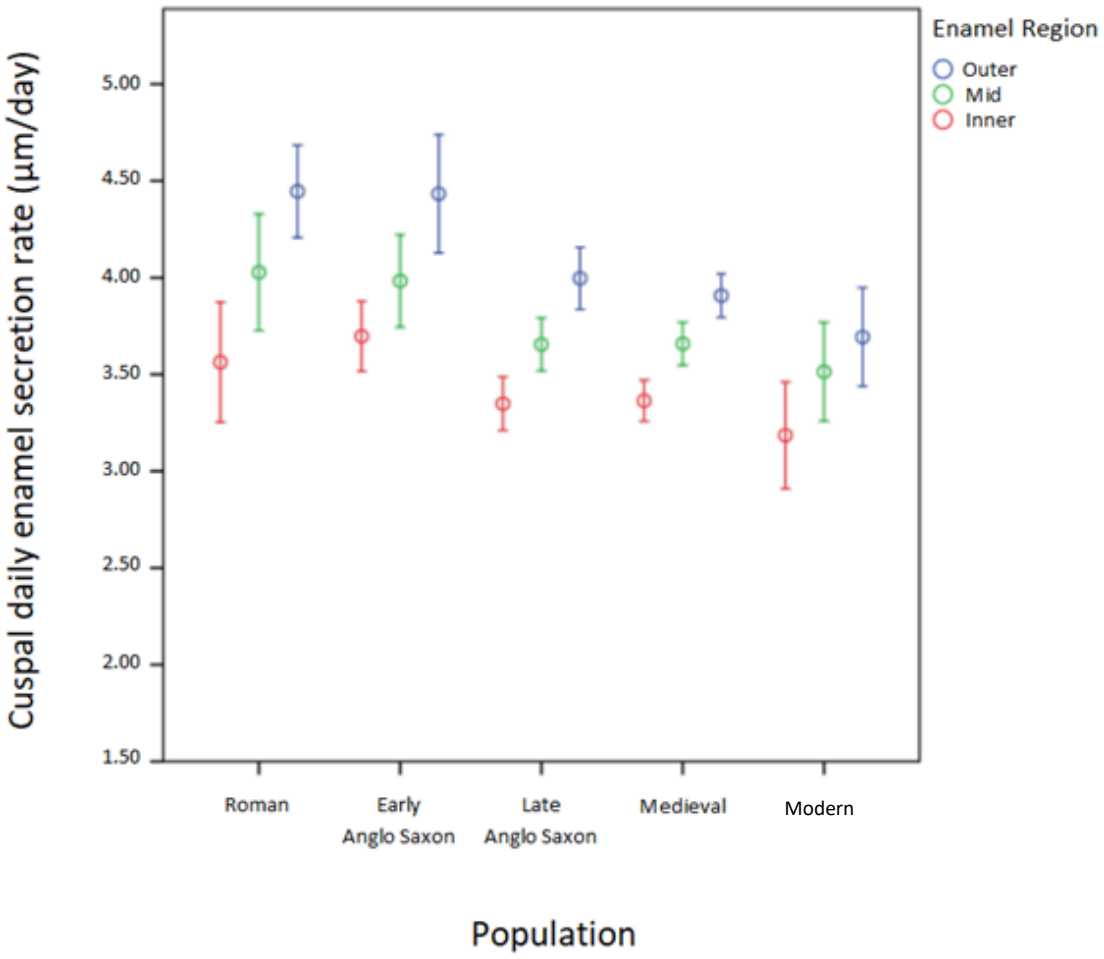

Fig. 5.5. *Circles depict the mean cuspal DSRs of each enamel region for each population. The figure illustrates the general decrease in secretion rates over time. The error bars display one standard deviation either side of the mean.*

5.3.4 Lateral daily secretion rates

Table 5.7 reports the mean inner, mid, and outer lateral DSRs of each population along with the standard deviations and results of the Kruskal-Wallis, post-hoc Dunn-Bonferroni pairwise comparisons, and Jonckheere-Terpstra tests.

The inner lateral DSR data show DSRs slow over time, with a significant negative trend from the more ancient to the modern period (p < 0.01; see Fig. 5.6). The modern day mean DSR is 0.52µm/day slower than the Roman mean. The Late Anglo-Saxon and Medieval populations again presented similar secretion rates with no significant differences. Dunn-Bonferroni revealed a single

difference where the Roman population presented significantly faster secretion rates compared to the modern day population (p<0.001).

Secretion rates slowed slightly between the more ancient to the modern period in the mid lateral DSR data, but the trend was insignificant (see Fig. 5.6). The mean secretion rate of the modern day population was 0.77µm/day slower than the Roman population. However, the Roman population presented significantly faster DSRs than the Late Anglo-Saxon (p<0.001), Medieval (p<0.001), and the modern day (p<0.001) populations. The Early Anglo-Saxon population also presented significantly faster secretion rates than the modern day population (p<0.001). The Medieval and Late Anglo-Saxon populations again presented similar secretion rates (only 0.02µm/day mean difference).

Outer lateral DSRs show a significant negative, slowing trend over time (p<0.001; see Fig. 5.6). The modern day mean rate was 0.86 µm/day slower than the Roman mean, which like the cuspal outer DSR, represents the most significant slowing. The mean DSRs were significantly faster in the Roman population compared to the Late Anglo-Saxon (p<0.001), Medieval (p<0.001), and modern day (p<0.001). The Early Anglo-Saxon population also presented significantly faster secretion rates than the Medieval (p = 0.03) and the modern day (p<0.001) populations. Again, the Late Anglo-Saxon and Medieval population pairs were nearly identical (0.01 µm/day mean difference).

Table 5.7. *Results of the Kruskal-Wallis, post-hoc pairwise Dunn-Bonferroni, and Jonckheere-Terpstra tests for the comparison of **lateral** DSRs (µm/day) between each population. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. Inclusion of - on Jonckheere-Terpstra results indicate a trend towards reduction. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	Min	Max	S.D	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.				Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Late Anglo-Saxon	Medieval	Modern day	
Inner	Roman	11	3.66	3.30	4.36	0.28	**0.00***	0.53	0.11	0.06	**0.00***	**-0.01**
	Early Anglo-Saxon	12	3.42	3.11	4.03	0.20			1.00	1.00	0.11	
	Late Anglo-Saxon	19	3.36	2.87	3.94	0.25				1.00	0.24	
	Medieval	29	3.36	2.84	3.81	0.26					0.09	
	Modern day	15	3.14	2.84	3.66	0.21						
Mid	Roman	11	4.19	3.67	4.78	0.33	**0.00***	1.00	**0.00***	**0.00***	**0.00***	-0.64
	Early Anglo-Saxon	12	3.85	3.54	4.21	0.20			0.78	0.48	**0.00***	
	Late Anglo-Saxon	19	3.63	2.86	4.14	0.33				1.00	0.65	
	Medieval	29	3.65	3.12	4.29	0.29					0.09	
	Modern day	15	3.42	2.82	4.04	0.32						
Outer	Roman	11	4.57	3,97	5.13	0.34	**0.00***	1.00	**0.00***	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	12	4.24	3.85	4.63	0.21			0.08	**0.03**	**0.00***	
	Late Anglo-Saxon	19	3.88	2.89	4.57	0.39				1.00	0.85	
	Medieval	27	3.89	3.47	4.50	0.26					0.83	
	Modern day	15	3.71	3.13	4.99	0.42						

*p<0.001

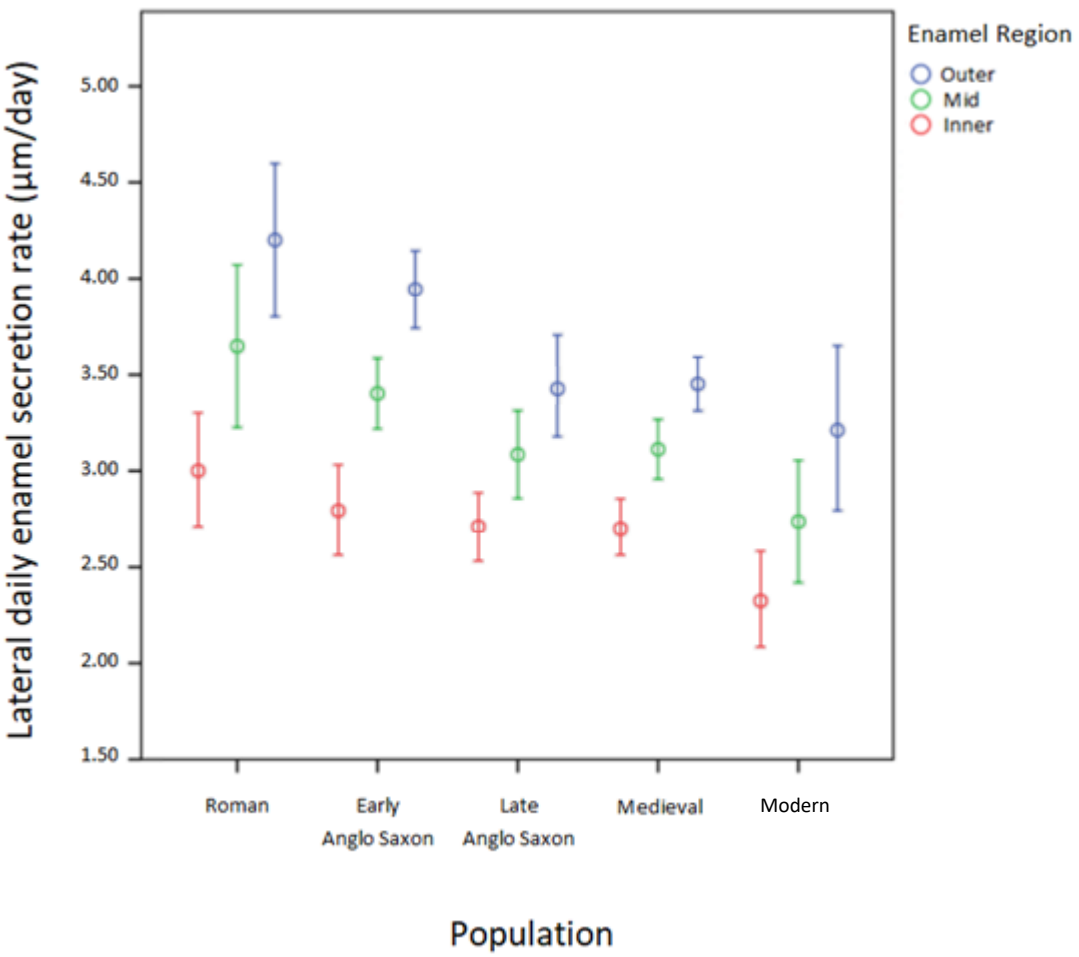

Fig. 5.6. *Circles depict the mean lateral DSRs of each region and population, displaying their general decrease over time. The error bars display one standard deviation around the mean.*

5.4 Discussion

With the exception of the Early Anglo Saxon sample, all three measures of first molar enamel thickness (RET, CT, and LT) remained generally similar when compared between the British populations that spanned a 2000-year period of time. In contrast, there was a significant trend towards slower DSRs in samples from the more recent periods when equivalent regions of enamel were compared between populations, with the exception of the mid-lateral region.

5.4.1 Relative enamel thickness

All measures of RET, CT, and LT lie within previously published ranges for human permanent first molars (Macho and Berner, 1993; Schwartz, 2000a; Suwa and Kono, 2005; Reid and Dean, 2006;

Smith et al., 2006a; Olejniczak et al., 2008; Mahoney, 2010) (Table 5.8). Compared to the other modern human groups that have been studied, all the British populations (except the Early Anglo-Saxons) have fairly thin first molar RET. Smith and colleagues (2006a) published mean RETs ranging from 20.16 (maxillary Medieval Danish M1s) to 22.62 (mandibular modern South African M1s). A study by Olejniczak and colleagues (2008) presented mandibular modern human mesial cusp RET ranges more similar to those presented here of 15.87 to 17.05. Here, the Early Anglo-Saxons had a mean RET of 21.52, and while the other populations ranged from 16.21 to 19.24 (see Table 5.5). The Early Anglo-Saxon values are still well within the range of published modern human RETs.

Table 5.8. *Ranges/means (± 1SD) for modern human M1 RET, CT, and LT.*

Source/Population	N	RET	CT (mm)	LT (mm)
Past Studies				
Smith et al., 2006a[†]	37	13.95-23.86		
Smith et al., 2006a[‡]	55	11.67-22.62		
Olejniczak et al., 2008[†*]	6	17.05		
Olejniczak et al., 2008[‡*]	1	15.87		
Schwartz, 2000a[¶]	9		1.77 ± 0.48	
Reid and Dean, 2006[§]	15		1.21 ± 0.241	
Reid and Dean, 2006[¶]	37		1.09 ± 0.113	
Mahoney, 2010[¶]	69		1.13 ± 0.22	1.43 ± 0.19
Suwa and Kono, 2005[§]	31		0.85 ± 0.206	1.47 ± 0.119
Suwa and Kono, 2005[¶]	37		1.12 ± 0.160	1.44 ± 0.113
Macho and Berner, 1993[§]	21			1.36 ± 0.17
This study				
Roman	6-10	17.02 ± 1.74	1.46 ± 0.20	1.34 ± 0.12
Early Anglo-Saxon	7-11	21.52 ± 2.05	1.18 ± 0.34	1.39 ± 0.19
Late Anglo-Saxon	8-19	16.21 ± 1.48	1.17 ± 0.23	1.44 ± 0.11
Medieval	22-27	16.30 ± 2.03	1.16 ± 0.22	1.44 ± 0.21
Modern day	10-15	19.24 ± 3.77	1.27 ± 0.57	1.68 ± 0.57

[†] data for maxillary M1s

[‡] data for mandibular M1s

[§] data for paracone cusp

[¶] data for metaconid cusp

* includes 3D measurements

Despite falling within previously reported ranges, the RET value for the Early Anglo-Saxon sample was significantly greater than Late Anglo-Saxon (p<0.001) and Medieval (p<0.001) populations (see Fig. 5.2; Table 5.5). It is possible that this thicker enamel in the Early Anglo-Saxon

population compared to the late Anglo-Saxon and Medieval periods might reflect a shift to a diet containing a high prevalence of harder or abrasive foodstuffs (e.g. Dumont, 1995; Hlusko et al., 2004; Grine, 2005; Pampush, et al., 2013). Alternatively, these slight but significant variations between populations could relate to differences in the linear dimensions of the tooth crown. This seems less likely though, as metric dimensions of first permanent molars for Anglo-Saxon and Medieval populations do not seem to vary greatly between these time periods (Aris et al., 2018).

Comparative analyses of non-human primate (e.g. Molnar and Gantt, 1977; Grine, 1981; Lucas, et al., 1985, 2008, 2013; Martin et al., 2003; Pampush et al., 2013) and human populations (Le Luyer et al., 2014) have found that those groups consuming foodstuffs that incur heavy wear tend to have thicker first molar enamel and greater RET. Given the coastal location of the Early Anglo-Saxon population, it is possible this population had a greater dependence on marine foods compared to the later Anglo-Saxon populations, resulting in greater attrition. Isotopic analysis by Mays and Beavan (2012) has identified increased levels of marine food dependency in Early Anglo-Saxon samples. High levels wear have also been linked to increased consumption of grit from sand in marine diets (Littleton and Frohlich, 1993). It is therefore possible that the greater RET of the Early Anglo Saxons could relate to a more abrasive diet.

Migration and gene flow might have also contributed to the difference in RET between the Early Anglo-Saxon and Late Anglo-Saxon and Medieval samples. Rapid increases in migration into Britain followed the end of Roman rule, with particular influx of Germanic people corresponding with political and cultural upheavals (Scull, 1993; Privat, O'Connell, and Richards, 2002; Lightfoot et al., 2009). This high level of immigration from mainland Europe may have also factored into the high RET observed in the Early Anglo-Saxon sample, if the immigrating groups had relatively thicker RET than those of the indigenous populations. Shifts towards more abrasive marine diets in the Anglo-Saxon period, and the migration and subsequent gene flow following the end of Roman rule in Britain provide two explanatory options for the high RET of the Early Anglo-Saxon population. However, further research into this period is required to support of refute these hypotheses.

5.4.2 Linear measures of enamel thickness

There were few differences in cuspal and lateral thickness between populations and no significant directional trends over time (see Table 5.5). The only significant difference (p = 0.04) was thicker lateral enamel in the modern sample (1.68 ± 0.57 mm) compared to the Roman sample (1.34 ± 0.12 mm) (see Table 5.5). Otherwise, both LT and CT remained relatively consistent across all populations

with means ranging from 1.34mm to 1.68mm and 1.16mm to 1.46mm respectively (see Table 5.5). Given the similarity of RET values for these populations, it is unsurprising that their linear enamel thickness measures are similarly indistinguishable. This is especially true considering that the maxillary paracone and mandibular metaconid experience less pressure during mastication compared to other mesial cusps of M1 teeth (protocone and protoconid) and are therefore less likely to undergo selective pressure (Macho and Berner, 1993; 1994; Shellis, et al., 1998; Grine, 2005; Kelley and Swanson, 2008).

5.4.2.1 Cuspal enamel thickness

Despite slight variation between populations, all CT means fell within the known ranges for modern human cuspal thickness (see Table 5.8), including those of previously analysed modern-day archaeological British (Mahoney, 2008) and Slavic (Schwartz, 2000a) populations and geographically diverse modern populations including those from southern Africa (Reid and Dean, 2006), North America, Japan, and Germany (Suwa and Kono, 2005).

The CT data shows no significant directional trends over the time periods considered here (see Table 5.5; Fig. 5.3). While the Roman mean CT was the thickest at 1.46mm, and the remaining archaeological populations were thinner, ranging from 1.16mm to 1.18mm, there were no discernible temporal trends. Although the Early Anglo-Saxon had high RET values compared to the other populations, it seems that they possessed a reduced volume of dentine encapsulated by the enamel cap (Table 5.2) resulting in a larger RET, rather than a noticeable increase in enamel at any crown region.

5.4.2.2 Lateral enamel thickness

The lateral thicknesses of the first molar cusps analysed here are comparable to those in the published literature (see Table 5.4) (Macho and Berner, 1993; Suwa and Kono, 2005; Mahoney, 2010). The Roman population presents significantly thinner LTs than the modern-day sample (p = 0.04, Table 5.5). The biological and evolutionary significance of this difference is difficult to identify. However, lateral enamel undergoes increased stress from increasing occlusal loads, resulting in higher levels of hoop stress that can cause margin fractures (Lawn and Lee, 2009). Interspecific analyses of lateral first molar thickness have suggested that the thickening of lateral aspects of first molars cusps could be related to dietary differences between modern humans and extinct members of the *Homo* genus (Pan et al., 2016). The significant thickening of enamel between the Roman and

modern-day LTs could relate to a shift in mechanical demands of food. However, there was no significant trend to the changes in LT across the 2000 year periods, so this possibility should be considered with caution.

5.4.3 Daily secretion rates

Daily secretion rates fall within the known ranges of published modern human enamel DSRs (Beynon et al., 1991a; Dean 1998; Lacruz et al., 2006; Lacruz and Bromage, 2006; Mahoney, 2008) (Table 5.9).

Table 5.9. *Mean modern human molar DSRs (µm/day ± 1SD) for cuspal and lateral regions.*

Source/Population	N	Inner	Mid	Outer
Cuspal DSRs				
Past studies				
Beynon et al (1991b)†	11-15	2.7 ± 0.4	4.3 ± 0.5	5.1 ± 0.7
Lacruz and Bromage (2006)†	10	2.80 ± 0.43	4.50 ± 0.55	5.20 ± 0.58
Smith et al (2007b)	21	2.55	4.34	5.45
Mahoney (2008)‡	13	3.01 ± 0.48	4.26 ± 0.54	4.61 ± 0.64
This study				
Roman†	7	3.72 ± 0.26	4.21 ± 0.42	4.56 ± 0.22
Early Anglo-Saxon†	6-7	3.69 ± 0.19	3.98 ± 0.25	4.43 ± 0.29
Late Anglo-Saxon†	17-18	3.34 ± 0.27	3.65 ± 0.32	3.99 ± 0.32
Medieval†	23-27	3.36 ± 0.24	3.65 ± 0.28	3.90 ± 0.28
Modern day†	14-15	3.20 ± 0.38	3.52 ± 0.35	3.75 ± 0.37
Lateral DSRs				
Past studies				
Beynon et al (1991b)	12-15	2.6 ± 0.5	4.0 ± 0.4	5.0 ± 0.5
Lacruz and Bromage (2006)	10	2.70 ± 0.42	4.30 ± 0.67	4.80 ± 0.67
This study				
Roman†	11	3.66 ± 0.28	4.19 ± 0.33	4.57 ± 0.34
Early Anglo-Saxon†	12	3.42 ± 0.20	3.85 ± 0.20	4.24 ± 0.21
Late Anglo-Saxon†	19	3.36 ± 0.25	3.63 ± 0.33	3.88 ± 0.39
Medieval†	27-29	3.36 ± 0.26	3.65 ± 0.29	3.89 ± 0.26
Modern day†	15	3.14 ± 0.21	3.42 ± 0.32	3.71 ± 0.42

†data for M1 enamel
‡data for the metaconid cusp

5.4.3.1 Daily secretion rates compared between enamel regions

The daily enamel growth rate of each population followed the expected pattern of ameloblast activity, with increasing DSRs from inner to outer enamel regions (e.g. Schwartz et al., 2001; Mahoney, 2008). Lacruz and Bromage (2006) reported an increase in the mean secretion rates of 10

modern human molars, when compared between the inner to mid, and mid to outer-cuspal enamel region (mean increase in the mean secretion rate of 1.70 and 0.70µm/day, respectively). Similar increases were observed by Mahoney (2008) in 13 British Bronze Age mandibular first molars, where metaconid secretion rates increased by 1.25µm/day between the inner and mid regions and 0.35µm/day between the mid and outer-cuspal areas. While this pattern was present in the modern clinical sample, the rate of increase was not nearly as steep as reported elsewhere (mean increase of 0.32 and 0.13µm/day between inner to mid and mid to outer, respectively; see Table 5.6). This suggests that such rapid increases in DSRs through the inner to outer regions are not always present in modern human first molars.

5.4.3.2 Cuspal daily secretion rates compared between populations

Cuspal enamel secretion rates decreased over time (p<0.001; see Table 5.6). A significant decrease in cuspal DSRs over such a short period of time has not been documented previously in modern human populations for permanent teeth. There was however a notable similarity between the mean cuspal DSRs of the Roman and Early Anglo-Saxon populations (respectively: inner +0.03µm/day, mid +0.23µm/day, outer +0.13µm/day), and between the Late Anglo-Saxon and Medieval populations (respectively: inner +0.02µm/day, mid 0.00µm/day, outer +0.09µm/day). Considering these population pairs are dated to similar periods (Roman/Early Anglo-Saxon – 70-400AD/500-600AD; Late Anglo-Saxon/Medieval – 800-1200AD/1100-1600AD) this is unsurprising.

5.4.3.3 Lateral daily secretion rates compared between populations

Like cuspal DSRs, lateral DSRs slowed over the last 2000 years. However, this trend was only significant in the inner and outer-cuspal regions (see Table 5.7). Similar to the cuspal DSRs, there was minimal difference between the mean DSRs of the Late Anglo-Saxon and Medieval populations across all regions of lateral enamel (inner 0.00µm/day; mid 0.02µm/day; outer 0.01µm/day). This is unsurprising, considering these two populations are dated to overlapping periods (800-1200AD and 1100-1600AD respectively). Analysis of the lateral thicknesses of the M1 cusps also identified minimal variation between the British populations presented here and those within published literature (see Table 5.8) (Macho and Berner, 1993; Suwa and Kono, 2005; Mahoney, 2010). Given the large number of significant variations observed in DSRs that are linked to enamel of the same relative thickness (both cuspal and lateral), it appears that underlying growth mechanisms of enamel

can change even though the final enamel thickness does not, when compared between human populations.

The volume of published modern human lateral enamel thickness data is considerably smaller than that of cuspal regions (see Table 5.9). The lateral DSRs for the British populations included here generally fall a bit above the published inner means, below the middle means, and below the outer means (see Table 5.9). However, the limited available data regarding M1 lateral enamel DSRs (Beynon et al., 1991b; Lacruz and Bromage, 2006) is too small to be considered representative of modern humans as a species. Thus, deviation from the published ranges is not surprising when analysing five new human populations.

5.4.4 Relationship between DSRs and linear thicknesses

Previous research has considered potential relationships between DSRs and enamel thickness within human first molars (Mahoney, 2008). Additional papers have considered other periodic biorhythms associated with enamel growth, in particular, Retzius periodicity (a circaseptum enamel growth pattern) has been associated with first molar enamel thickness. In permanent first molar slower periodicities have been found to predict thicker enamel (Mahoney et al., 2018). The opposite has been observed in deciduous molars (Mahoney et al., 2018). However, the results presented here suggest that there is not a strong connection between daily enamel secretion rates and thickness when compared between samples of first molars from different periods. While lateral and cuspal DSRs slowed significantly over this 2000 year time period in Britain, there was no complimentary change in RET, LT, and CT over the same period of time (see Tables 5.5, 5.6, and 5.7).

Lacruz and Bromage (2006) found that thicker enamel in fossil hominins is associated with faster secretion rates. However, the results presented here suggest that, while first molar enamel thickness can remain generally similar across time between human populations, DSRs may fluctuate independently. This is most evident in the lateral first molar enamel, where the modern-day sample was found to have significantly thicker lateral enamel than Romans (see Table 5.5) yet have significantly slower DSRs than Romans in every single enamel region (see Tables 5.6 and 5.7), which is opposite of what would be expected given the results of Lacruz and Bromage (2006). Similarly, the Roman and Medieval populations show no significant RET, LT, or CT differences (see Table 5.5), but the Romans had significantly faster DSRs in the cuspal and lateral middle and outer enamel regions (see Tables 5.6 and 5.7).

Given the numerous significant differences observed in DSRs and the lack of significant differences in enamel thickness measures from the same populations, it appears that inter-population variations in DSRs and enamel thickness are not tightly correlated. This could be the result of a number of factors, including but not exclusive to: features that influence enamel thickness changes between groups (e.g. dietary changes) do not affect DSRs in the same way or at all; enamel DSRs are more readily plastic than thickness and are therefore able to significantly change between groups over shorter periods of time; difference in genetic diversity between the populations analysed had a greater influence on enamel DSRs than thicknesses. Future research should therefore consider if other developmental variables such as ameloblast lifespan, crown formation time, periodicity, and/or enamel extension rate, are more strongly linked to enamel thickness when compared between different populations.

5.4.5 Conclusions

Modern human enamel thickness has remained relatively stable across British populations throughout the last 2000 years. Conversely, the underlying enamel secretion rates of both the cuspal and lateral enamel regions present a significant slowing trend from the Romano-British period to present. Thus, it seems secretion rates can vary significantly over relatively short periods of time in modern human populations. Finally, the differences in DSRs were independent of enamel thickness, suggesting that factors influencing the rate of enamel growth do not necessarily influence the final thickness of enamel, when compared between populations.

CHAPTER 6: RESEARCH CHAPTER – ENAMEL GROWTH RATES OF ANTERIOR TEETH IN MALES AND FEMALES FROM MODERN AND ANCIENT BRITISH POPULATIONS

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Co-authored with: Dr Patrick Mahoney and Dr Chris Deter

Abstract

Objectives

This study explored biological sex differences in the regional daily growth rates of human anterior enamel from modern and ancient populations in Britain.

Methods

Maxillary permanent first incisors (*n*=80) and upper canine (*n*=69) from Roman, Anglo-Saxon, Medieval, and Modern day populations were analysed using histological methods. Daily secretion rates (DSRs) were collected for inner, mid, and outer regions of cuspal and lateral enamel. Modern day samples were of known sex, archaeological individuals had sex determined using standard osteological methods. Variation in DSRs between the sexes, both between and within populations, was tested for using parametric and non-parametric tests.

Results

When all samples were pooled, there was no significant difference between males and females. Similarly no significant differences in DSRs were identified between male and females within each population. When DSRs were compared between the populations, DSRs decreased from the more ancient to the more recent populations for males, and for females. More inter-population differences were observed in males.

Discussion

This study presents evidence for the relative consistency of enamel DSRs between male and female groups within each British population. Inter-population analyses found DSRs slowed significantly between Roman and modern day populations for both sexes, with male DSRs showing the greatest variation between populations.

6.1 Introduction

Studies of enamel daily secretion rates (DSRs) of human teeth have tended to focus on permanent molars (e.g. Beynon et al., 1991b; Lacruz and Bromage, 2006; Mahoney, 2008; Aris et al., 2020b) and deciduous dentition (e.g. Birch and Dean, 2009; Mahoney, 2012, 2015). Relatively less research has been undertaken on growth rates of the anterior dentition (incisors and canines) (e.g. Fitzgerald, 1998; Reid et al., 1998a; Schwartz et al., 2001). Of these studies only a few tested for biological sex differences in the daily rate at which enamel forms (Schwartz et al., 2001). Schwartz and colleagues (2001) tested for sex differences in permanent canine DSRs in a sample of humans and non-human hominoids. Their analysis of 16 mandibular human canines revealed no difference in DSRs when compared between the sexes (Schwartz et al., 2001), though whether there are sex differences in incisor enamel growth rates has not been examined.

The aim of this study is to explore sex differences in DSRs from anterior teeth in ancient and modern populations. First, DSRs from equivalent enamel regions of permanent upper first incisors and upper canines will be compared between males and females using a pooled sample of all British populations. Second, DSRs will be compared between the sexes within each population. Third, DSRs will be compared between the populations, for males, and then for females.

6.1.1 Intraspecific study of human enamel secretion rates

The majority of human anterior tooth DSR analyses have focussed on deciduous dentition. Research by Fitzgerald and Hillson (2009) conducted histological analysis on 36 infants from 1st century AD Greece in order to study variations in appositional growth rates of enamel. Birch and Dean (2009) conducted a similar analysis with the aim of mapping the differences in DSRs across the varying regions of the enamel cap for mandibular deciduous tooth types including anterior teeth. They found that deciduous enamel DSRs varied similarly to permanent enamel, with DSRs increasing with proximity to both the cuspal and outer enamel areas. More recently Mahoney presented deciduous anterior tooth DSRs (Mahoney, 2012, 2015). Across Medieval British (Mahoney, 2012, 2015) and modern day Swedish samples (Mahoney, 2015), the mean DSRs presented were notably slower than those previously presented (Birch and Dean, 2009; Fitzgerald and Hillson, 2009). While these papers only concern deciduous teeth, they do highlight the inter-population differences present within anterior tooth types concerning their daily growth rates for modern humans.

Schwartz and colleagues (2001) compared DSRs between human males and females as part of a study into the developmental mechanisms underlying canine dimorphism in extant hominoids. Their analysis of 16 mandibular human canines revealed the expected pattern of enamel secretion whereby rates were fastest in the cuspal region and with distance from the EDJ. Rates were consistent when compared between human males and females. It was also found that there was no significant difference between the DSRs of equivalent regions between the sexes (Schwartz et al., 2001). Incisor enamel DSRs were not an aim of their study, so little is known about this aspect of daily enamel growth in this tooth type. In particular, variation between male and female groups within a wider selection of human populations, between the same populations, and for data gathered from incisor enamel, has yet to be researched.

6.2 Materials and methods

6.2.1 Dental sample

Maxillary permanent anterior teeth (*n* = 149) were selected from British populations that date to archaeological and modern periods. The incisor sample (*n* = 80) consisted of maxillary first incisors: Roman (*n* = 10); Early Anglo Saxon (*n* = 22); Late Anglo-Saxon (*n* = 10); Medieval (*n* = 26); Modern day (*n* = 12). The maxillary canine sample (*n* = 69) consisted of Roman (*n* = 11), Early Anglo Saxon (*n* = 20), Late Anglo-Saxon (*n* = 10), Medieval (*n* = 16) and Modern day (*n* = 12). Right teeth were selected unless they were unavailable or the left was better preserved.

6.2.2 Estimating sex

The modern day dental samples were all of known biological sex. The archaeological samples were assigned sex using established osteological methods of the skull and pelvis, utilising a 1-5 scale (1 = definitely female; 2 = likely female; 3 = indeterminate; 4 likely male; 5 = definitely male; see Appendix B) (Phenice, 1969; Ferembach, 1980; Krogman and Iscan, 1986; Schwartz, 1995; Loth and Henneberg, 1996; Patriquin et al., 2005). Sex assessment using the skull involved assessing 25 features known to be sexually dimorphic (as defined by: Ferembach, 1980; Krogman and Iscan, 1986; Schwartz, 1995; Loth & Henneberg, 1996). Assessment of the pelvis involved analysing a further 20 sexually dimorphic skeletal features (as defined by: Phenice, 1969; Ferembach, 1980; Krogman and Iscan, 1986; Schwartz, 1995) were also analysed. In addition, where the pelvis was not fragmented metric analyses were also used to give a sex determination score (Patriquin et al., 2005). Once all viable features of the skull and pelvis had been assigned a 1-5 score, all scores for an individual were

given an average which equated to the overall sex assessment. Individuals with a clear sex determination (i.e. not indeterminate) were then used for further analyses. Where possible all sex assessment methods were utilised, however in some cases methods could not be used due to the preservation of skeletal remains. For this reason only individuals with at least well preserved cranial or pelvic features were utilised.

6.2.3 Daily secretion rates

Using standard methods, the DSRs for both the upper first incisors and upper canine were calculated for the inner, mid, and outer areas of the lateral and cuspal enamel of each tooth (Fig. 6.1) (e.g. Beynon et al., 1991a; Schwartz et al., 2001; Mahoney, 2008). Please note that the angle of prism pathways vary slightly between anterior tooth types and molars. As inner, mid, and outer areas of enamel cap regions are defined by these pathways they are located slightly differently in dental cross sections; as a result differences can be noted between Figures 5.1 and 6.1. All cross striation measurements were taken between 20x and 40x magnification (see Fig. 6.2).

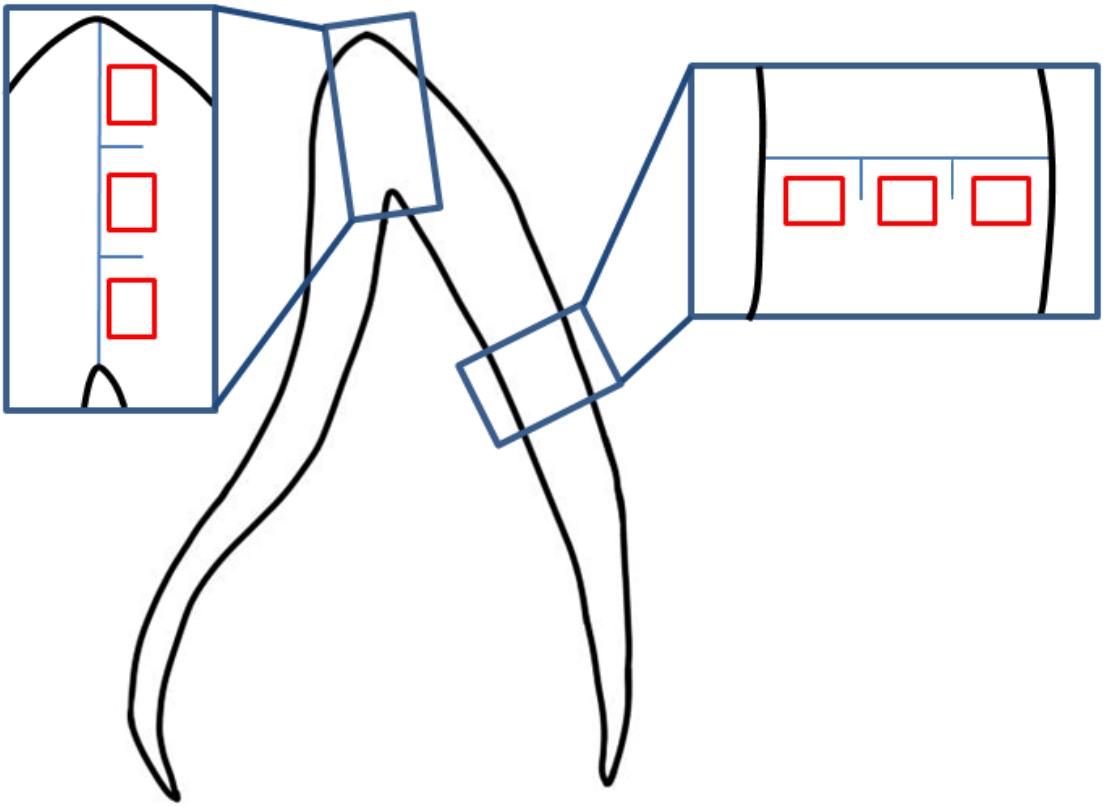

Fig. 6.1. *Cross-sectional diagram of an upper first incisor displaying the breakdown of cuspal and lateral enamel into areas for DSR calculation. The* ***left*** *superimposition shows the cuspal enamel. The* ***right*** *superimposition shows lateral enamel. The red squares indicate the regions where DSR measurements were taken for the, moving upwards, inner, mid, and outer areas.*

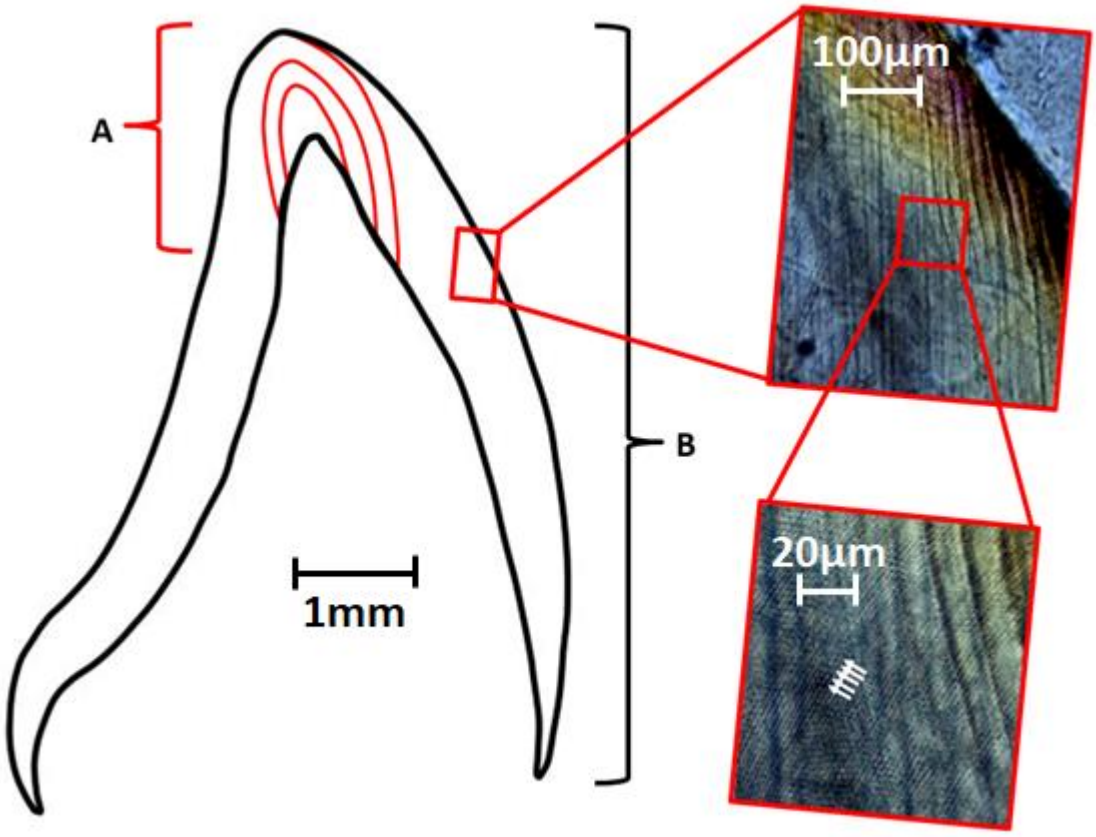

Fig. 6.2. *Cross-sectional diagram of an upper canine.* ***A****. Appositional enamel and* ***B****. Imbricational enamel. The* ***top*** *superimposition shows the mid-outer lateral region. The* ***bottom*** *superimposition shows cross striations, indicated by the small white arrows, of the outer lateral region. Both images were captured at 20x magnification under polarised light.*

6.2.4 Statistical analysis

Mann-Whitney U tests were run to identify any differences between the sexes in regional DSRs of upper first incisors and upper canines. Where regional DSRs presented adequate sample sizes, and were consistent between tooth types in all populations, they were pooled into a single upper anterior tooth sample set for subsequent analyses. A series of Independent Samples T-tests were then conducted to test for differences in DSRs between the male and female groups across all populations. Further Mann-Whitney U tests were then conducted to search for differences between males and females within each of the five populations separately.

Kruskal-Wallis tests with Dunn-Bonferroni pairwise comparisons and Jonckheere-Terpstra tests (for the majority of DSRs), and a series of Mann-Whitney U tests (for cuspal DSRs), one each for the male and female samples, were conducted to compare DSRs between the populations. This was

undertaken to identify significant in DSRs in males when compared between the time periods, and in females when compared between the same periods. In the few cases where $n<5$ for a given sample (where n = number of teeth), mean values and standard deviations were compared between groups.

While non-parametric tests were required in most cases, parametric tests were conducted where sample sizes allowed in order to strengthen the statistical analyses where possible. All statistical analyses were performed using SPSS 24.0.

6.3 Results

6.3.1 Differences between tooth types

Results of the Mann-Whitney U tests (see Table 6.1) revealed DSRs from equivalent enamel regions did not differ significantly when compared between upper first incisors and upper canines within any of the British populations. As a result, the data for both tooth types were pooled to create DSRs from anterior teeth for each population. These DSRs from anterior teeth were used for all subsequent statistical analyses.

Table 6.1. *Results of the Mann-Whitney U tests for variation between the upper first incisor and upper canine regional DSRs for each population.*

Population	Feature	Tooth (upper)	N	Mean	S.D	Sig.
Roman	Inner Lateral DSR	Canine	11	3.46	0.21	0.61
		Incisor	9	3.86	0.23	
	Mid Lateral DSR	Canine	11	3.97	0.20	0.49
		Incisor	10	4.08	0.13	
	Outer Lateral DSR	Canine	11	4.41	0.24	0.34
		Incisor	10	4.50	0.16	
	Inner Cuspal DSR	Canine	9	3.52	0.21	0.30
		Incisor	9	3.78	0.23	
	Mid Cuspal DSR	Canine	9	4.17	0.27	0.27
		Incisor	10	4.12	0.11	
	Outer Cuspal DSR	Canine	9	4.65	0.26	0.11
		Incisor	10	4.79	0.12	
Early Anglo-Saxons	Inner Lateral DSR	Canine	20	3.26	0.19	0.36
		Incisor	22	3.26	0.21	
	Mid Lateral DSR	Canine	20	3.83	0.20	0.26
		Incisor	22	3.77	0.26	
	Outer Lateral DSR	Canine	19	4.28	0.21	0.96
		Incisor	20	4.29	0.28	
	Inner Cuspal DSR	Canine	16	3.27	0.14	1.00
		Incisor	9	3.31	0.20	
	Mid Cuspal DSR	Canine	15	3.86	0.20	0.65
		Incisor	9	3.89	0.28	
	Outer Cuspal DSR	Canine	13	4.46	0.38	0.69
		Incisor	8	4.47	0.22	
Late Anglo-Saxons	Inner Lateral DSR	Canine	10	3.40	0.18	0.34
		Incisor	10	3.58	0.11	
	Mid Lateral DSR	Canine	10	3.86	0.19	0.25
		Incisor	10	4.00	0.25	
	Outer Lateral DSR	Canine	10	4.12	0.15	0.39
		Incisor	10	4.36	0.23	
	Inner Cuspal DSR	Canine	9	3.51	0.21	0.20
		Incisor	8	3.34	0.17	
	Mid Cuspal DSR	Canine	8	3.94	0.15	0.14
		Incisor	9	4.07	0.26	
	Outer Cuspal DSR	Canine	6	4.35	0.16	0.35
		Incisor	7	4.49	0.22	
Medieval	Inner Lateral DSR	Canine	14	3.11	0.26	0.57
		Incisor	25	3.13	0.22	
	Mid Lateral DSR	Canine	15	3.53	0.27	0.93
		Incisor	25	3.54	0.27	
	Outer Lateral DSR	Canine	15	3.91	0.24	0.47
		Incisor	25	3.88	0.24	
	Inner Cuspal DSR	Canine	11	3.12	0.16	0.90
		Incisor	16	3.13	0.18	
	Mid Cuspal DSR	Canine	12	3.70	0.20	0.96
		Incisor	20	3.73	0.22	
	Outer Cuspal DSR	Canine	9	4.10	0.25	0.24
		Incisor	19	4.01	0.28	
Modern day	Inner Lateral DSR	Canine	12	2.85	0.25	0.80
		Incisor	12	3.04	0.21	
	Mid Lateral DSR	Canine	12	3.30	0.21	0.27
		Incisor	12	3.49	0.27	
	Outer Lateral DSR	Canine	11	3.60	0.23	0.15
		Incisor	12	3.72	0.25	
	Inner Cuspal DSR	Canine	10	2.90	0.29	0.93
		Incisor	8	3.20	0.23	
	Mid Cuspal DSR	Canine	10	3.18	0.22	0.21
		Incisor	8	3.54	0.22	
	Outer Cuspal DSR	Canine	10	3.57	0.19	0.18
		Incisor	8	3.89	0.23	

6.3.2 Differences in DSRs between biological sex groups

Table 6.2 reports the results of DSRs compared between biological male and females when all of the British populations were pooled. Independent Samples T-tests revealed no significant differences in DSRs when compared between males and females. Table 6.3 reports the same tests conducted separately for each population. There was no significant difference between the sexes when DSRs from equivalent regions were compared within each population.

Table 6.2. *Results of the Independent Samples T-tests for variation in DSRs when the sexes were pooled for all populations.*

Feature	Sex	N	Mean	S.D	Min	Max	Sig.
Inner Lateral DSR	M	44	3.16	0.33	2.47	4.11	0.19
	F	39	3.31	0.32	2.45	4.24	
Mid Lateral DSR	M	44	3.65	0.33	2.99	4.42	0.13
	F	41	3.80	0.31	2,86	4.29	
Outer Lateral DSR	M	42	4.05	0.37	3.35	4.75	0.11
	F	40	4.15	0.40	3.03	4.81	
Inner Cuspal DSR	M	32	3.24	0.33	2.41	4.16	0.69
	F	32	3.26	0.32	2.51	3.26	
Mid Cuspal DSR	M	33	3.72	0.38	2.91	4.58	0.22
	F	33	3.81	0.31	2.86	4.29	
Outer Cuspal DSR	M	27	4.17	0.50	3.42	5.05	0.25
	F	31	4.31	0.48	3.16	5.37	

Table 6.3. *Results of the Mann-Whitney U tests for variation in DSRs compared between the sexes for the upper anterior tooth sample of each population.*

Population	Feature	Sex	N	Mean	S.D	Min	Max	Sig.
Roman	Inner Lateral DSR	M	8	3.62	0.22	3.35	4.11	0.79
		F	8	3.58	0.36	3.12	4.24	
	Mid Lateral DSR	M	8	4.04	0.18	3.67	4.28	0.84
		F	9	4.04	0.18	3.68	4.29	
	Outer Lateral DSR	M	8	4.48	0.17	4.28	4.75	0.77
		F	9	4.41	0.25	3.88	4.81	
	Inner Cuspal DSR	M	7	3.65	0.28	3.35	4.16	0.33
		F	7	3.57	0.38	3.22	4.23	
	Mid Cuspal DSR	M	8	4.17	0.21	3.94	4.58	0.90
		F	7	4.13	0.23	3.67	4.35	
	Outer Cuspal DSR	M	8	4.74	0.28	4.16	5.05	0.52
		F	7	4.72	0.12	4.53	4.92	
Early Anglo-Saxons	Inner Lateral DSR	M	9	3.18	0.15	3.05	3.43	0.60
		F	13	3.39	0.13	3.11	3.62	
	Mid Lateral DSR	M	9	3.76	0.29	3.36	4.42	0.90
		F	13	3.96	0.19	3.48	4.17	
	Outer Lateral DSR	M	8	4.33	0.22	3.94	4.74	0.28
		F	12	4.40	0.19	3.96	4.69	
	Inner Cuspal DSR	M	5	3.25	0.15	3.05	3.43	0.35
		F	9	3.32	0.19	2.85	3.32	
	Mid Cuspal DSR	M	5	3.75	0.25	3.47	3.99	0.38
		F	9	3.92	0.19	3.38	4.29	
Medieval	Inner Lateral DSR	M	7	2.96	0.19	2.72	3.22	0.24
		F	5	3.18	0.16	2.97	3.43	
	Mid Lateral DSR	M	7	3.53	0.14	3.35	3.74	0.28
		F	6	3.64	0.18	3.42	3.91	
	Outer Lateral DSR	M	7	3.84	0.18	3.61	4.11	0.15
		F	6	4.04	0.28	3.60	4.49	
	Inner Cuspal DSR	M	7	3.10	0.12	2.91	3.29	0.56
		F	5	3.09	0.20	2.94	3.45	
	Mid Cuspal DSR	M	7	3.71	0.08	3.62	3.85	0.11
		F	6	3.81	0.26	3.34	4.09	
Modern day	Inner Lateral DSR	M	14	2.92	0.23	2.47	3.22	0.53
		F	10	2.99	0.28	2.45	3.32	
	Mid Lateral DSR	M	14	3.32	0.19	2.99	3.65	0.10
		F	10	3.46	0.31	2.86	3.80	
	Outer Lateral DSR	M	13	3.66	0.17	3.35	3.84	0.55
		F	10	3.66	0.33	3.03	4.06	
	Inner Cuspal DSR	M	9	3.00	0.32	2.41	3.34	0.86
		F	9	3.06	0.30	2.51	3.43	
	Mid Cuspal DSR	M	9	3.25	0.21	2.91	3.59	0.15
		F	9	3.41	0.33	2.81	3.86	
	Outer Cuspal DSR	M	9	3.66	0.17	3.42	3.95	0.25
		F	9	3.77	0.32	3.16	4.09	

6.3.3 Differences in DSRs between biological sex groups and between populations

Tables 6.4 and 6.5 report mean inner, mid, and outer DSRs for cuspal and lateral regions (respectively) of the upper anterior tooth samples from the male individuals of each population. In addition the tables include the descriptive data and results of the Kruskal Wallis and post-hoc Dunn-Bonferroni pairwise comparisons.

Mean DSRs from the inner cuspal enamel region of male anterior teeth were significantly slower in the modern day sample compared to the Roman sample. The inner region mean DSRs of the male cuspal upper anterior tooth enamel slowed between the Roman and modern day samples by 0.40 µm/day. This slowing displayed a followed a significant trend through time (p<0.001). The Roman mean DSRs were significantly faster than that of the Medieval and modern day populations (p<0.001). The mean DSRs of the mid cuspal male upper anterior tooth enamel between the Roman and modern day samples slowed by a rate of 0.92 µm/day. The trend towards slowing for the enamel region was also significant (p<0.001). In addition, the Roman mean DSRs were significantly faster than that of the modern day (p<0.001) population. The mean outer cuspal male upper anterior tooth DSRs also alluded to a slowing in secretions rates over time between populations. The mean Roman DSRs were significantly faster than the modern day population (p<0.001), with a mean difference of 1.08 µm/day.

The inner region mean DSRs of the lateral upper anterior tooth enamel slowed between the Roman and modern day samples by 0.70 µm/day, and displayed a significant slowing trend through time (p<0.001). The Roman mean DSRs were significantly faster than that of the Medieval and modern day (both at p<0.001) populations. The mean DSRs of the mid lateral upper anterior tooth enamel between the Roman and modern day samples slowed by a rate of 0.72 µm/day, and with a significant trend towards slowing (p<0.001). The Roman and Early Anglo-Saxon mean DSRs were significantly faster than that of the modern day (p<0.001 and p = 0.01 respectively) population. The mean Roman DSRs were also significantly faster than in the Medieval (p = 0.03) population. The mean DSRs of the outer lateral upper anterior tooth enamel between the Roman and modern day samples slowed by an increased rate of 0.82 µm/day. The region also displayed a significant trend towards slowing (p<0.001). The Roman and Early Anglo-Saxon mean DSRs were significantly faster than that of the modern day (both at p<0.001) population. The mean Roman DSRs were also significantly faster than in the Medieval (p<0.001) population.

Table 6.4. *Results of the Mann-Whitney U and independent samples Kruskal-Wallis tests with post-hoc pairwise Dunn-Bonferroni analysis, and Jonckheere-Terpstra testing for variation between the* ***male cuspal*** *DSRs (µm/day) across all populations. Mann-Whitney U tests were used in the case of the outer enamel data as only two populations supported statistical analysis (as other samples were ≥n=5). Where populations presented sample sizes too small for statistical analysis they are still presented here for future comparisons. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. The negative Jonckheere-Terpstra result shows the trend towards a reduction in DSRs. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	S.D	Min	Max	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.			Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Medieval	Modern day	
Inner	Roman	7	3.65	0.28	3.35	4.16	**0.01**	0.45	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	5	3.25	0.15	3.05	3.43					
									0.51	0.43	
	Medieval	7	3.10	0.12	2.91	3.29				1.00	
	Modern day	9	3.00	0.32	2.41	3.34					
Mid	Roman	8	4.17	0.21	3.94	4.58	**0.00***	0.52	0.07	**0.00***	**-0.00***
	Early Anglo-Saxon	5	3.75	0.25	3.47	3.99					
									1.00	0.11	
	Medieval	7	3.71	0.08	3.62	3.85				0.32	
	Modern day	9	3.25	0.21	2.91	3.59					
							Mann-Whitney U Sig.				
Outer	Roman	8	4.74	0.28	4.16	5.05	**0.00***				
	Modern day	9	3.66	0.17	3.42	3.95					
	Early Anglo-Saxon	4	4.35	0.33	3.99	4.64					
	Medieval	4	3.88	0.09	3.77	4.01					

*p<0.001

Table 6.5. *Results of the independent samples Kruskal-Wallis, post-hoc pairwise Dunn-Bonferroni analysis, and Jonckheere-Terpstra testing for variation between the **male lateral** DSRs (µm/day) across all populations. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. The negative Jonckheere-Terpstra result shows the trend towards a reduction in DSRs. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	S.D	Min	Max	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.			Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Medieval	Modern day	
Inner	Roman	8	3.62	0.22	3.35	4.11	**0.00***	0.11	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	9	3.18	0.15	3.05	3.43			1.00	0.54	
	Medieval	7	2.96	0.19	2.72	3.22				1.00	
	Modern day	14	2.92	0.23	2.47	3.22					
Mid	Roman	8	4.04	0.18	3.67	4.28	**0.00***	1.00	**0.03**	**0.00***	**-0.00***
	Early Anglo-Saxon	9	3.76	0.29	3.36	4.42			1.00	**0.01**	
	Medieval	7	3.53	0.14	3.35	3.74				1.00	
	Modern day	14	3.32	0.19	2.99	3.65					
Outer	Roman	8	4.48	0.17	4.28	4.75	**0.00***	1.00	**0.00***	**0.00***	**-0.00***
	Early Anglo-Saxon	8	4.33	0.22	3.94	4.74			0.09	**0.00***	
	Medieval	7	3.84	0.18	3.61	4.11				1.00	
	Modern day	13	3.66	0.17	3.35	3.84					

*p<0.001

Tables 6.5 and 6.6 report the mean inner, mid, and outer DSRs for the cuspal and lateral region (respectively) of the upper anterior tooth samples compromised of the female individuals of each population. In addition the tables include the descriptive data and results of the Kruskal Wallis and post-hoc Dunn-Bonferroni pairwise comparisons.

The inner region mean DSRs of the female cuspal upper anterior tooth enamel slowed by a rate of 0.51 µm/day between the Roman and modern day samples. Pairwise analyses found the mean Roman DSRs to be significantly faster than that of the modern day (p = 0.05), and the trend towards slowing through time was also significant (p<-0.001). The mean DSRs of the mid cuspal female upper anterior tooth enamel between the Roman and modern day samples slowed by a rate of 0.72 µm/day and the trend towards slowing DSRs was significant (p<0.001). In addition, the Roman mean DSRs were significantly faster than that of the modern day (p<0.001) population, with a mean difference of 0.95 µm/day. The mean outer cuspal DSRs also slowed between populations, at a rate of 0.95 µm/day between the Roman and modern day populations. The trend towards slowing was again significant (p<-0.001). Male modern day mean outer cuspal DSRs were also significantly slower than those of both the Roman and Early Anglo-Saxon populations (both at p<0.001).

The inner region mean DSRs of the lateral upper anterior tooth enamel slowed between the Roman and modern day samples by 0.59 µm/day. Changes in DSRs between populations through time, again followed a significant slowing trend (p<0.001). The Roman and Early Anglo-Saxon mean DSRs were significantly faster than that of the modern day (p<0.001 and p = 0.01 respectively) population. The mean DSRs of the mid lateral upper anterior tooth enamel between the Roman and modern day samples slowed by a similar rate of 0.58 µm/day, and with a significant trend towards slowing (p<0.001). The Roman and Early Anglo-Saxon mean DSRs were significantly faster than that of the modern day (p<0.001 and p = 0.01 respectively) population. The mean Roman DSRs were also significant faster than in the Medieval (p = 0.02) population. The mean DSRs of the outer lateral upper anterior tooth enamel between the Roman and modern day samples slowed by an accelerated rate of 0.74 µm/day, and displayed a significant trend towards slowing (p<0.001). The Roman and Early Anglo-Saxon mean DSRs were again significantly faster than that of the modern day (both at p<0.001) population.

Table 6.6. *Results of the independent samples Kruskal-Wallis tests with post-hoc pairwise Dunn-Bonferroni analysis, and Jonckheere-Terpstra testing for variation between the* ***female cuspal*** *DSRs (µm/day) across all populations. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. The negative Jonckheere-Terpstra result shows the trend towards a reduction in DSRs. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	S.D	Min	Max	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.			Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Medieval	Modern day	
Inner	Roman	7	3.57	0.38	3.22	4.23	**0.04**	1.00	0.19	**0.05**	**-0.00***
	Early Anglo-Saxon	9	3.32	0.19	2.85	3.32			1.00	0.64	
	Medieval	5	3.09	0.20	2.94	3.45				1.00	
	Modern day	9	3.06	0.30	2.51	3.43					
Mid	Roman	7	4.13	0.23	3.67	4.35	**0.00***	1.00	0.89	**0.00***	**-0.00***
	Early Anglo-Saxon	9	3.92	0.19	3.38	4.29			1.00	0.07	
	Medieval	6	3.81	0.26	3.34	4.09				0.67	
	Modern day	9	3.41	0.33	2.81	3.86					
Outer	Roman	7	4.72	0.12	4.53	4.92	**0.00***	1.00	0.31	**0.00***	**-0.00***
	Early Anglo-Saxon	12	4.40	0.35	3.96	5.37			1.00	**0.00***	
	Medieval	6	4.26	0.16	4.08	4.41				0.65	
	Modern day	9	3.77	0.32	3.16	4.09					

*p<0.001

Table 6.7. *Results of the independent samples Kruskal-Wallis, post-hoc pairwise Dunn-Bonferroni analysis, and Jonckheere-Terpstra testing for variation between the **female lateral** DSRs (µm/day) across all populations. Mann-Whitney U tests were used where sample sizes only supported the analysis of two populations. The Dunn-Bonferroni significance values have been adjusted to account for Bonferroni corrections. The negative Jonckheere-Terpstra result shows the trend towards a reduction in DSRs. Significant results are marked in bold.*

Enamel Region	Population	N	Mean	S.D	Min	Max	Kruskal-Wallis Sig.	Dunn-Bonferroni Adjusted Sig.			Jonckheere-Terpstra Sig.
								Early Anglo-Saxon	Medieval	Modern day	
Inner	Roman	8	3.58	0.36	3.12	4.24	**0.00***	1.00	0.17	**0.00***	**-0.00***
	Early Anglo-Saxon	13	3.39	0.13	3.11	3.62			0.73	**0.01**	
	Medieval	5	3.18	0.16	2.97	3.43				1.00	
	Modern day	10	2.99	0.28	2.45	3.32					
Mid	Roman	9	4.04	0.18	3.68	4.29	**0.00***	1.00	**0.02**	**0.00***	**-0.00***
	Early Anglo-Saxon	13	3.96	0.19	3.48	4.17			0.07	**0.00***	
	Medieval	6	3.64	0.18	3.42	3.91				1.00	
	Modern day	10	3.46	0.31	2.86	3.80					
Outer	Roman	9	4.41	0.25	3.88	4.81	**0.00***	1.00	0.47	**0.00***	**-0.00***
	Early Anglo-Saxon	12	4.40	0.19	3.96	4.69			0.42	**0.00***	
	Medieval	6	4.04	0.28	3.60	4.49				1.00	
	Modern day	10	3.66	0.33	3.03	4.06					

*p<0.001

6.4 Discussion

This study compared DSRs of anterior teeth between biological male and females, from modern and archaeological populations from Britain. These comparisons revealed no significant difference in DSRs when males were compared to females, either when British populations were combined into a single sample, or within each population. However, there was a significant trend towards a slowing of DSRs across the 2000 year period, from the Romano-British to the modern day populations, for males, and for females. There were a greater number of significant differences between the populations when males were compared, in comparison to the number of significant differences observed in females.

6.4.1 Daily secretion rates compared between the sexes within each populations

There was no significant difference between the anterior teeth for males and females in this study. This analysis reveals that the daily enamel growth of permanent human upper anterior tooth enamel is consistent between the sexes, within these ancient and modern British populations. Our findings for permanent upper anterior tooth enamel DSRs are consistent with findings for DSRs from permanent canines from a single human population (Schwartz et al., 2001).

6.4.2 Daily secretion rates compared between the sexes, from ancient to modern populations

While all regional DSRs from male and female groups slowed over time (see Tables 6.5, 6.6 and 6.7), the male samples displayed a higher volume of significant differences when compared between populations. Cuspal enamel analyses revealed four such variations, whereas the equivalent analyses of the female samples identified only two pairwise significant differences. Analyses of lateral enamel DSRs displayed a similar trend with eight significant differences between the enamel of male groups, with slightly fewer significant differences in the female groups.

6.4.3 Daily secretion rates between populations

Pairwise analysis of both male and female samples found a number of significant inter-population differences in DSRs of cuspal and lateral upper anterior tooth enamel. All these differences display an additional significant trend towards a slowing trajectory in DSRs between populations from the Roman to modern day populations. Indeed, only in the single case of the female inner cuspal DSRs

were the mean Roman values not significantly faster than the modern day (p = 0.07; see Table 6.6). While the male sample displayed the most pairwise significant differences between populations, the trends towards slowing were consistent for all enamel regions across both male and female analyses (see Tables 6.5, 6.6, and 6.7). However, the five British populations presented here represent a 2000 year period of history, and each possessed unique differentiating cultural practices. These factors could have potentially caused enamel growth to significantly slow over the last 2000 years in Britain. However, additional research would be required to test this theory, and how this might explain the variation between the inter-population differences in male and female groups.

6.4.4 Daily secretion rates compared to posterior teeth

Past research utilising the same British populations, has identified a significant trend towards slowing enamel DSRs from the Roman to modern period in permanent first molar teeth (Aris et al., 2020b). This trend is similar to those observed here in both male and female samples. These results show the trend towards the slowing of daily enamel growth in Britain over the last 2000 years, has been consistent in both anterior and posterior teeth. Where the differences in enamel growth rates between populations were similar in the anterior teeth and first molars of the British populations, this was not the case specifically in differences between the Roman and Early Anglo-Saxon populations. In their study, Aris and colleagues (2020b) commented on the similarities in the growth rates of the two populations, most notably in the cuspal enamel where mean rates were near identical. Conversely, in almost all upper anterior tooth enamel regions presented here, both male and female DSRs can be observed to vary between the Roman and Early Anglo-Saxon population. Only in the mid and outer lateral enamel of the female sample can comparable similarities to that seen in the first molar teeth of the sample populations (Aris et al., 2020b) be observed, between the Roman and Early Anglo-Saxon populations. These differences in findings between upper anterior teeth and first molar growth rates suggest that, within a consistent trend of slowing overtime, variation between tooth types has also occurred. This further suggests that no single tooth type should necessarily be considered representative of the dental arcade when investigating differences in enamel growth rates between human populations. Future analysis of premolar enamel would be valuable.

Comparison of DSRs for equivalent enamel regions and British populations between tooth types identifies further variation growth patterns. Comparing mean regional DSRs calculated from male and female upper anterior tooth samples (presented here) to mean DSRs for first molar regions

(see: Tables 2 and 3 in Aris et al., 2020b; alternatively see Tables 5.6 and 5.7 in *Chapter 5*) shows permanent first molar enamel, in the majority of 24 comparisons, to have been secreted at a faster rate than that of anterior teeth. In 11 of these cases first molar regions grew faster, but only by a rate of ≤0.15 µm/day. In four of the seven cases where first molar enamel secreted at a slower rate, the difference in DSRs between first molar and upper anterior teeth was also only ≤0.15 µm/day. Interestingly these were almost always in outer region DSRs. In the remaining six cases of the initial 24 comparisons, more notable differences were seen between the regional DRSs of first molar and upper anterior teeth. One difference was isolated to a single case of the Early Anglo-Saxon population in which inner cuspal first molar enamel was secreted at a rate of 0.40 µm/day faster than that of the anterior teeth. In another single case, the mid lateral upper anterior tooth enamel of the modern-day population grew at a faster rate by 0.23 µm/day. For the remaining four cases where notable differences between tooth types were observed, all were within comparisons of the medieval population. In the inner lateral and cuspal regions Medieval first molar enamel secreted at a faster rate (by 0.29 and 0.26 µm/day respectively), where conversely the outer lateral and cuspal regions Medieval upper anterior tooth enamel secreted faster (by 0.23 and 0.40 µm/day respectively). While preliminary, the findings of comparing upper anterior tooth and first molar enamel DSRs within British populations does allude to variable growth patterns between tooth types. Overall, permanent first molar enamel appears to develop faster, particularly in the inner and mid regions. Conversely, upper anterior tooth types can develop faster in the outer regions, most notably the cuspal outer regions. The cause for this difference is as of yet unknown, but appears to be most active in the Medieval British population.

The discovery of variation in permanent enamel DSRs, both between tooth types within populations and within tooth types between populations, provides further evidence to the idea that permanent enamel DSRs are highly variable in humans, even over as short a period as 2000 years. Furthermore, the reasons underlying the slower DSRs from the more ancient to modern period have probably influenced the anterior and posterior teeth, as both of these tooth types show a similar trend.

6.4.5 Conclusions

Results presented here display a consistency in DSRs when compared between biologically male and female groups, in both archaeological and modern British populations. In contrast to these findings, DSRs have varied to a greater degree between British populations. Daily secretion rates, from cuspal

and lateral enamel regions, were observed to have significantly slowed throughout the last 2000 years in Britain. This pattern is consistent to that previously observed for DSRs in first molar enamel. Future research would benefit from integrating life history, genetic, and environmental factors in order to widen our understanding of how diversity in human enamel growth has, and may continue to, evolve.

CHAPTER 7: DISCUSSION

7.1 Enamel growth variation across the tooth row and between groups

7.1.1 Roman DSRs

In all cases except one, the mean upper canine DSRs were slower compared to equivalent regions in the upper first incisors and first molars in the Roman sample. Mean DSRs showed only slight variation between the upper first incisors and first molars when regions were compared. Differences were largest between the first molars and upper first incisors in the outer cuspal and inner lateral regions (see Table 7.1). Mean outer cuspal upper first incisor DSRs were faster by a rate 0.23µm/day compared to the same in first molars. Conversely, mean inner lateral first molar DSRs were faster by a rate of 0.20µm/day compared to the same in upper first incisors. Overall, the permanent enamel growth of the upper canines was slower than that of first molars and upper first incisors.

The differences between tooth types agrees with the comments in the discussion of Chapter 6 (Aris et al., 2020a) that variation in regional mean DSRs between tooth types has occurred. However, in Chapter 6 first molar DSRs were compared to a pooled upper anterior tooth DSRs sample. As the comparisons here consider all tooth types, the differences observed show a further level of variation in enamel DSRs not previously noted.

Table 7.1. *Regional mean DSRs (±SD) for all tooth types of the Roman population.*

Cuspal DSRs Mean (µm/day)				
Tooth	N	Region		
		Inner	Mid	Outer
First molars	7	3.72 ± 0.26	4.21 ± 0.42	4.56 ± 0.22
Upper canines	9	3.52 ± 0.21	4.12 ± 0.27	4.56 ± 0.26
Upper first incisors	10	3.78 ± 0.23	4.17 ± 0.11	4.79 ± 0.12
Lateral DSRs Mean (µm/day)				
First molars	11	3.66 ± 0.28	4.19 ± 0.33	4.57 ± 0.34
Upper canines	11	3.46 ± 0.21	3.97 ± 0.20	4.41 ± 0.24
Upper first incisors	9-10	3.86 ± 0.23	4.08 ± 0.13	4.50 ± 0.16

7.1.2 Early Anglo-Saxon DSRs

The first molars presented faster mean DSRs than the equivalent regions in the upper anterior teeth for the inner and mid regions of both cuspal and lateral enamel. The upper canines presented the slowest mean DSRs for the cuspal inner and mid regions. The difference between the upper canine and first molar mean DSRs was most notable in the inner regions, where upper canines were slower by a rate of 0.42µm/day in cuspal enamel and 0.16µm/day in the lateral enamel. The mean DSRs for the inner and mid regions of lateral enamel were slowest in upper first incisors. Conversely, the first molars presented the slowest mean DSRs for the outer regions of both cuspal and lateral enamel (although by slim margins of 0.03µm/day and 0.04µm/day respectively; see Table 7.2).

In the Early Anglo-Saxon population, variation in mean DSRs differed between tooth types when compared to that previously seen in the Roman population, particularly in the slower growing upper first incisors. No past research has yet investigated for variation in enamel DSRs between tooth types, and whether this could itself vary between populations. As a result, the data for the Early Anglo-Saxon population shows an additional new trend in human enamel growth, while displaying additional inter-population differences to those previously observed in Chapters 5 and 6 (Aris et al., 2020a, 2020b).

Table 7.2. *Regional mean DSRs (±SD) for all tooth types of the Early Anglo-Saxon population.*

Cuspal DSRs Mean (µm/day)				
Tooth	N	Region		
		Inner	Mid	Outer
First molars	6-7	3.69 ± 0.19	3.98 ± 0.25	4.43 ± 0.29
Upper canines	13-20	3.27 ± 0.14	3.86 ± 0.20	4.46 ± 0.38
Upper first incisors	8-9	3.31 ± 0.20	3.89 ± 0.28	4.47 ± 0.22
Lateral DSRs Mean (µm/day)				
First molars	12	3.42 ± 0.20	3.85 ± 0.20	4.24 ± 0.21
Upper canines	19-20	3.26 ± 0.19	3.83 ± 0.20	4.28 ± 0.21
Upper first incisors	20-22	3.26 ± 0.21	3.77 ± 0.26	4.29 ± 0.28

7.1.3 Late Anglo-Saxon DSRs

Within the cuspal regions the upper anterior teeth displayed the fastest growing enamel. This was fastest in the upper first incisors for the mid and outer regions, and upper canines in the inner region (see Table 7.3). Cuspal first molar enamel was therefore the slowest, although first molars were equal slowest with upper first incisors in the cuspal inner region.

The lateral region DSRs showed more consistent differences between tooth types. Lateral upper first incisor enamel was the fastest growing enamel between the tooth types for all regions. Lateral first molar enamel was the slowest growing of all tooth types analysed. Differences between the tooth types was most notable in the outer lateral region, where the first molars grew slower by a mean rate of 0.24µm/day compared to upper canines, and 0.48µm/day compared to upper first incisors.

The variations between the Late Anglo-Saxon tooth types are more similar to those seen in the Romans, and are thereby also different to those of the Early Anglo-Saxon population. In Chapter 6 sample sizes did not permit for observations to be made between posterior and upper anterior tooth types (Aris et al., 2020a). Therefore, the observations made here are novel, and provide further evidence for the newly discovered high level of variation which exists within human enamel growth, both between populations and tooth types.

Table 7.3. *Regional mean DSRs (±SD) for all tooth types of the Late Anglo-Saxon population.*

Cuspal DSRs Mean (µm/day)				
Tooth	N	Region		
		Inner	Mid	Outer
First molars	17-18	3.34 ± 0.27	3.65 ± 0.32	3.99 ± 0.32
Upper canines	6-9	3.51 ± 0.21	3.94 ± 0.15	4.35 ± 0.16
Upper first incisors	7-9	3.34 ± 0.17	4.07 ± 0.26	4.49 ± 0.22
Lateral DSRs Mean (µm/day)				
First molars	19	3.36 ± 0.25	3.63 ± 0.33	3.88 ± 0.39
Upper canines	10	3.40 ± 0.18	3.86 ± 0.19	4.12 ± 0.25
Upper first incisors	10	3.58 ± 0.11	4.00 ± 0.25	4.36 ± 0.23

7.1.4 Medieval DSRs

The inner cuspal and lateral DSRs were fastest in the first molars. While slower, the upper anterior tooth types presented little difference between upper first incisors and upper canines, with mean DSRs for the inner cuspal and lateral regions only differing by rates of 0.01µm/day and 0.02µm/day (both faster in upper first incisors; see Table 7.4) respectively. Within the cuspal enamel specifically, the differences between upper canine and incisor DSRs were more varied in the mid and outer regions, but were both were faster than first molars. Within the lateral enamel specifically the differences between upper canine and incisor DSRs remained minimal in the mid and outer regions; with first molars presenting the fastest DSRs again for the mid region, but all teeth possessing similar DSRs for the outer region (all within a mean range of 0.03µm/day; see Table 7.4).

In Chapter 6 it was shown that the mean first molar DSRs of the Medieval population were faster in inner regions, and slower in outer regions, when compared to the pooled upper anterior tooth sample (Aris et al., 2020a). This finding holds here when observing inner enamel regions but is less obvious in the outer regions – particularly when observing the similarity between the mean DSRs of the outer regions of first molars and upper first incisors. While differences between tooth types are still evident in the data here, the results of the comparisons show that it is inaccurate to examine differences between tooth types when pooling anterior or posterior tooth types. Future research should therefore ensure individual tooth types are kept separate when comparing growth rates data between teeth.

Table 7.4. *Regional mean DSRs (±SD) for all tooth types of the Medieval population.*

Cuspal DSRs Mean (µm/day)				
Tooth	N	Region		
		Inner	Mid	Outer
First molars	23-27	3.36 ± 0.24	3.65 ± 0.28	3.90 ± 0.28
Upper canines	9-12	3.12 ± 0.26	3.70 ± 0.20	4.10 ± 0.25
Upper first incisors	16-20	3.13 ± 0.18	3.73 ± 0.22	4.01 ± 0.28
Lateral DSRs Mean (µm/day)				
First molars	27-29	3.36 ± 0.26	3.65 ± 0.29	3.89 ± 0.26
Upper canines	15	3.11 ± 0.26	3.53 ± 0.27	3.91 ± 0.24
Upper first incisors	25	3.13 ± 0.22	3.54 ± 0.27	3.88 ± 0.24

7.1.5 Modern-day DSRs

Mean DSRs for the first molar and upper first incisor tooth types were similar. In the outer cuspal enamel region, the upper first incisor DSRs were faster than the first molars by a mean rate of 0.14µm/day – this is the largest difference seen in mean DSRs between tooth types (see Table 7.5). Upper canine mean DSRs were the slowest for all regions of both enamel areas. When compared to first molars (the second fastest DSRs) the upper canine mean DSRs were slower by a rate ranging from 0.11µm/day in the outer lateral region, to 0.34µm/day in the mid cuspal regions.

In Chapter 6 it was observed that only in the mid lateral region did modern-day upper anterior tooth enamel grow faster by mean than first molars (Aris et al., 2020a). When upper anterior tooth types were separated here this was not the case. This suggests that the differences previously observed in Chapter 6 were the result of slow growing modern-day upper canines specifically. As a result, it is even more imperative that future research testing for differences between the growth rates between tooth types ensure that upper first incisors and upper canines are not pooled.

Table 7.5. *Regional mean DSRs (±SD) for all tooth types of the Modern-day population.*

Cuspal DSRs Mean (µm/day)				
Tooth	N	Region		
		Inner	Mid	Outer
First molars	14-15	3.20 ± 0.38	3.52 ± 0.35	3.75 ± 0.37
Upper canines	10	2.90 ± 0.29	3.18 ± 0.22	3.57 ± 0.19
Upper first incisors	8	3.20 ± 0.23	3.54 ± 0.22	3.89 ± 0.23
Lateral DSRs Mean (µm/day)				
First molars	15	3.14 ± 0.21	3.42 ± 0.32	3.71 ± 0.42
Upper canines	11-12	2.85 ± 0.25	3.30 ± 0.21	3.60 ± 0.23
Upper first incisors	12	3.04 ± 0.21	3.49 ± 0.27	3.72 ± 0.25

7.1.6 Enamel growth variations between the biological sexes

Chapter 6 investigated for differences between the biological sexes in regional mean DSRs (Aris et al., 2020a). No significant differences between biological males and females in regional mean DSRs were found when analyses tested a pooled British population, or within each individual populations. However, within the Early Anglo-Saxon population, the female sample had consistently faster mean DSRs than the male (by rates of: 0.21µm/day in the inner lateral region, 0.20µm/day in the mid lateral region, and 0.17µm/day in the mid and outer cuspal regions). There were only two other cases where DSRs differed between the sexes by a notable rate: in the outer lateral region of the Medieval population and the mid cuspal region of Modern-day samples (see Table 6.3). Sample sizes were too small to further analyse sex differences between first molars, but it is possible first molar enamel could present differences between males and females. Given the increased regional differences observed in the Early Anglo-Saxon population, a larger sample of first molar enamel would be ideal for future investigations into sex differences in human enamel growth across the tooth row.

7.2 First molar enamel thickness compared to known ranges

For this section please note that thickness data is presented from a pooled sample of upper and lower first molars. Relative enamel thickness was collected from the paracone and protocone cusps for the upper molars, and metaconid and protocone cusps for the lower molars. Linear measurements of CT and LT were collected from the paracone and protoconid.

7.2.1 Roman upper molar enamel thickness

Observation of the Roman first molar sample RET shows the greatest similarity to data published by Olejniczak and colleagues (2008), differing in mean by only 0.02 (see Table 7.6). This data represents a modern-day sample of maxillary first molars (different to that collected and analysed from in past chapters here) with mostly unknown provenance, but that which is thought to be derived from North American and European extractions (Olejniczak et al., 2008). This similarity is unexpected given the larger variation in first molar RET (by 2.22; see also Table 7.6) observed between the Roman population and Modern-day sample, newly analysed in this research. These differences further highlight the intraspecific variation which exists between human populations, first identified in here in Chapter 5.

The mean CT of the Roman population was thicker, on average, than all published human ranges by a degree of >0.20mm, except in the single case of the population studied by Schwartz (2000a). These samples were from a modern British population (Schwartz, 2000a), and had thicker mean CT (by 0.31mm; see Table 7.6). This difference is particularly interesting, as the Roman population presented thicker CT than the Modern-day population analysed here (by 0.19mm; see Table 7.6). This is a similar trend to that previously seen in the RET of the Roman and modern-day first molar samples. This not only alludes to wide variation in linear enamel thickness between populations, but further indicates that high levels of variation exist between modern-day populations. This concurs with comments in Chapter 5 that enamel thickness variations are not tightly related to difference in time periods (Aris et al., 2020b).

The mean LT of the Roman population is more comparable to other published human populations (Macho and Berner, 1993; Suwa and Kono, 2005; Mahoney, 2010). While the Roman mean values were thinner than all other known populations, this was only by a degree of 0.13mm compared to the thickest published mean (Suwa and Kono, 2005), and by 0.02mm to the previous thinnest published mean (Macho and Berner, 1993). These results show CT to be more variable between human populations, whereas LT remains more consistent.

7.2.2 Early Anglo-Saxon first molar enamel thickness

The mean first molar RET of the Early Anglo-Saxon population was notably higher than other published data, with a value 4.47 higher than data published by Olenjniczak et al (2008) (see Table 7.6). However, ranges by Smith and colleagues (2006a) includes mean maxillary first molar RET larger than the Early Anglo-Saxon population in this research. This variability in RET highlights the

different ranges of RET that exist between human populations. In Chapter 5, the high mean RET of the Early Anglo-Saxon population is attributed to possible shifts in diet resulting in thickening of enamel between populations over short periods of time, notably in response to highly abrasive diets (Aris et al., 2020b). This was concluded to be the result of their coastal location (Millard et al., 1969) and additional dietary research of the early Anglo-Saxon period (Littleton and Frohlich, 1993; Mays and Beavan, 2012). While similar trends in changing enamel thickness have been observed, these have compared populations over a larger span of 7000 years (Le Luyer et al., 2014). It is therefore plausible that the high RET of the Early Anglo-Saxon population shows that enamel thickness is plastic enough as to vary over far shorter periods of time. Unfortunately, no dietary data is available for the Early Anglo-Saxon population. Therefore, future research should endeavour to collect such data from the same or a similar population to test this theory.

The mean CT of the Early Anglo-Saxon population is similar to several different populations (Suwa and Kono, 2005; Reid and Dean, 2006; Mahoney, 2010), differing by only ≤±0.09mm. One publication in particular varied from the Early Anglo-Saxon data, Schwartz (2000a), where the mean CT of a modern-day population was 0.59mm thicker than in the Early Anglo-Saxon population. Conversely, the Modern-day sample presented here possesses thicker mean CT by only 0.09mm. This is considerably less difference than those noted in Schwartz's (2000a) population. This suggests that while changes in first molar CT have occurred between the Early Anglo-Saxon and modern periods, the cause for the changes is not necessarily strongly tied to the difference in time period.

The mean first molar LT values are more similar to the published values of other populations. Early Anglo-Saxon mean LT was only 0.08mm thinner than the thickest published mean (Suwa and Kono, 2005), and 0.03mm thicker than the thinnest published mean (Macho and Berner, 1993). These results agree with the conclusions of Chapter 6, and further show CT to be more variable between human populations where LT is relatively consistent.

7.2.3 Late Anglo-Saxon first molar enamel thickness

Relative enamel thickness mean from of the Late Anglo-Saxon first molar was similar to data published by Olejniczak and colleagues (2008), differing in mean by only 0.84 and 0.34 (see Table 7.6). As previously stated, Olejniczak and colleagues' (2008) data represents a modern-day sample of maxillary first molars thought to be extractions from North America and Europe. This is unusual given the larger difference of 3.03 (see Table 7.6) between the mean first molar RET of Late Anglo-Saxon and the British Modern-day population. These observations continue to highlight the

intraspecific variation which exists between human populations identified in here in Chapter 6, and the emerging variation observable within the modern period.

Mean CT of the Late Anglo-Saxon population shows little difference to the ranges published by other human populations (Suwa and Kono, 2005; Reid and Dean, 2006; Mahoney, 2010), differing to these by only ≤±0.05mm. Conversely, the data published by Schwartz (2000a) was 0.60mm thicker than the Late Anglo-Saxon mean (see Table 7.6). The Modern-day sample analysed here presented thicker CT, suggesting changes in first molar CT has occurred between the Anglo-Saxon period as a whole and the modern period. However, the CT data collected here was only 0.10mm thicker in the Modern-day population compared to the Late Anglo-Saxon population. This is again, far less of a difference than when the Late Anglo-Saxon population's CT is compared to that of Schwartz's (2000a) population.

The Late Anglo-Saxon population possessed similar mean first molar LT values to all other published values of human populations. Late Anglo-Saxon mean LT was only 0.03mm thinner than the thickest published mean (Suwa and Kono, 2005), and 0.08mm thicker than the thinnest published mean (Macho and Berner, 1993). These results build upon previous observations that suggest first molar CT is more variable between human populations, compared to more consistent LT between populations (Chapter 5; Aris et al., 2020b).

7.2.4 Medieval first molar enamel thickness

The mean RET of the Medieval first molar sample presented a similar mean value to the Late Anglo-Saxon, with a difference of only 0.09 (larger in the Medieval population). The Medieval RET was therefore also similar to the equivalent data published by Olejniczak and colleagues (2008), with differences of by only 0.75 and 0.43 (see Table 7.6). The variable differences between the Medieval sample, and the British Modern-day and Olejniczak and colleagues' modern population, are the same as previously observed in the Roman and Late Anglo-Saxon populations. These comparisons thereby highlight the variation which exists between human populations identified here, and previously in Chapter 5. Conversely, they also highlight the minimal changes which occurred in human first molar RET throughout the Late Anglo-Saxon to Medieval periods in Britain. This finding concurs with the comments of Chapter 5 that the time period between the two populations was potentially too short as to permit significant changes in enamel thickness.

The mean CT of the Medieval population were also similar to that presented by the Late Anglo-Saxon population, differing by only 0.01mm (thinner in Medieval). Mean medieval CT was also

similar to the equivalent values reported for other human populations (Reid and Dean, 2006; Mahoney, 2010; Suwa and Kono, 2005), differing to these by only ≤±0.05mm (see Table 7.6). The modern CT data published by Schwartz (2000a) was 0.59mm thicker in mean when compared to the Medieval population. The Modern-day sample analysed here also presented thicker mean CT, but only by 0.11mm. These differences alone could suggest similar changes occurred between the modern and Medieval periods, as between the Medieval and Anglo-Saxon periods. However, the variable difference between Schwartz's (2000a) modern-day sample, and the Modern-day population presented here, are notable. This variance concurs with comments in Chapter 5 which suggest that differences in enamel thickness between populations may not be tightly linked to difference in time period (Aris et al., 2020b).

The mean LT of the Medieval population was also similar to that of the Late Anglo-Saxon population, differing only in standard deviation. Mean Medieval LT was similar to values of all other published data. Medieval mean first molar LT was only 0.03mm thinner than the thickest published mean (Suwa and Kono, 2005), and 0.08mm thicker than the thinnest published mean (Macho and Berner, 1993). These results build upon previous observations, suggesting human first molar CT can be highly variable between populations, whereas LT remains more consistent.

7.2.5 Modern-day first molar enamel thickness

The mean RET for the Modern-day population is the second highest value ever reported for humans, with only the Early Anglo-Saxon population presenting higher values (by 2.28; see Table 7.6). In Chapter 5 this value is discussed within the context of changes over time, where little difference was observed between the British populations (Aris et al., 2020b). While those results do suggest little change within a temporal transect, a new highest mean RET for a human population is notable. The results presented here expand the previously known range of human molar RET.

The mean CT of the British Modern-day sample was thicker than all past populations, with the exception of the modern-day sample researched by Schwartz (2000a) which had a 0.50mm thicker mean. Schwartz's (2000a) population was of Slavic origin, suggesting the difference between the two modern populations was the result of the factors tied to geographic variation. However, the previous second thickest mean human molar CT value (seen in the maxillary molars of a Northern European sample; Reid and Dean, 2006) is only 0.06mm thinner that of the British Modern-day sample (see Table 7.6). This suggests that while modern-day human populations could possess thicker first molar cuspal enamel than more ancient populations, the thickness within modern-day

populations can vary to a high degree. This finding concurs with previous comments within this chapter.

The mean LT of the Modern-day first molars is the highest value ever reported. This value is 0.21mm thicker by mean than the previous thickest value (Suwa and Kono, 2005). This difference is of particular note, as until now the range of human molar mean LT values was only ±0.11mm. The value reported here for the British Modern-day population further highlights the more extreme variation in first molar enamel thickness which can exist between human populations. This is similar to the variation potential highlighted by the mean CT values published by Schwartz (2000a), also collected from a modern-human population. This finding concurs with previous comments in Chapter 5 relating to variation between the Roman and Modern-day populations, and possible links to dietary shifts (Aris et al., 2020b). However, little research has been conducted on human molar LT measures, and thus an evidenced cause for this variation is difficult to ascertain.

Table 7.6. *Mean molar enamel thickness measures (±SD) for the British and other published human populations.*

Population	N	Thickness feature		
		RET	CT (mm)	LT (mm)
This study				
Roman	6-10	17.02 ± 1.74	1.46 ± 0.20	1.34 ± 0.12
Early Anglo-Saxon	7-11	21.52 ± 2.05	1.18 ± 0.34	1.39 ± 0.19
Late Anglo-Saxons	8-19	16.21 ± 1.48	1.17 ± 0.23	1.44 ± 0.11
Medieval	22-27	16.30 ± 2.03	1.16 ± 0.22	1.44 ± 0.21
Modern-day	10-15	19.24 ± 3.77	1.27 ± 0.57	1.68 ± 0.57
Past studies				
Smith et al., 2006a[†]	37	13.95-23.86		
Smith et al., 2006a[‡]	55	11.67-22.62		
Olejniczak et al., 2008[†*]	6	17.05		
Olejniczak et al., 2008[‡*]	1	15.87		
Schwartz, 2000a[¶]	9		1.77 ± 0.48	
Reid and Dean, 2006[§]	15		1.21 ± 0.241	
Reid and Dean, 2006[¶]	37		1.09 ± 0.113	
Mahoney, 2010[¶]	69		1.13 ± 0.22	1.43 ± 0.19
Suwa and Kono, 2005[§]	31		0.85 ± 0.206	1.47 ± 0.119
Suwa and Kono, 2005[¶]	37		1.12 ± 0.160	1.44 ± 0.113
Macho and Berner, 1993[§]	21			1.36 ± 0.17

[†] data for maxillary M1s

[‡] data for mandibular M1s

[§] data for paracone cusp

[¶] data for metaconid cusp

* includes 3D measurements

7.3 Discussion – concluding remarks

7.3.1 Variation in enamel growth

In Chapters 5 and 6 it was found that there were significant trends of slowing enamel growth over time between British populations, for both upper anterior (Aris et al., 2020a) and posterior first molar (Aris et al., 2020b) teeth. These address the first research question of this project regarding inter-population variation in enamel growth, with the results showing a previously unknown level of plasticity in the growth rates of human enamel, as they can now be observed to significantly vary between populations within a time period as short as 2000 years. As previously discussed in these chapters, it is important that future studies now consider this when using single human populations as representative samples in comparative analyses of additional human and non-human primate DSRs.

Conversely to inter-population analysis, subsequent analyses found no direct significant difference between mean DSRs of biological male or female samples, directly addressing the second research question of this project. These results concur with similar past research (Schwartz et al., 2001). These findings are of particular interest within the context of the inter-population differences over time, as this suggests that the factor(s) resulting in varying enamel growth rates over time was not discriminatory between male and female populations.

To address the third research question of this project, when DSR data is viewed from across the tooth row, previously unknown trends in human enamel growth variation can be observed. Typically, first molars appear to be the fastest growing teeth. This is particularly evident in the Roman, Early Anglo-Saxon, Medieval, and Modern-day populations. Upper first incisors appear to also grow at faster rates than upper canines in the majority of cases, and are similar to first molars (or marginally faster in isolated cases) in the Roman, Late Anglo-Saxon, and Medieval populations. Upper canines were the slowest growing teeth in the majority of cases as seen in the Roman, Early Anglo-Saxon, Medieval, and Modern-day populations. Similarly to the consistency in DSRs between the biological sexes, the differences between tooth types are consistent within the trend of changes over time between populations. This suggests also that the factor(s) resulting in changes in DSRs over time between populations have had an equal influence across permanent upper first incisors, upper canines, and first molars.

While the trends of faster growing first molar and slower growing upper canine enamel are consistent in four of the British populations, the Early-Anglo Saxon population presents contradictory data. In particular, Early Anglo-Saxon first molars were the slowest growing teeth

across all enamel areas and regions. The Early Anglo-Saxon population also presented the highest degree of variation between the biological sexes in the anterior teeth. This suggests that an additional factor influenced the growth rates of Early Anglo-Saxon permanent enamel, which did not persist to the Late Anglo-Saxon period. Given the regional differences between the British populations this difference in growth across the tooth row could be unique to the Ozengell population. If true this could suggest that enamel growth is even more plastic than evidence here indicates, and able to vary within single populations over even shorter periods of time. It would therefore be interesting for future research to investigate additional Early Anglo-Saxon populations for differences in DSRs between tooth types, populations, and biological sexes.

The potential for the impact of genetic diversity must be considered. While collection of genetic data was outside of the ability and scope of this project, there is a real possibility of genetic factors causing variation within populations, and thereby influencing the differences in enamel growth rates between populations. High levels genetic diversity could for instance explain the unique variation seen in the Early Anglo-Saxon population. As discussed in Chapter 5 the period related to the Roman and Anglo-Saxon populations analysed here is associated with rapid increases in migration into Britain (e.g. Lightfoot et al., 2009), with the same being true for the current period of time directly associated with the Modern-day population. Both these periods of migration will have resulted in significant gene flow into British populations. While the clear trends in enamel growth rates over time between populations could suggest a minimal impact of these periods of high levels of migration, the potential influence of genetic drift should not be ignored. It would be valuable for future research to further investigate the genetics and origins of these populations to help develop an understating of how far migration and resulting generic diversity has influenced enamel grow rate variation. However, due to legal restrictions associated with the Modern-day extracted samples this is not viable for the specific collection analysed here.

7.3.2 Molar enamel thickness variation between populations

Analysis of the first molar thickness measures collected address the fourth research question of this project. The features of first molar enamel thickness showed little variation between British populations; particularly LT and CT, especially within the context of temporal variation (see Chapter 5). However, comparison of previously published British populations alludes to a degree of variation existing between human populations not seen until now. Relative enamel thickness in particular is highly variable between populations, as well as between time periods. While the Roman, Late Anglo-

Saxon, and Medieval populations had relatively larger mean RET values, all these were still within known ranges of previously published data. In contrast, the mean RET values of the Early Anglo-Saxon now represent the highest mean RET values for human first molars. The results comparing the Early Anglo-Saxon population to known ranges further supports comments from Chapter 5 that a highly abrasive, sea-food heavy diet, resulted in changes in enamel thickness between the British populations (Aris et al., 2020b).

The mean CT values of the British populations remained within the previously published literature. However, this is only true given the incredibly high mean CT value published by Schwartz (2000a). That value notwithstanding, the Roman and Modern-day populations had mean CT values thicker than previously known ranges. These observations concur with those of Chapter 5, which comment on the high variability of CT which can now be seen to exist between populations within as short a period as 2000 years (Aris et al., 2020b).

Mean LT values of the archaeological British populations all fall with known published ranges. Conversely, the Modern-day sample possessed the thickest mean molar LT ever published for a human population. However, rather than being an indication of large inter-population differences, this is likely the result of the low volume of data published for human molar LT. This supports the findings of Chapter 5 which only observed a single significant difference between the Roman and Modern-day populations (p = 0.04; Aris et al., 2020b).

The potential impact of genetic diversity must again be considered here for enamel thickness. As discussed previously in section *7.3.1*, and in Chapter 5 when specifically relating to enamel thickness, the period of time associated with the Roman, Anglo-Saxon, and Modern-day populations analysed here is associated with rapid increases in migration, and thereby gene flow, into Britain (e.g. Lightfoot et al., 2009). Unlike with enamel growth rates, no clear trends in enamel thickness variation were seen between populations over time. This could potentially allude to a more significant impact of increased genetic diversity. It would therefore be even more valuable for future research to further investigate the genetics and origins of these populations to help develop our understating of how migration and increased generic diversity enamel thickness expression both within and between populations.

7.3.3 Summary

The aims of the research presented here were to expand upon our understanding of the evolutionary plasticity of modern human permanent enamel. By analysing changes in features of

enamel thickness and growth rates, this has been achieved by discussing any variation within the context of: changes over time between populations, differences between biological male and female groups, and across the tooth row within populations.

In regards to evolutionary plasticity of enamel growth, it appears to be consistent between the sexes (Aris et al., 2020a), which concurs with past similar findings (Schwartz et al., 2001). However, variation between populations show a series of significant changes, most notably significant trends of slowing DSRs over time in both first molar (Aris et al., 2020b) and upper anterior teeth (Aris et al., 2020a). Such trends have never been observed until now, and show a new level of plasticity in modern human permanent enamel.

Within the context of changes over time, enamel thickness was shown to be less plastic than DSRs, with less inter-population differences (Aris et al., 2020b). However, the significant changes observed do allude to a correlation with the shift towards more abrasive diets in Early Anglo-Saxon populations. This agrees with past research which has suggested that enamel thickness in human molars can significantly differ over time between populations as the result of dietary shifts (Le Luyer et al., 2014; Le Luyer and Bayle, 2017). Moreover, the ranges of LT and RET for human first molars have been increased with the publication of British population's mean thicknesses. While this does not necessarily increase our understanding of enamel plasticity between populations, it does widen our understanding of existing ranges in human molar thickness.

Even though the mean first molar enamel thickness measures gathered here have been discussed for their differences to previously published ranges, it should still be noted that these remained relatively consistent between the British populations (Aris et al., 2020b). Conversely, the mean DSRs varied significantly between populations over time for all tooth types and enamel regions analysed. This suggests that changes in enamel growth and thickness between populations are not tightly linked.

7.4 Limitations

7.4.1 Limited tooth types and only one tooth analysed per individual

While this project analysed 285 different teeth, these were isolated to permanent first molars, and upper first incisors and canines, all of which came from different individuals. It could be argued this represents a good number of tooth types, spanning both the anterior and posterior regions of the dental arcade. Nevertheless, it must be considered that second incisors, second and third molars,

the majority of mandibular teeth, and the entire premolar tooth type have not be used in any novel analysis. Moreover, at no point could the different teeth analysed here be compared within the context of a single individual. While this is a significant number of unconsidered tooth types, and missed potential analyses, these omissions are purely the result of time and resource constraints, and institutional request during sample collections which impacted this project.

To summarise:

- Institutions regularly permitted only the sampling of small sample sizes per population (typically 10 teeth per collection). Expanding to additional tooth types would have limited already reduced samples in some cases (discussed later in this section) to unacceptable levels for strong statistical analysis.
- Institutions consistently requested that individual sets of remains only have one tooth sampled. This was commonly reasoned as to reduce the impact of destructive analysis conducted for this project on future research using the same collections.
- The extended time required to section and subsequently collect all measurements from a single tooth, via histology and light microscopy, would have meant the inclusion of suitably large samples from other tooth types would not have been possible within the timeframe of this project.
- The cost of sampling additional teeth, incurred through the need to collect additional teeth and then use consumables to section them, could not have been supported through available research funds.

While these factors limiting the use of additional tooth types were unavoidable, given a significant portion of the later discussion presented here concerns inter-tooth difference in regional DSRs, this limitation must still be considered. The conclusions of these discussions must therefore be noted as preliminary, and observations must be noted to only have been made between first molars, and upper anterior tooth types (defined as first incisors and canines). Future research would therefore be valuable in expanding the research and analysis here into different tooth types and further into the mandibular dental arcade.

Similarly, the impact of only analysing one tooth per individual must be mentioned considering the previous discussion regarding inter-tooth differences. With analyses including 285 teeth from 285 different individuals, no analysis being conducted across a single individual's dental arcade limits how far observations of differences in regional DSRs between teeth can be interpreted. A potential line of future research would be to return to the same individuals analysed here and collect data

from a different tooth than already sampled (i.e. an upper first incisor/canine is collected where the molar is presented here), to test whether inter-tooth DSR difference are consistent when compared in individuals. Such analyses would also help account for differences in diversity of population genetics and ancestry. This was another factor that could not be accounted for due to the availability of genetic data from the archaeological populations and ethnicity of the modern-day population (see section 4.1).

7.4.2 Disparity in sample sizes

Due to the destructive nature and temporal transect of this project, some populations could be represented by larger sample sizes. Notably, the Medieval and Anglo-Saxon populations presented here are represented by larger samples as a result of such populations being abundant and readily available for analysis in the United Kingdom. Conversely, Roman and modern-day dental collections are rarer, and as a result gaining permission to conduct destructive sampling of such collections was difficult, even when the collections could be located. In the case of sourcing Roman teeth, collections are less common as Roman Britain is associated with high levels of cremation as a burial practice (e.g. Williams, 2004), which rarely leaves dental remains suitable for the analyses conducted here. As a result of suitable Roman collections being rarer the relative value of the remains is increased, which in turn reduces the chance of destructive analysis being permitted. Similar limitations surrounded accessing modern-day dentition, as modern extraction samples are rarely available to anthropological research. This is due to each modern-day collections sourced from living individuals requiring ethical approval for destructive analysis from the United Kingdom National Health Service research ethics committee. Despite these issues surrounding sourcing and sampling Roman and modern-day teeth, it was ensured that ten of each tooth type analysed in this project (first molar, upper canine, upper first incisor) were sampled to support the subsequent statistical analysis.

7.4.3 Low number of dental growth measures

One further limitation of this project was the limited number of measures relating to enamel growth which were collected and presented. While regional DSR collection and calculations involved a large number of measurements being taken, and their analysis allowed for discussion regarding enamel growth variation throughout the enamel cap, a number of commonly analysed enamel growth

measures were not collected as a part of this project. Most notably crown formation times (CFTs), crown initiation, and enamel extension rates (EERs) are not presented here.

Crown formation time is a valuable measure as it allows for calculation of the exact time (in days) it takes for a crown to fully mature. This could be analysed alone with a research project, or alongside DSRs, to investigate enamel growth differences between tooth types and/or populations (e.g. Antoine et al., 2009). Unfortunately, calculating CFTs is a very time consuming process. This typically involves creating a high-resolution composite image of an entire dental crown, and subsequently counting over 1000 cross striations per tooth. Crown initiation relates to the age at which a tooth begins to mineralise, and is typically calculated by counting the number of cross striations, counted along an enamel prism originating from the dentine horn, between the EDJ and neonatal line (e.g. Antoine and Hillson, 2016; Reid and Guatelli-Steinberg, 2017). While there is value in comparing this measure between tooth types and populations, this measure routinely involves counting upwards of 200 cross striations per tooth, and is thus a highly time consuming process. Finally, EERs account for the rate of ameloblast activation as enamel matrix secretion begins along the EDJ during amelogenesis (e.g. Antoine et al., 2018). It is calculated by counting cross striations along a prism to a striae of Rezius, and then measuring the distance between the prism's origin and intersection of the Retzius line with the EDJ, thus allowing the calculation of the rate of cellular differentiation along the EDJ (e.g. Guatelli-Steinberg et al., 2012b; Antoine et al., 2018). Enamel extension rates are not a singular measure, and can be continuously measured along the EDJ to show changes in rates of differentiation along the length of the EDJ. Depending on the number of cross striations counted for each EER calculation, this method can also involve counting over 1000 lines per tooth analysed.

Overall, it was the constraints (as previously outlined in section *7.4.1*) which resulted in these additional growth measures of enamel not being included in this project. It is worth noting that crown formation times and enamel extension rates were initially planned to be collected for this project, and preliminary data collection was started. However, due to limits of data collection time focus was kept on DSRs to ensure large and reliable sample sizes were obtained for each region. Valuable future research could now be conducted, returning to the teeth analysed here, by examining whether similar trends to those seen in DSRs can be observed in changes in CFTs, EERs and initiation rates over time between populations.

CHAPTER 8: CONCLUSIONS

8.1 First molar enamel

The first research question of this project posed whether enamel growth rates of first molars had varied over time between populations. The first molars of the British populations showed a significant trend in the slowing of DSRs over a 2000 year period. This was seen in all regions of the first molar enamel. Until now, molar DSRs have only been recorded within an intra-population context which has suggested minimal intraspecific variation (Beynon et al., 1991b; Lacruz and Bromage, 2006; Smith et al., 2007b; Mahoney, 2008). This research showed that permanent first molar DSRs can instead significantly vary between British populations. Future research would be well served extending research on the temporal variation of British populations to those more ancient than Roman, including Bronze and Iron Age samples. This could show how expansive the gradual slowing of DSRs between British populations truly is. In addition, a replica study of the one presented here using populations of different geographic origin, but still spanning the last 2000 years, would aluminate as to whether the trend of slowing DSRs is isolated to British populations.

The fourth research question asked whether enamel thickness of the same first molars had varied over time between populations. First molar enamel thickness is concluded to have varied between British populations to a far lesser extent compared to DSRs, with no clear trend between populations. First molar enamel thickness varied most notably between the Early and Late Anglo-Saxon populations. The Early period had larger RET and thicker LT compared to the Late Anglo-Saxon population. Past research has suggested variation in molar thickness had been recorded between populations differing by a minimum of 7000 years (Le Luyer et al., 2014). Therefore, data presented here suggests that human first molar enamel thickness is even more evolutionarily plastic than previously thought, as the Early and Late Anglo Saxon populations differ in date by a range of only 200-700 years. Moreover, the increased thickness of the Early Anglo-Saxon first molars is attributed to a more abrasive marine diet. While this theory is based on historical data of the period in comparison to thickness measures collected here, it does agree with past research of human molar enamel thickness which suggested variations between populations to be the result of differential dietary practices (e.g. Le Luyer et al., 2014; Le Luyer and Bayle, 2017). Future research should aim to address this concept by analysing thickness data of an Early Anglo-Saxon and/or coastally-based population in conjunction with collecting dietary data, in order to make direct comparisons between the two.

8.2 Upper anterior tooth enamel

The first research question of this project also posed whether the enamel growth rates of upper anterior teeth had varied over time between populations. Daily secretion rates for the upper canines and upper first incisors showed variation between the British populations, with a significant trend towards the slowing of enamel in more recent populations. The trend was also observed in both male and female samples during independent inter-population analyses. It is difficult to assess the DSR ranges for the permanent British anterior teeth in the same manner as the first molar data, as there is little available equivalent published data, unlike that available for canines (Schwartz et al., 2001). However, what can be said is that high degrees of variation between populations can occur in these DSRs, even when the populations share a dated origin within a 2000 year period.

The third research question further asked if enamel growth had varied between different tooth types. When isolated, Anglo-Saxon analyses found the Late Anglo-Saxon population to possess faster mean upper first incisor DSRs on average than that of the Early Anglo-Saxon population. These were significantly faster in the inner and mid lateral upper first incisor regions. However, the upper canine enamel DSRs presented variation akin to both the first molars and upper first incisors. While the inner and mid cuspal, and inner and mid lateral upper canine DSRs of the Late Anglo-Saxon population were also on average faster, the cuspal regions of both enamel areas were faster in the Early Anglo-Saxon population. Future research focussing on the Anglo-Saxon period could therefore help widen our understanding of what caused the slowing of DSRs between human populations over time, as well as the increase in some regional DSRs between the early and Late Anglo-Saxon periods.

8.3 Enamel sexual dimorphism

The second research question presented in the introduction to this thesis expanded on questions regarding enamel growth variation, asking if growth had varied between biological male and female groups. Associated analyses found no significant difference between the biological male and female upper anterior tooth DSRs in any of the British populations. This agrees with past research by Schwartz and colleagues (2001) who found DSRs to be consistent between males and females in a modern sample. While we cannot conclude whether this is also true of the first molar enamel DSRs, the similar inter-population differences between the first molars and upper anterior tooth types would suggest first molars are unlikely to display any differentiating significant sexual dimorphism within their DSRs. Given human permanent molars have been found to possess significant sexual dimorphism (e.g. Schwartz and Dean, 2005; Cardoso, 2008; Aris et al., 2018), this could mean that

differences in size are due to longer periods of enamel growth. The evidence provided is in not substantial enough to support this theory fully, with committed future research now being required.

8.4 Enamel growth and thickness

The fifth and final research question of this project asked if any enamel growth and thickness variation correlated. While first molar enamel thickness varied between the Anglo-Saxon populations, there was little other variation between the British populations. Conversely, inter-population variation in regional DSRs occurred in all enamel areas and tooth types analysed. Overall, what was found were similarly thick teeth (outside of the Early Anglo-Saxon population) growing at significantly different speeds according between populations. While DSRs do not appear tightly linked to enamel thickness, past research has found slower Retzius periodicity to predict thicker teeth (Mahoney et al., 2018). As a result, it should not be assumed that all enamel growth data variation does not correlate with changes in thickness. Future research should thus consider if other developmental factors of enamel are more strongly linked to enamel thickness when compared to different populations.

8.5 Future research directions

A number of questions arose during the course of this research, particularly pertaining to the inconsistent enamel thickness differences between populations. Comments have been made as to the potential influence of dietary changes influencing enamel thickness, most notably in regards to the Early Anglo-Saxon population. Future research would therefore be best served conducting analyses of stable nitrogen and carbon isotopes from both bone and enamel, preferably from the same individuals from whom dental cross-sections were produced. It is thus suggested that future research should test the utility of combining isotope and histological methods within research projects. Only by doing so can the true inter-population differences in diet be identified, and the physiological changes occurring in enamel be tracked for the same populations and individuals.

The results of the inter-population DSR analyses raise further questions. While the results were undeniably significant and consistent across tooth types, it should not be assumed that the analyses of the five British populations are representative of the human species at a global or temporal scale. It would be of great benefit to conduct similar studies to those presented here on more diverse populations. The literature concerning enamel thickness and DSRs is dominated by

data gathered from European and Southern African samples. The most obvious and consistent finding of this project as a whole, is that dental development via enamel growth is highly variable between human populations with minimal difference through a temporal transect. Expansion into new populations, from different from regions of the world and time periods, is therefore vital in order to develop our understanding as to how and why enamel growth can significantly vary between populations over relatively short periods of time. For instance, the trend of slowing enamel growth between populations in Britain may not be consistent on a global scale, or even when the 2000 year time span analysed here is expanded further into the more distant past of Britain.

Given the preference for European and Southern African in research to date, future research would ideally be conducted on Asian, Indonesian, and/or Australasian samples. This would expand the available data for human enamel features to be more geographically and ethnically inclusive. Moreover, such projects would be best conducted with local and indigenous experts. This project proposed that socio-cultural factors could potentially influence enamel physiology. With expert knowledge and input, our understanding of how enamel can vary in modern humans between populations can only be expanded. Furthermore, more regular inclusion of indigenous experts in bio-archaeological projects will improve ethical standards and help expand the field outside of the current main spheres of influence in Europe and North America.

REFERENCES

Alba, D. M., Fortuny, J., & Moyà-Solà, S. (2010). Enamel thickness in the Middle Miocene great apes Anoiapithecus, Pierolapithecus and Dryopithecus. *Proceedings of the Royal Society of London B: Biological Sciences*, *277*(1691), 2237-2245.

AlQahtani, S. J., Hector, M. P., & Liversidge, H. M. (2010). Brief communication: the London atlas of human tooth development and eruption. *American Journal of Physical Anthropology*, *142*(3), 481-490.

Alvesalo, L., Tammisalo, E., & Hakola, P. (1985). Enamel thickness in 47, XYY males' permanent teeth. *Annals of Human Biology*, *12*(5), 421-427.

Anemone, R. L., Mooney, M. P., & Siegel, M. I. (1996). Longitudinal study of dental development in chimpanzees of known chronological age: implications for understanding the age at death of Plio-Pleistocene hominids. *American Journal of Physical Anthropology*, *99*(1), 119-133.

Anemone, R. L., Watts, E. S., & Swindler, D. R. (1991). Dental development of known-age chimpanzees, Pan troglodytes (primates, pongidae). *American Journal of Physical Anthropology*, *86*(2), 229-241.

Antoine, D. (2000). *Evaluating the periodicity of incremental structures in dental enamel as a means of studying growth in children from past human populations* (Doctoral dissertation, University College London).

Antoine, D., FitzGerlad, C. M., & Rose, J. C. (2018). Incremental Structures in Teeth: Keys to Unlocking and Understanding Dental Growth and Development. *Biological Anthropology of the Human Skeleton*, 225.

Antoine, D., & Hillson, S. (2016). Enamel structure and properties. *A companion to dental anthropology*, 223-243.

Antoine, D., Hillson, S., & Dean, M. C. (2009). The developmental clock of dental enamel: a test for the periodicity of prism cross-striations in modern humans and an evaluation of the most likely sources of error in histological studies of this kind. *Journal of Anatomy*, *214*(1), 45-55.

Aris, C. (2020). The Histological Paradox: Methodology and Efficacy of Dental Sectioning. *Papers from the Institute of Archaeology*, 29(1).

Aris, C., Mahoney, P., & Deter, C. (2020a). Enamel thickness and growth rates in modern human permanent first molars over a 2000 year period in Britain. *American Journal of Physical Anthropology*.

Aris, C., Mahoney, P., O'Hara, M. C., & Deter, C. (2020b). Enamel growth rates of anterior teeth in males and females from modern and ancient British populations. *American Journal of Physical Anthropology*.

Aris, C., & Street, E. (2021). Growth rates of accessory human enamel: a histological case study of a modern-day incisor from Northern England. *Dental Anthropology Journal*, *34*(1), 3-12.

Arnold, C. J. (1988). Early Anglo-Saxon Pottery in 'Illington-Lackford' Type 1. *Oxford Journal of Archaeology*, 7(3), 343-359.

Arnold, C. J. (2005). *An archaeology of the early Anglo-Saxon kingdoms*. Routledge.

Asper, H., (1916). Uber die ''braune Retzius'sche Parallelsteifung'' im Schmelz der menschlichen Zahne; Schweiz Vjschr. Zahnhlk. 26(1), 275-314

Barrett, J. H., Locker, A. M., & Roberts, C. M. (2004). The origins of intensive marine fishing in medieval Europe: the English evidence. *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 271(1556), 2417-2421.

Baumgartner, A., Dichtl, S., Hitzenberger, C. K., Sattmann, H., Robl, B., Moritz, A.., & Sperr, W. (2000). Polarization–sensitive optical coherence tomography of dental structures. *Caries Research*, 34(1), 59-69.

Beek, G. C. van. (1983). *Dental Morphology: an Illustrated Guide*. Wright.

Bellisari, A., Duren, D. L., & Sherwood, R. J. (2005). Sex differences in emergence of deciduous dentition in captive lowland gorillas (*Gorilla gorilla gorilla). American Journal of Physical Anthropology,* 72-72.

Bellisari, A., Newman, T. K., Greenberg, C., Rogers, J., & Towne, B. (2001). Individual variation in the growth of captive infant gorillas. *American Journal of Physical Anthropology*, 115(2), 110-132.

Benazzi, S., Panetta, D., Fornai, C., Toussaint, M., Gruppioni, G., & Hublin, J. J. (2014). Guidelines for the digital computation of 2D and 3D enamel thickness in hominoid teeth. *American Journal of Physical Anthropology*, 153(2), 305-313.

Benefit BR. (1987). *The molar morphology, natural history, and phylogenetic position of the middle Miocene monkey Victoriapithecus*. (Unpublished doctoral thesis). New York University, New York.

Berkovitz, B. K. B., Holland, G. R., & Moxham, B. J. (2002). *Oral Anatomy, Embryology and Histology*. Mosby Incorporated.

Beynon, A. D. (1992). Circaseptan rhythms in enamel development in modern humans and Plio-Pleistocene hominids. In: *Structure, Function and Evolution of Teeth*, pp. 295-309.

Beynon, A. D., & Dean, M. C. (1987). Crown-formation time of a fossil hominid premolar tooth. *Archives of Oral Biology*, 32(11), 773-780.

Beynon, A. D., & Dean, M. C. (1988). Distinct dental development patterns in early fossil hominids. *Nature*, *335*(6190), 509-514.

Beynon, A. D., & Reid, D. J. (1987). Relationships between perikymata counts and crown formation times in the human permanent dentition. *Journal of Dental Research,* 66(4), 889-889).

Beynon, A. D., & Wood, B. A. (1986). Variations in enamel thickness and structure in East African hominids. *American Journal of Physical Anthropology*, 70(2), 177-193.

Beynon, A. D., & Wood, B. A. (1987). Patterns and rates of enamel growth in the molar teeth of early hominids. *Nature*, 326(6112), 493-496.

Beynon, A. D., Clayton, C. B., Ramirez Rozzi, F. V. R., & Reid, D. J. (1998a). Radiographic and histological methodologies in estimating the chronology of crown development in modern humans and great apes: a review, with some applications for studies on juvenile hominids. *Journal of Human Evolution*, 35(4), 351-370.

Beynon, A. D., Dean, M. C., & Reid, D. J. (1991a). Histological study on the chronology of the developing dentition in gorilla and orangutan. *American Journal of Physical Anthropology*, 86(2), 189-203.

Beynon, A. D., Dean, M. C., & Reid, D. J. (1991b). On thick and thin enamel in hominoids. *American Journal of Physical Anthropology*, 86(2), 295-309.

Beynon, A. D., Dean, M. C., Leakey, M. G., Reid, D. J., & Walker, A. (1998b). Comparative dental development and microstructure of Proconsul teeth from Rusinga Island, Kenya. *Journal of Human Evolution*, 35(2), 163-209.

Beynon, A.D. & Wood, B.A. (1986). Variations in enamel thickness and structure in East African hominids. *American Journal of Physical Anthropology*, 70, 177-194

Birch, W. J. (2012). *Incremental Growth of Deciduous Tooth Enamel* (Doctoral dissertation, University College London).

Birch, W., & Dean, M. C. (2009). Rates of enamel formation in human deciduous teeth. In: *Comparative Dental Morphology* (Vol. 13, pp. 116-120). Karger Publishers.

Birch, W., & Dean, M. C. (2014). A method of calculating human deciduous crown formation times and of estimating the chronological ages of stressful events occurring during deciduous enamel formation. *Journal of Forensic and Legal Medicine*, 22, 127-144.

Bodecker, C. F. (1953). Enamel lamellae and their origin. *Journal of Dental Research*, 32(2), 239-245.

Bolter, D. R., & Zihlman, A. L. (2003). Morphometric analysis of growth and development in wild-collected vervet monkeys (Cercopithecus aethiops), with implications for growth patterns in Old World monkeys, apes and humans. *Journal of Zoology*, 260(1), 99-110.

Bowes, J. H., & Murray, M. M. (1935). The chemical composition of teeth: The composition of human enamel and dentine. *Biochemical Journal*, 29(12), 2721.

Boyde, A. (1963). Estimation of age at death of young human skeletal remains from incremental lines in the dental enamel. In: *Third International Meeting in Forensic Immunology, Medicine, Pathology and Toxicology*, 1, 36-37.

Boyde, A. (1967). The development of enamel structure.

Boyde, A. (1979). Carbonate concentration, crystal centers, core dissolution, caries, cross striations, circadian rhythms, and compositional contrast in the SEM. *Journal of Dental Research*, 58(2), 981-983.

Boyde, A. (1989). Enamel. In *Teeth*. Springer, Berlin, Heidelberg.

Boyde, A., (1990). Developmental interpretations of dental microstructure. In: DeRousseau, J.C. (Ed.), *Primate Life History and Evolution*. Wiley-Liss, New York, pp. 229-267.

Boyde, A. (1997). Microstructure of enamel. In: *Ciba Foundation Symposium* (pp. 18-31).

Boyde, A., & Martin, L. (1984). The microstructure of primate dental enamel. In: *Food Acquisition and Processing in Primates*, pp. 341-367. Springer, Boston, MA.

Boyde, A., (1963). Estimation of age at death from young human skeletal remains from incremental lines in dental enamel. *Third International Meeting in Forensic Immunology, Medicine, Pathology and Toxicology*, London, pp. 36-46

Boyde, A., (1990). Developmental interpretations of dental microstructure. In: DeRousseau, J.C. (Ed.), *Primate Life History and Evolution*. Wiley-Liss, New York, pp. 229-267.

Boyle, A., Barcley, A., Dodd, A., Miles, D., & Mudd, A. (1995). *Two Oxfordshire Anglo-Saxon Cemeteries: Berinsfield and Didcot*. Oxford University committee for archaeology.

Brace, C. L., & Nagai, M. (1982). Japanese tooth size: past and present. *American Journal of Physical Anthropology*, 59(4), 399-411.

Brace, C. L., & Ryan, A. S. (1980). Sexual dimorphism and human tooth size differences. *Journal of Human Evolution*, 9(5), 417-435.

Brace, C. L., Rosenberg, K. R., & Hunt, K. D. (1987). Gradual change in human tooth size in the late Pleistocene and post-Pleistocene. *Evolution*, 41(4), 705-720.

Bromage, T. G. (1987). The biological and chronological maturation of early hominids. *Journal of Human Evolution*, 16(3), 257-272.

Bromage, T. G. (1991). Enamel incremental periodicity in the pig-tailed macaque: A polychrome fluorescent labeling study of dental hard tissues. *American Journal of Physical Anthropology*, 86(2), 205-214.

Bromage, T. G. (1992). Microstructural organization and biomechanics of the macaque circumorbital region. *Structure, function and evolution of teeth. London: Freund Publishing House,* pp. 257-272.

Bromage, T. G., & Dean, M. C. (1985). Re-evaluation of the age at death of immature fossil hominids. *Nature*, 317(6037), 525-527.

Bromage, T. G., Hogg, R. T., Lacruz, R. S., & Hou, C. (2012). Primate enamel evinces long period biological timing and regulation of life history. *Journal of Theoretical Biology*, 305, 131-144.

Bromage, T. G., Perez-Ochoa, A., & Boyde, A. (2005). Portable confocal microscope reveals fossil hominid microstructure. *Journal of Microscopy*, 19, 5-7.

Brunet, M., Guy, F., Pilbeam, D., Mackaye, H. T., Likius, A., Ahounta, D., & De Bonis, L. (2002). A new hominid from the Upper Miocene of Chad, Central Africa. *Nature*, 418(6894), 145.

Campbell, J., John, E., Wormald, P., Addyman, P. V., & Hawkes, S. C. (1982). *The Anglo-Saxons*. Oxford: Phaidon.

Cape, A. T., & Kitchin, P. C. (1930). Histologic Phenomena of Tooth Tissues as Observed under Polarized Light; with a note on the Roentgen-Ray Spectra of Enamel and Dentin From the research laboratory of the dental school of the Ohio State University, Paul C. Kitchin, director. Read before the Research Session at the Sixth Annual Meeting of the American Association of Dental Schools, Chicago, Ill., March 26, 1929. *The Journal of the American Dental Association (1922)*, 17(2), 193-227.

Cardoso, H. F. (2008). Sample-specific (universal) metric approaches for determining the sex of immature human skeletal remains using permanent tooth dimensions. *Journal of Archaeological Science*, *35*(1), 158-168.

Cathey, W. J. (1963). A plastic embedding medium for thin sectioning in light microscopy. *Stain Technology*, 38(4), 213-216.

Chandrasekera, M. S., Reid, D. J., & Beynon, A. D. (1993). Dental chronology in chimpanzee (Pan troglodytes). *Journal of Dental Research*, 72, 729.

Chenery, C., Müldner, G., Evans, J., Eckardt, H., & Lewis, M. (2010). Strontium and stable isotope evidence for diet and mobility in Roman Gloucester, UK. *Journal of Archaeological Science*, 37(1), 150-163.

Christensen, G. J., & Kraus, B. S. (1965). Initial calcification of the human permanent first molar. *Journal of Dental Research*, 44(6), 1338-1342.

Conroy, G. C. (1988). Alleged synapomorphy of the M1/I1 eruption pattern in robust australopithecines and Homo: Evidence from high-resolution computed tomography. *American Journal of Physical Anthropology*, 75(4), 487-492.

Conroy, G. C., & Vannier, M. W. (1987). Dental development of the Taung skull from computerized tomography. *Nature*, 329(6140), 625-627.

Conroy, G. C., & Vannier, M. W. (1988). The nature of Taung dental maturation continued. *Nature*, 333(6176), 808-808.

Conroy, G. C., & Vannier, M. W. (1991). Dental development in South African australopithecines. Part II: Dental stage assessment. *American Journal of Physical Anthropology*, 86(2), 137-156.

Conroy, M. W. (1989). The Taung skull revisited: New evidence from high-resolution computed tomography. *South African Journal of Science*, 85(1), 30.

Constantino, P. J., Lee, J. J. W., Chai, H., Zipfel, B., Ziscovici, C., Lawn, B. R., & Lucas, P. W. (2010). Tooth chipping can reveal the diet and bite forces of fossil hominins. *Biology Letters*, 6(6), 826-829.

Crabb, H. S. (1966). Enamel caries. Observations on the histology and pattern of progress of the approximal lesion. *British Dental Journal*, 121(3), 115.

Crawford, S. (1999). *Childhood in Anglo-Saxon England*. Sutton Publishing.

Daegling, D. J., McGraw, W. S., Ungar, P. S., Pampush, J. D., Vick, A. E., & Bitty, E. A. (2011). Hard-object feeding in sooty mangabeys (Cercocebus atys) and interpretation of early hominin feeding ecology. *PLoS One*, 6(8), e23095.

Dancey, J. T., Deubelbeiss, K. A., Harker, L. A., & Greenaway, J. (1976). Section preparation of human marrow for light microscopy. *Journal of Clinical Pathology*, 29(8), 704-710

Danielson, D. R., & Reinhard, K. J. (1998). Human dental microwear caused by calcium oxalate phytoliths in prehistoric diet of the lower Pecos region, Texas. *American Journal of Physical Anthropology*, 107(3), 297-304

Darling, A. I. (1958). Studies of the early lesion of enamel caries. *British Dental Journal*, *105*, 119-134.

Daubert, D. M., Kelley, J. L., Udod, Y. G., Habor, C., Kleist, C. G., Furman, I. K., & Roberts, F. A. (2016). Human enamel thickness and ENAM polymorphism. *International Journal of Oral Science*, 8(2), 93.

De Castro, J. M. B. (1985). *La dentición de los pobladores prehistóricos de las Islas Canarias: estudio antropológico* (Doctoral dissertation).

De Castro, J. M. B. (1989a). The Carabelli trait in human prehistoric populations of the Canary Islands. *Human Biology*, 117-131.

De Castro, J. M. B. (1989b). Third molar agenesis in human prehistoric populations of the Canary Islands. *American Journal of Physical Anthropology*, 79(2), 207-215.

De Menezes Oliveira, M. A. H., Torres, C. P., Gomes-Silva, J. M., Chinelatti, M. A., De Menezes, F. C. H., Palma-Dibb, R. G., & Borsatto, M. C. (2010). Microstructure and mineral composition of dental enamel of permanent and deciduous teeth. *Microscopy Research and Technique*, 73(5), 572-577.

Dean, C. (2009). Extension rates and growth in tooth height of modern human and fossil hominin canines and molars. In: *Comparative Dental Morphology* (Vol. 13, pp. 68-73). Karger Publishers.

Dean, C., Leakey, M. G., Reid, D., Schrenk, F., Schwartz, G. T., Stringer, C., & Walker, A. (2001). Growth processes in teeth distinguish modern humans from Homo erectus and earlier hominins. *Nature*, 414(6864), 628.

Dean, H. T. (1933). Distribution of Mottled Enamel in the United States*†* Read before the Section on Histology. Physiology, Pathology, Bacteriology and Chemistry (Research) at the Seventy-Fourth Annual Session of the American Dental Association, Buffalo, NY, Sept. 15, 1932.† Dental surgeon, US Public Health Service. *The Journal of the American Dental Association (1922)*, 20(2), 319-333.

Dean, J. A., Avery, D. R., & Swartz, M. L. (1986). Attachment of anterior tooth fragments. *Paediatric Dentistry*, 8(3), 139-143.

Dean, M. C. (1987a). The dental developmental status of six East African juvenile fossil hominids. *Journal of Human Evolution*, 16(2), 197-213.

Dean, M. C. (1987b). Growth layers and incremental markings in hard tissues; a review of the literature and some preliminary observations about enamel structure in Paranthropus boisei. *Journal of Human Evolution*, 16(2), 157-172.

Dean, M. C. (1995). The nature and periodicity of incremental lines in primate dentine and their relationship to periradicular bands in OH 16 (Homo habilis). *Aspects of Dental Biology: Paleontology, Anthropology and Evolution*, 239-265.

Dean, M. C. (1998). A comparative study of cross striation spacings in cuspal enamel and of four methods of estimating the time taken to grow molar cuspal enamel in Pan, Pongo and Homo. *Journal of Human Evolution*, 35(4), 449-462.

Dean, M. C. (2000). Incremental markings in enamel and dentine: what they can tell us about the way teeth grow. *Development, Function and Evolution of Teeth*, 119-130.

Dean, M. C., & Beynon, A. D. (1991). Histological reconstruction of crown formation times and initial root formation times in a modern human child. *American Journal of Physical Anthropology*, 86(2), 215-228.

Dean, M. C., & Lucas, V. S. (2009). Dental and skeletal growth in early fossil hominins. *Annals of Human Biology*, 36(5), 545-561.

Dean, M. C., & Reid, D. J. (2001). Perikymata spacing and distribution on hominid anterior teeth. *American Journal of Physical Anthropology* 116(3), 209-215.

Dean, M. C., & Scandrett, A. E. (1996). The relation between long-period incremental markings in dentine and daily cross-striations in enamel in human teeth. *Archives of Oral Biology*, 41(3), 233-241.

Dean, M. C., & Schrenk, F. (2003). Enamel thickness and development in a third permanent molar of Gigantopithecus blacki. *Journal of Human Evolution*, 45(5), 381-388.

Dean, M. C., & Smith, B. H. (2009). Growth and development of the Nariokotome youth, KNM-WT 15000. In: *The First Humans – Origin and Early Evolution of the Genus Homo* (pp. 101-120). Springer Netherlands.

Dean, M. C., & Wood, B. A. (1981). Developing pongid dentition and its use for ageing individual crania in comparative cross-sectional growth studies. *Folia Primatologica*, 36(1-2), 111-127.

Dean, M. C., Beynon, A. D., Reid, D. J., & Whittaker, D. K. (1993a). A longitudinal study of tooth growth in a single individual based on long-and short-period incremental markings in dentine and enamel. *International Journal of Osteoarchaeology*, 3(4), 249-264.

Dean, M. C., Beynon, A. D., Thackeray, J. F., & Macho, G. A. (1993b). Histological reconstruction of dental development and age at death of a juvenile Paranthropus robustus specimen, SK 63, from Swartkrans, South Africa. *American Journal of Physical Anthropology*, 91(4), 401-419.

Dean, M.C., (1995). The nature and periodicity of incremental lines in primate dentine and their relationship to periradicular bands in OH 16 (Homo habilis). In: Moggi-Cecchi, J. (Ed.), *Aspects of Dental Biology, Paleontology, Anthropology and Evolution*. International institute for the study of man, Angelo Pontecorboli, Florence, pp. 239-265.

Deter, C. A. (2009). Gradients of occlusal wear in hunter-gatherers and agriculturalists. *American Journal of Physical Anthropology*, 138(3), 247-254.

Dirks, W. (1998). Histological reconstruction of dental development and age at death in a juvenile gibbon (Hylobates lar). *Journal of Human Evolution*, 35(4-5), 411-425.

Dirks, W., & Bowman, J. E. (2007). Life history theory and dental development in four species of catarrhine primates. *Journal of Human Evolution*, 53(3), 309-320.

Dirks, W., Humphrey, L. T., Dean, M. C., & Jeffries, T. E. (2010). The relationship of accentuated lines in enamel to weaning stress in juvenile baboons (Papio hamadryas anubis). *Folia Primatologica*, 81(4), 207-223.

Dirks, W., Reid, D. J., Jolly, C. J., Phillips-Conroy, J. E., & Brett, F. L. (2002). Out of the mouths of baboons: stress, life history, and dental development in the Awash National Park hybrid zone, Ethiopia. *American Journal of Physical Anthropology*, 118(3), 239-252.

Driessens, F. C. M., Heijligers, H. J. M., Borggreven, J. M. P. M., & Wöltgens, J. H. M. (1984). Variations in the mineral composition of human enamel on the level of cross-striations and striae of Retzius. *Caries Research*, 18(3), 237-241.

Dumont ER. (1995). Enamel thickness and dietary adaptation among extant primates and chiropterans. *Journal of Mammalogy*, 76:1127– 1136.

Eisenmann. (1998). *Enamel structure***.** R.T. Cate (Ed.), Oral Histology, Mosby, St. Louis, Missouri.

Elhechmi, I., Braga, J., Dasgupta, G., & Gharbi, T. (2013). Accelerated measurement of perikymata by an optical instrument. *Biomedical Optics Express*, 4(10), 2124-2137.

Eli, I., Sarnat, H., & Talmi, E. (1989). Effect of the birth process on the neonatal line in primary tooth enamel. *Paediatric Dentistry*, 11(3), 220-223.

El-Najjar, M. Y., Desanti, M. V., & Ozebek, L. (1978). Prevalence and possible etiology of dental enamel hypoplasia. *American Journal of Physical Anthropology*, 48(2), 185-192.

Farooq, I., Ali, S., & Anderson, P. (2021). Dental Enamel. *An Illustrated Guide to Oral Histology*, 15-34.

Featherstone, J. D. B., L. Holmen, A. Thylstrup, L. Fredebo, and M. Shariati. (1985). "Chemical and histological changes during development of artificial caries." *Caries Research*, 19(1), 1-10.

Feeney, R. N., Zermeno, J. P., Reid, D. J., Nakashima, S., Sano, H., Bahar, A., & Smith, T. M. (2010). Enamel thickness in Asian human canines and premolars. *Anthropological Science*, 118(3), 191-198.

Fejerskov, O., Silverstone, L. M., Melsen, B., & Møller, I. J. (1975). Histological features of fluorosed human dental enamel. *Caries Research*, 9(3), 190-210.

Fisher, D. C. (1987). Mastodont procurement by Paleoindians of the Great Lakes region: hunting or scavenging. *The Evolution of Human Hunting*, 309-421.

FitzGerald, C. M. (1995). *Tooth crown formation and the variation of enamel microstructural growth markers in modern humans* (Doctoral dissertation, University of Cambridge).

FitzGerald, C. M. (1998). Do enamel microstructures have regular time dependency? Conclusions from the literature and a large-scale study. *Journal of Human Evolution*, 35(4-5), 371-386.

FitzGerald, C. M., & Hillson, S. (2009). Deciduous tooth growth in an ancient Greek infant cemetery. In: *Comparative Dental Morphology* (Vol. 13, pp. 178-183). Karger Publishers.

FitzGerald, C. M., & Saunders, S. R. (2005). Test of histological methods of determining chronology of accentuated striae in deciduous teeth. *American Journal of Physical Anthropology*, 127(3), 277-290.

Fukuhara, T. (1959). Comparative anatomical studies of the growth lines in the enamel of mammalian teeth. *Acta Anat. Nipp*, 34, 322-332.

Fuller, B. T., Molleson, T. I., Harris, D. A., Gilmour, L. T., & Hedges, R. E. (2006). Isotopic evidence for breast feeding and possible adult dietary differences from late/sub-Roman Britain. *American Journal of Physical Anthropology*, 129(1), 45-54.

Gantt, D. G. (1979). Comparative enamel histology of primate teeth. *Journal of Dental Research*, 58(2), 1002-1003.

Gantt, D. G. (1980). Implications of enamel prism patterns for the origin of the New World monkeys. In: *Evolutionary Biology of the New World Monkeys and Continental Drift* (pp. 201-217). Springer US.

Gantt, D. G., Harris, E. F., Rafter, J. A., & Rahn, J. K. (2001). Distribution of enamel thickness on human deciduous molars. *Dental Morphology*, 167-190.

Gantt, D.G. (1983). The enamel of Neogene hominoids: structural and phyletic implications. In: *Ciochon, R. and Corruccini, R. Eds., New Interpretations of Ape and Human Ancestry*. 249-298. New York, Plenum

Gantt, D.G., (1977). *Enamel of Primate Teeth: its Thickness and Structure with Reference to Functional and Phylogenetic Implications* (Doctoral dissertation, Washington University).

Goodman, A. H., & Armelagos, G. J. (1985a). Factors affecting the distribution of enamel hypoplasias within the human permanent dentition. *American Journal of Physical Anthropology*, 68(4), 479-493.

Goodman, A. H., & Armelagos, G. J. (1985b). The chronological distribution of enamel hypoplasia in human permanent incisor and canine teeth. *Archives of Oral Biology*, 30(6), 503-507.

Goodman, A. H., & Rose, J. C. (1990). Assessment of systemic physiological perturbations from dental enamel hypoplasias and associated histological structures. *American Journal of Physical Anthropology*, 33(S11), 59-110.

Grine, F. E. (1981). Trophic differences between 'gracile' and 'robust' australopithecines: a scanning electron microscope analysis of occlusal events. *South African Journal of Science*, 77(5), 203-230.

Grine, F. E. (1988). Enamel thickness and development in Australopithecus and Paranthropus. *Evolutionary History of the "Robust" Australopithecines*, 3-42.

Grine, F. E. (2002). Scaling of tooth enamel thickness, and molar crown size reduction in modern humans. *South African Journal of Science*, 98(9-10), 503-509.

Grine, F. E. (2005). Enamel thickness of deciduous and permanent molars in modern Homo sapiens. *American Journal of Physical Anthropology*, 126(1), 14-31.

Grine, F. E., Stevens, N. J., & Jungers, W. L. (2001). An evaluation of dental radiograph accuracy in the measurement of enamel thickness. *Archives of Oral Biology*, *46*(12), 1117-1125.

Grine, F.E. and Martin, L.B. (1988). Enamel thickness and development in Australopithecus and Paranthropus. In: Grine, F.E. Ed., *Evolutionary History of the "Robust" Australopithecines*, 3-42. New York, Aldine de Gruyter.

Guatelli-Steinberg, D. (2001). What can developmental defects of enamel reveal about physiological stress in nonhuman primates?. *Evolutionary Anthropology: Issues, News, and Reviews*, 10(4), 138-151.

Guatelli-Steinberg, D., & Lukacs, J. R. (1999). Interpreting sex differences in enamel hypoplasia in human and non-human primates: Developmental, environmental, and cultural considerations. *American Journal of Physical Anthropology*, 110(S29), 73-126.

Guatelli-Steinberg, D., & Reid, D. J. (2008). What molars contribute to an emerging understanding of lateral enamel formation in Neandertals vs. modern humans. *Journal of Human Evolution*, 54(2), 236-250.

Guatelli-Steinberg, D., & Skinner, M. (2000). Prevalence and etiology of linear enamel hypoplasia in monkeys and apes from Asia and Africa. *Folia Primatologica*, 71(3), 115-132.

Guatelli-Steinberg, D., Ferrell, R. J., & Spence, J. (2012a). Linear enamel hypoplasia as an indicator of physiological stress in great apes: reviewing the evidence in light of enamel growth variation. *American Journal of Physical Anthropology*, 148(2), 191-204.

Guatelli-Steinberg, D., Floyd, B. A., Dean, M. C., & Reid, D. J. (2012b). Enamel extension rate patterns in modern human teeth: two approaches designed to establish an integrated comparative context for fossil primates. *Journal of Human Evolution*, 63(3), 475-486.

Guatelli-Steinberg, D., Irish, J. D., & Lukacs, J. R. (2001). Canary islands-north African population affinities: measures of divergence based on dental morphology. *HOMO-Journal of Comparative Human Biology*, 52(2), 173-188.

Guatelli-Steinberg, D., Reid, D. J., & Bishop, T. A. (2007). Did the lateral enamel of Neandertal anterior teeth grow differently from that of modern humans?. *Journal of Human Evolution*, 52(1), 72-84.

Guatelli-Steinberg, D., Reid, D. J., Bishop, T. A., & Larsen, C. S. (2005). Anterior tooth growth periods in Neandertals were comparable to those of modern humans. *Proceedings of the National Academy of Sciences*, 102(40), 14197-14202.

Gustafson, A. G. (1959). *A morphologic investigation of certain variations in the structure and mineralization of human dental enamel*. Berlingska boktryckeriet.

Gustafson, G. (1957). The histopathology of caries of human dental enamel with special reference to the division of the carious lesion into zones. *Acta Odontologica Scandinavica*, 15(1), 13-55.

Gustafson, G., & Gustafson, A. G. (1961). Human dental enamel in polarized light and contact micro-radiography. *Acta Odontologica Scandinavica*, 19(2), 259-287.

Gustafson, G., & Koch, G. (1974). Age estimation up to 16 years of age based on dental development. *Odontologisk Revy*, 25(3), 297.

Gwinnett, A. J. (1971). Histologic changes in human enamel following treatment with acidic adhesive conditioning agents. *Archives of Oral Biology*, 16(7), 731IN13737IN15-736IN14738.

Gwinnett, A. John. (1966). Normal enamel. I. Quantitative polarized light study. *Journal of Dental Research*, 45(1), 120-127.

Gysi, A. (1931). Metabolism in adult enamel. *Dental Dig*, 37, 661-668

Halberg, F., Halberg, E., Barnum, C.P., Bittner, J.J., (1959). Photoperiodism and Related Phenomena in Plants and Animals, vol. 55. American Association for the Advancement of Science, Washington, DC, pp. 803 – 878.

Hallsworth, A. S., Weatherell, J. A., & Robinson, C. (1973). Loss of carbonate during the first stages of enamel caries. *Caries Research*, 7(4), 345-348.

Härke, H. (1997) Early Anglo-Saxon social structure. In: *HINES, J. (ed) The Anglo-Saxons from the Migration Period to the Eighth Century. An Ethnographic Perspective Woodbridge*, Boydell Press: 125-170.

Harris, E. F., Hicks, J. D., & Barcroft, B. D. (1998). Absence of sexual dimorphism in enamel thickness of human deciduous molars. *Dental Morphology*, 338-349.

Haydock, H., Clarke, L., Craig-Atkins, E., Howcroft, R., & Buckberry, J. (2013). Weaning at Anglo-Saxon raunds: Implications for changing breast feeding practice in britain over two millennia. *American Journal of Physical Anthropology*, 151(4), 604-612.

He, P., Zhang, Y., Kim, S. O., Radlanski, R. J., Butcher, K., Schneider, R. A., & DenBesten, P. K. (2010). Ameloblast differentiation in the human developing tooth: effects of extracellular matrices. *Matrix Biology*, 29(5), 411-419.

Helmcke, J. G. (1967). Ultrastructure of enamel. *Structural and Chemical Organization of Teeth*, 2, 135-163.

Hicks, M., & Hicks, A. (2001). *St Gregory's Priory, Northgate, Canterbury: Excavations 1988-1991* (Vol. 2). Canterbury Archaeological Trust Limited.

Hiller, C. R., Robinson, C., & Weatherell, J. A. (1975). Variations in the composition of developing rat incisor enamel. *Calcified Tissue Research*, 18(1), 1-12.

Hillson, S. (1996). *Dental anthropology*. Cambridge University Press.

Hillson, S. (2001). Recording dental caries in archaeological human remains. *International Journal of Osteoarchaeology*, 11(4), 249-289.

Hillson, S., & Bond, S. (1997). Relationship of enamel hypoplasia to the pattern of tooth crown growth: a discussion. *American Journal of Physical Anthropology*, *104*(1), 89-103.

Hlusko, L. J., Suwa, G., Kono, R. T., & Mahaney, M. C. (2004). Genetics and the evolution of primate enamel thickness: a baboon model. *American Journal of Physical Anthropology*, 124(3), 223-233.

Hogg, R. T., Godfrey, L. R., Schwartz, G. T., Dirks, W., & Bromage, T. G. (2015). Lemur biorhythms and life history evolution. *PloS One*, 10(8), e0134210.

Holmen, L., Mejare, I., Malmgren, B., & Thylstrup, A. (1988). The effect of regular professional plaque removal on dental caries in vivo. *Caries Research*, 22(4), 250-256.

Holmen, L., Thylstrup, A., & Årtun, J. (1987). Clinical and histological features observed during arrestment of active enamel carious lesions in vivo. *Caries Research*, 21(6), 546-554.

Holmen, L., Thylstrup, A., Øgaard, B., & Kragh, F. (1985). A polarized light microscopic study of progressive stages of enamel caries in vivo. *Caries Research*, 19(4), 348-354.

Holst, M. (2005). "Artefacts and Environmental Evidence: Human Bone. Summary of Skeletal Analysis". In Spall, C.A. and Toop, N.J. (eds.) Blue Bridge Lane and Fishergate House, York. Report on Excavations: July 2000 to July 2002. Archaeological Planning Consultancy Ltd.

Holt, S. A., Reid, D. J., & Guatelli-Steinberg, D. (2012). Brief communication: premolar enamel formation: completion of figures for aging LEH defects in permanent dentition. *Dental Anthropology Journal*, 25(1), 4-7.

Hooton, E. A. (1925). *The Ancient Inhabitants of the Canary Islands* (Vol. 7). Corinthian Press.

Hoppa, R. D. (1992). Evaluating human skeletal growth: an Anglo-Saxon example. *International Journal of Osteoarchaeology*, 2(4), 275-288.

Horvath, J. E., Ramachandran, G. L., Fedrigo, O., Nielsen, W. J., Babbitt, C. C., Clair, E. M. S., & Wall, C. E. (2014). Genetic comparisons yield insight into the evolution of enamel thickness during human evolution. *Journal of Human Evolution*, 73, 75-87.

Huda, T. F., & Bowman, J. E. (1994). Variation in cross-striation number between striae in an archaeological population. *International Journal of Osteoarchaeology*, 4(1), 49-52.

Huda, T. F., & Bowman, J. E. (1995). Age determination from dental microstructure in juveniles. *American Journal of Physical Anthropology*, *97*(2), 135-150.

Humphrey, L.T. (2010) Weaning behaviour in human evolution. *Seminars in Cell & Developmental Biology*, 21(4), 453-461.

Hylander, W. L. (1975). The human mandible: lever or link?. *American Journal of Physical Anthropology*, 43(2), 227-242.

Ingram, G. S., & Silverstone, L. M. (1981). A chemical and histological study of artificial caries in human dental enamel in vitro. *Caries Research*, 15(5), 393-398.

Islam, A., & Frisch, B. (1985). Plastic embedding in routine histology I: Preparation of semi-thin sections of undecalcified marrow cores. *Histopathology*, 9(12), 1263-1274.

Jay, M. (2009). Breast feeding and weaning behaviour in archaeological populations: evidence from the isotopic analysis of skeletal materials. *Childhood in the Past*, 2(1), 163-178.

Jiang, N., Zhou, J., Chen, M., Schiff, M. D., Lee, C. H., Kong, K., & Mao, J. J. (2014). Postnatal epithelium and mesenchyme stem/progenitor cells in bioengineered amelogenesis and dentinogenesis. *Biomaterials*, 35(7), 2172-2180.

Jolly, C. J. (1970). A new model of hominid differentiation based on a baboon analogy. *Man*, 5(1), 5-26.

Jonckheere, A. R. (1954). A distribution-free k-sample test against ordered alternatives. *Biometrika*, *41*(1/2), 133-145.

Kajiyama, S. (1965). Total number of regular incremental lines (Regulare Parallelstreifen nach Asper) in the enamel of human permanent teeth. *Journal of Nihon University School of Dentistry*, 39, 77-83.

Kay, R. F. (1981). The nut-crackers–a new theory of the adaptations of the Ramapithecinae. *American Journal of Physical Anthropology*, 55(2), 141-151.

Keiter, M. D. (1981). Hand-rearing and development of a Lowland gorilla at Woodland Park Zoo, Seattle. *International Zoo Yearbook*, 21(1), 229-235.

Kelley, J. L., & Swanson, W. J. (2008). Dietary change and adaptive evolution of enamelin in humans and among primates. *Genetics*, 178, 1595-1603.

Kelley, J., & Schwartz, G. T. (2010). Dental development and life history in living African and Asian apes. *Proceedings of the National Academy of Sciences*, 107(3), 1035-1040.

Komai, S., (1942). A study of enamel prism cross striations in human teeth. *J. Jap. Stomatol. Soc. (Kobyo-shi),* 16, 279-292.

Kono, R. T. (2004). Molar enamel thickness and distribution patterns in extant great apes and humans: new insights based on a 3-dimensional whole crown perspective. *Anthropological Science*, 112(2), 121-146.

Kono, R. T., & Suwa, G. (2000). Appearance patterns of molar enamel thickness in tooth sections: analysis based on 3-dimensional data. *Anthropological Science*, 108(1), 102-102.

Kono, R. T., Suwa, G., & Tanijiri, T. (2002). A three-dimensional analysis of enamel distribution patterns in human permanent first molars. *Archives of Oral Biology*, 47(12), 867-875.

Kuykendall, K. L. (1996). Dental development in chimpanzees (Pan troglodytes): the timing of tooth calcification stages. *American Journal of Physical Anthropology*, 99(1), 135-157.

Kuykendall, K. L. (2003). Reconstructing australopithecine growth and development: What do we think we know?. *Cambridge Studies in Biological and Evolutionary Anthropology*, 191-218.

Kuykendall, K. L., & Conroy, G. C. (1996). Permanent tooth calcification in chimpanzees (Pan troglodytes): Patterns and polymorphisms. *American Journal of Physical Anthropology*, 99(1), 159-174.

Lacruz, R. S. (2007). Enamel microstructure of the hominid KB 5223 from Kromdraai, South Africa. *American Journal of Physical Anthropology*, 132(2), 175-182.

Lacruz, R. S., & Bromage, T. G. (2006). Appositional enamel growth in molars of South African fossil hominids. *Journal of Anatomy*, 209(1), 13-20.

Lacruz, R. S., Brookes, S. J., Wen, X., Jimenez, J. M., Vikman, S., Hu, P., & Paine, M. L. (2013). Adaptor protein complex 2–mediated, clathrin-dependent endocytosis, and related gene activities, are a prominent feature during maturation stage amelogenesis. *Journal of Bone and Mineral Research*, 28(3), 672-687

Lacruz, R. S., Dean, M. C., Ramirez-Rozzi, F., & Bromage, T. G. (2008). Megadontia, striae periodicity and patterns of enamel secretion in Plio-Pleistocene fossil hominins. *Journal of Anatomy*, 213(2), 148-158.

Lacruz, R. S., Ramirez-Rozzi, F. R., & Bromage, T. G. (2006). Variation in enamel development of South African fossil hominids. *Journal of Human Evolution*, 51(6), 580-590.

Lacruz, R. S., Smith, C. E., Moffatt, P., Chang, E. H., Bromage, T. G., Bringas, P., & Kurtz, I. (2012). Requirements for ion and solute transport, and pH regulation during enamel maturation. *Journal of Cellular Physiology*, 227(4), 1776-1785.

Lambert, J. E., Chapman, C. A., Wrangham, R. W., & Conklin-Brittain, N. L. (2004). Hardness of cercopithecine foods: implications for the critical function of enamel thickness in exploiting fallback foods. *American Journal of Physical Anthropology*, 125(4), 363-368.

Larsen CS. 1995. Biological changes in human populations with agriculture. *Annual Review of Anthropology*, 24(1), 185–213.

Larsen CS. 1997. Masticatory and nonmasticatory functions: craniofacial adaptation. In: Larsen CS, editor. *Bio archaeology interpreting behavior from the human skeleton*. Cambridge: Cambridge University Press, pp. 226–270.

Lavelle, C:. L. B., Shellis, K. P. and Poole, D. F. G., 1977. *Evolutionary Changes to the Primate Skull and Dentition*. Springfield: Thomas.

Lawn, B. R., & Lee, J. J. (2009). Analysis of fracture and deformation modes in teeth subjected to occlusal loading. *Acta Biomaterialia*, 5(6), 2213-2221.

Le Cabec, A., Tang, N., & Tafforeau, P. (2015). Accessing developmental information of fossil hominin teeth using new synchrotron microtomography-based visualization techniques of dental surfaces and interfaces. *PloS One*, 10(4), e0123019.

Le Luyer, M., & Bayle, P. (2017). Microevolution of outer and inner structures of upper molars in Late Pleistocene and Early Holocene humans. *Comptes Rendus Palevol*, 16(5-6), 632-644.

Le Luyer, M., Rottier, S., & Bayle, P. (2014). Brief communication: Comparative patterns of enamel thickness topography and oblique molar wear in two early neolithic and medieval population samples. *American Journal of Physical Anthropology*, 155(1), 162-172.

Lee, J. J. W., Morris, D., Constantino, P. J., Lucas, P. W., Smith, T. M., & Lawn, B. R. (2010). Properties of tooth enamel in great apes. *Acta Biomaterialia*, 6(12), 4560-4565.

Leigh, Steven R., and Brian T. Shea. (1995). Ontogeny and the evolution of adult body size dimorphism in apes. *American Journal of Primatology.* 36(1): 37-60.

Lester, K. S. (1965). The bands of Schreger: The role of reflexion. *Archives of Oral Biology*, 10(3), 361-IN14.

Lewis, C. (2010). Exploring black holes: Recent investigations in currently occupied rural settlements in Eastern England. *The Landscape Archaeology of Anglo-Saxon England*, 83-106.

Li, C., & Risnes, S. (2004). SEM observations of Retzius lines and prism cross-striations in human dental enamel after different acid etching regimes. *Archives of Oral Biology*, 49(1), 45-52.

Lightfoot, E., O'Connell, T. C., Stevens, R. E., Hamilton, J., Hey, G., & Hedges, R. E. (2009). An investigation into diet at the site of Yarnton, Oxfordshire, using stable carbon and nitrogen isotopes. *Oxford Journal of Archaeology*, 28(3), 301-322.

Littleton, J., & Frohlich, B. (1993). Fish-eaters and farmers: dental pathology in the Arabian Gulf. *American Journal of Physical Anthropology*, 92(4), 427-447.

Liversidge, H. M. (1995). Crown formation times of the permanent dentition and root extension rate in humans. *Aspects of Dental Biology: Palaeontology, Anthropology and Evolution*, 267-275.

Lockwood, C. A., Menter, C. G., Moggi-Cecchi, J., & Keyser, A. W. (2007). Extended male growth in a fossil hominin species. *Science*, 318(5855), 1443-1446.

Lubell, D., Jackes, M., Schwarcz, H., Knyf, M., & Meiklejohn, C. (1994). The Mesolithic-Neolithic transition in Portugal: isotopic and dental evidence of diet. *Journal of Archaeological Science*, 21, 201-201.

Lucas, P. W. (1989). Significance of Mezzettia leptopoda fruits eaten by orang-utans for dental microwear analysis. *Folia Primatologica*, 52(3-4), 185-190.

Lucas, P. W., Constantino, P., Wood, B., & Lawn, B. (2008). Dental enamel as a dietary indicator in mammals. *BioEssays*, 30(4), 374-385.

Lucas, P. W., Corlett, R. T., & Luke, D. A. (1985). Plio-Pleistocene hominid diets: an approach combining masticatory and ecological analysis. *Journal of Human Evolution*, 14(2), 187-202.

Lucas, P. W., Omar, R., Al-Fadhalah, K., Almusallam, A. S., Henry, A. G., Michael, S., & Atkins, A. G. (2013). Mechanisms and causes of wear in tooth enamel: implications for hominin diets. *Journal of the Royal Society Interface*, 10(80), 20120923.

Lucas, P. W., Omar, R., Al-Fadhalah, K., Almusallam, A. S., Henry, A. G., Michael, S., & Atkins, A. G. (2013). Mechanisms and causes of wear in tooth enamel: implications for hominin diets. *Journal of the Royal Society Interface*, 10(80), 20120923.

Lucas, P. W., Peters, C. R., & Arrandale, S. R. (1994). Seed-breaking forces exerted by orang-utans with their teeth in captivity and a new technique for estimating forces produced in the wild. *American Journal of Physical Anthropology*, 94(3), 365-378.

Lucas, P., Constantino, P., Wood, B., & Lawn, B. (2008). Dental enamel as a dietary indicator in mammals. *BioEssays*, 30(4), 374-385.

Lukacs, J. R. (1991). Localized enamel hypoplasia of human deciduous canine teeth: prevalence and pattern of expression in rural Pakistan. *Human Biology*, 513-522.

Lukacs, J. R. (1992). Dental paleopathology and agricultural intensification in South Asia: new evidence from Bronze Age Harappa. *American Journal of Physical Anthropology*, 87(2), 133-150.

Lukacs, J. R. (1999). Enamel hypoplasia in deciduous teeth of great apes: Do differences in defect prevalence imply differential levels of physiological stress?. *American Journal of Physical Anthropology*, 110(3), 351-363.

Lukacs, J. R., & Guatelli-Steinberg, D. (1994). Daughter neglect in India: LEH prevalence and the question of female biological superiority. *American Journal of Physical Anthropology Supplement*, 18, 132.

Lukacs, J. R., & Joshi, M. R. (1992). Enamel hypoplasia prevalence in three ethnic groups of northwest India: A test of daughter neglect and a framework for the past. Recent contributions to the study of enamel developmental defects. *Journal of Paleopathology Monograms Publications*, 2, 359-372.

Lukacs, J. R., & Pal, J. N. (1993). Mesolithic subsistence in North India: inferences from dental attributes. *Current Anthropology*, 34(5), 745-765.

Lukacs, J. R., & Walimbe, S. R. (1998). Physiological stress in prehistoric India: new data on localized hypoplasia of primary canines linked to climate and subsistence change. *Journal of Archaeological Science*, 25(6), 571-585.

Lukacs, J. R., Schultz, M., & Hemphill, B. E. (1985). Dental pathology and dietary patterns in Iron Age northern Pakistan. *South Asian Archaeology*, 475-96.

Macchiarelli, R., Bondioli, L., Debénath, A., Mazurier, A., Tournepiche, J. F., Birch, W., & Dean, M. C. (2006). How Neanderthal molar teeth grew. *Nature*, 444(7120), 748-751.

Macho, G. A. (1994). Variation in enamel thickness and cusp area within human maxillary molars and its bearing on scaling techniques used for studies of enamel thickness between species. *Archives of Oral Biology*, 39(9), 783-792.

Macho, G. A. (2001). Primate molar crown formation times and life history evolution revisited. *American Journal of Primatology*, 55(4), 189-201.

Macho, G. A., & Berner, M. E. (1993). Enamel thickness of human maxillary molars reconsidered. *American Journal of Physical Anthropology*, 92(2), 189-200.

Macho, G. A., & Shimizu, D. (2009). Dietary adaptations of South African australopiths: inference from enamel prism attitude. *Journal of Human Evolution*, 57(3), 241-247.

Macho, G. A., & Thackeray, J. F. (1992). Computed tomography and enamel thickness of maxillary molars of Plio-Pleistocene hominids from Sterkfontein, Swartkrans, and Kromdraai (South Africa): An exploratory study. *American Journal of Physical Anthropology*, 89(2), 133-143.

Macho, G. A., & Wood, B. A. (1995). The role of time and timing in hominid dental evolution. *Evolutionary Anthropology: Issues, News, and Reviews*, 4(1), 17-31.

Macpherson, P. M. (2005). *Tracing Change: an Isotopic Investigation of Anglo-Saxon Childhood Diet* (Doctoral dissertation, University of Sheffield).

Mahoney, P. (2008). Intraspecific variation in M1 enamel development in modern humans: implications for human evolution. *Journal of Human Evolution*, 55(1), 131-147.

Mahoney, P. (2010). Two-dimensional patterns of human enamel thickness on deciduous (dm1, dm2) and permanent first (M1) mandibular molars. *Archives of Oral Biology*, 55(2), 115-126.

Mahoney, P. (2011). Human deciduous mandibular molar incremental enamel development. *American Journal of Physical Anthropology*, 144(2), 204-214.

Mahoney, P. (2012). Incremental enamel development in modern human deciduous anterior teeth. *American Journal of Physical Anthropology*, 147(4), 637-651.

Mahoney, P. (2013). Testing functional and morphological interpretations of enamel thickness along the deciduous tooth row in human children. *American Journal of Physical Anthropology*, 151(4), 518-525.

Mahoney, P. (2015). Dental fast track: prenatal enamel growth, incisor eruption, and weaning in human infants. *American Journal of Physical Anthropology*, *156*(3), 407-421.

Mahoney, P., Miszkiewicz, J. J., Pitfield, R., Schlecht, S. H., Deter, C., & Guatelli-Steinberg, D. (2016). Biorhythms, deciduous enamel thickness, and primary bone growth: a test of the Havers-Halberg Oscillation hypothesis. *Journal of Anatomy*, 228(6), 919-928.

Mahoney, P., Smith, T. M., Schwartz, G. T., Dean, C., & Kelley, J. (2007). Molar crown formation in the late Miocene Asian hominoids, Sivapithecus parvada and Sivapithecus indicus. *Journal of Human Evolution*, 53(1), 61-68.

Mahoney, Patrick, Justyna J. Miszkiewicz, Rosie Pitfield, Chris Deter, and Debbie Guatelli-Steinberg (2017). Enamel biorhythms of humans and great apes: the Havers-Halberg Oscillation hypothesis reconsidered. *Journal of Anatomy* 230(2), 272-281.

Mann, A. E. (1975). *Some paleodemographic aspects of the South African australopithecines* (No. 1). Dept. of Anthropology, University of Pennsylvania.

Mann, A. E., Monge, J. M., & Lampl, M. (1991). Investigation into the relationship between perikymata counts and crown formation times. *American Journal of Physical Anthropology*, 86(2), 175-188.

Mann, A., Lampl, M., & Monge, J. (1990). Patterns of ontogeny in human evolution: Evidence from dental development. *American Journal of Physical Anthropology*, 33(S11), 111-150.

Martin, L. (1985). Significance of enamel thickness in hominoid evolution. *Nature*, 314(6008), 260-263.

Martin, L. B. (1983). *The Relationships of the Later Miocene Hominoidea* (Doctoral dissertation, University of London).

Martin, L. B., Olejniczak, A. J., & Maas, M. C. (2003). Enamel thickness and microstructure in pitheciin primates, with comments on dietary adaptations of the middle Miocene hominoid Kenyapithecus. *Journal of Human Evolution*, 45(5), 351-367.

Martin, L. B., Olejniczak, A. J., & Maas, M. C. (2003). Enamel thickness and microstructure in pitheciin primates, with comments on dietary adaptations of the middle Miocene hominoid Kenyapithecus. *Journal of Human Evolution*, 45(5), 351-367.

Martin, S. A., Guatelli-Steinberg, D., Sciulli, P. W., & Walker, P. L. (2008). Brief communication: comparison of methods for estimating chronological age at linear enamel formation on anterior dentition. *American Journal of Physical Anthropology,* 135(3), 362-365.

Massler, M., & Schour, I. (1946). Growth of the child and the calcification pattern of the teeth. *American Journal of Orthodontics and Oral Surgery*, 32(9), 495-517.

Mays, S., & Beavan, N. (2012). An investigation of diet in early Anglo-Saxon England using carbon and nitrogen stable isotope analysis of human bone collagen. *Journal of Archaeological Science*, 39(4), 867-874.

Mays, S., Elders, J., Humphrey, L., White, W., & Marshall, P. (2013). *Science and the dead: a guideline for the destructive sampling of archaeological human remains for scientific analysis*. English Heritage Publishing with the Advisory Panel on the Archaeology of Burials in England.

McFarlane, G., Littleton, J., & Floyd, B. (2014). Estimating striae of Retzius periodicity nondestructively using partial counts of perikymata. *American Journal of Physical Anthropology*, 154(2), 251-258.

McGraw, W. S., Pampush, J. D., & Daegling, D. J. (2012). Brief communication: Enamel thickness and durophagy in mangabeys revisited. *American Journal of Physical Anthropology*, 147(2), 326-333.

McKinley, J. I., Leivers, M., Schuster, J., & Marshall, P. (2015). *Cliffs End Farm Isle of Thanet, Kent: A mortuary and ritual site of the Bronze Age, Iron Age and Anglo-Saxon period with evidence for long-distance maritime mobility*. Wessex Archaeology.

McWhirr, A., Viner, L., & Wells, C. (1982). Romano-British Cemeteries at Cirencester: Cirencester Excavations II. *Cirencester Excavation Committee. Cirencester*.

Millard, L., Jarman, S., & Hawkes, S. C. (1969). *Anglo-Saxon Burials Near the Lord of the Manor, Ramsgate: New Light on the Site of Ozengell?* Headley Bros.

Miszkiewicz, J. J. (2015). Linear enamel hypoplasia and age-at-death at medieval (11th–16th Centuries) St. Gregory's Priory and Cemetery, Canterbury, UK. *International Journal of Osteoarchaeology*, *25*(1), 79-87.

Molnar S. 1972. Tooth wear and culture: a survey of tooth functions among some prehistoric populations. *Current Anthropology*, 13, 511–526

Molnar, S., & Gantt, D. G. (1977). Functional implications of primate enamel thickness. *American Journal of Physical Anthropology*, 46(3), 447-454.

Molnar, S., Hildebolt, C., Molnar, I. M., Radovcic, J., & Gravier, M. (1993). Hominid enamel thickness: I. The Krapina neandertals. *American Journal of Physical Anthropology*, 92(2), 131-138.

Montgomery, J., Evans, J. A., Powlesland, D., & Roberts, C. A. (2005). Continuity or colonization in Anglo-Saxon England? Isotope evidence for mobility, subsistence practice, and status at West Heslerton. *American Journal of Physical Anthropology*, 126(2), 123-138.

Moorrees, C. F., Fanning, E. A., & Hunt Jr, E. E. (1963). Age variation of formation stages for ten permanent teeth. *Journal of Dental Research*, 42(6), 1490-1502.

Mukherjee, K., Ruan, Q., Nutt, S., Tao, J., De Yoreo, J. J., & Moradian-Oldak, J. (2018). Peptide-based bioinspired approach to regrowing multilayered aprismatic enamel. *ACS Omega*, 3(3), 2546-2557.

Müldner, G. (2013). Stable isotopes and diet: their contribution to Romano-British research. *Antiquity*, 87(335), 137-149.

Mummery, J. H. (1924). *The microscopic & general anatomy of the teeth: human and comparative*. H. Milford.

Nanci, A. (2003). Enamel: composition, formation, and structure. In: *Oral Histology, Development Structure and Function*. Mosby Year Book, Inc., St. Louis, Missouri, pp. 145-191

Nanci, A. and Smith. C. E. (1992). Development and calcification of enamel. In: *Calcification in Biological Systems*. CRC Press, Boca Raton, pp. 313-343

Needham, A.E. (1964). *The Growth Process in Animals*, pp. 522. London: Pitman 1964

Nelson, D. G. A., Wefel, J. S., Jongebloed, W. L., & Featherstone, J. D. B. (1987). Morphology, histology and crystallography of human dental enamel treated with pulsed low-energy infrared laser radiation. *Caries Research*, 21(5), 411-426.

Neville, A. C. (1967). Daily growth layers in animals and plants. *Biological Reviews*, 42(3), 421-439.

Newell-Morris, L., & Sirianni, J. E. (1982). Parameters of bone growth in the fetal and infant macaque (Macaca nemestrina) humerus as documented by trichromatic bone labels. *Progress in Clinical and Biological Research*, 101, 243-258.

Newman, H. N., & Levers, B. G. H. (1979). Tooth eruption and function in an early Anglo-Saxon population. *Journal of the Royal Society of Medicine*, 72(5), 341-350.

Newman, H. N., & Poole, D. F. G. (1974). Observations with scanning and transmission electron microscopy on the structure of human surface enamel. *Archives of Oral Biology*, 19(12), 1135-1143.

Nyström, M., & Ranta, H. (2003). Tooth formation and the mandibular symphysis during the first five postnatal months. *Journal of Forensic Sciences*, 48(2), 373-378.

Okada, M. (1943). Hard tissues of animal body. Highly interesting details of Nippon studies in periodic patterns of hard tissues are described. *Shanghai Evening Post. Medical Edition of September 1943*, pp. 15-31

Olejniczak, A. J., Smith, T. M., Feeney, R. N., Macchiarelli, R., Mazurier, A., Bondioli, L., & Radovčić, J. (2008a). Dental tissue proportions and enamel thickness in Neandertal and modern human molars. *Journal of Human Evolution*, *55*(1), 12-23.

Olejniczak, A. J., Smith, T. M., Skinner, M. M., Grine, F. E., Feeney, R. N., Thackeray, J. F., & Hublin, J. J. (2008b). Three-dimensional molar enamel distribution and thickness in Australopithecus and Paranthropus. *Biology Letters*, 4(4), 406-410.

Olejniczak, A. J., Tafforeau, P., Feeney, R. N., & Martin, L. B. (2008). Three-dimensional primate molar enamel thickness. *Journal of Human Evolution*, 54(2), 187-195.

Owen, R. (1840). Odontography; or a treatise on the comparative anatomy of the teeth: their physiological relations, mode of development, and microscopic structure in the vertebrate animals. 2. *Atlas*. Baillière.

Pampush, J. D., Duque, A. C., Burrows, B. R., Daegling, D. J., Kenney, W. F., & McGraw, W. S. (2013). Homoplasy and thick enamel in primates. *Journal of Human Evolution*, 64(3), 216-224.

Pan, L., Dumoncel, J., De Beer, F., Hoffman, J., Thackeray, J. F., Duployer, B., & Braga, J. (2016). Further morphological evidence on South African earliest Homo lower postcanine dentition: Enamel thickness and enamel dentine junction. *Journal of Human Evolution*, 96, 82-96.

Pate, F. D. (1994). Bone chemistry and paleodiet. *Journal of Archaeological Method and Theory*, 1(2), 161-209.1: 161

Pérez-Pérez, A., Espurz, V., de Castro, J. M. B., de Lumley, M. A., & Turbón, D. (2003). Non-occlusal dental microwear variability in a sample of Middle and Late Pleistocene human populations from Europe and the Near East. *Journal of Human Evolution*, 44(4), 497-513

Primeau, C., Arge, S. O., Boyer, C., & Lynnerup, N. (2015). A test of inter-and intra-observer error for an atlas method of combined histological data for the evaluation of enamel hypoplasia. *Journal of Archaeological Science: Reports*, 2, 384-388.

Privat, K. L., O'connell, T. C., & Richards, M. P. (2002). Stable isotope analysis of human and faunal remains from the Anglo-Saxon cemetery at Berinsfield, Oxfordshire: dietary and social implications. *Journal of Archaeological Science*, 29(7), 779-790.

Rando, J. C., Cabrera, V. M., Larruga, J. M., Hernández, M., González, A. M., Pinto, F., & Bandelt, H. J. (1999). Phylogeographic patterns of mtDNA reflecting the colonization of the Canary Islands. *Annals of Human Genetics*, 63(5), 413-428.

Redfern, R. C., Hamlin, C., & Athfield, N. B. (2010). Temporal changes in diet: a stable isotope analysis of late Iron Age and Roman Dorset, Britain. *Journal of Archaeological Science*, 37(6), 1149-1160.

Reid, D. J., & Dean, M. C. (2000). Brief communication: the timing of linear hypoplasias on human anterior teeth. *American Journal of Physical Anthropology*, 113(1), 135-139.

Reid, D. J., & Dean, M. C. (2006). Variation in modern human enamel formation times. *Journal of Human Evolution*, 50(3), 329-346.

Reid, D. J., & Ferrell, R. J. (2006). The relationship between number of striae of Retzius and their periodicity in imbricational enamel formation. *Journal of Human Evolution*, 50(2), 195-202.

Reid, D. J., Beynon, A. D., & Ramirez Rozzi, F. V. R. (1998a). Histological reconstruction of dental development in four individuals from a medieval site in Picardie, France. *Journal of Human Evolution*, 35(4-5), 463-477.

Reid, D. J., & Guatelli-Steinberg, D. (2017). Updating histological data on crown initiation and crown completion ages in southern Africans. *American Journal of Physical Anthropology*, *162*(4), 817-829.

Reid, D. J., Guatelli-Steinberg, D., & Walton, P. (2008a). Variation in modern human premolar enamel formation times: Implications for Neandertals. *Journal of Human Evolution*, 54(2), 225-235.

Reid, D. J., Schwartz, G. T., Dean, C., & Chandrasekera, M. S. (1998b). A histological reconstruction of dental development in the common chimpanzee, Pan troglodytes. *Journal of Human Evolution*, 35(4-5), 427-448.

Reitsema, L. J. (2013). Beyond diet reconstruction: stable isotope applications to human physiology, health, and nutrition. *American Journal of Human Biology*, 25(4), 445-456.

Retzius, A. (1837). Bemerkungen über den inneren Bau der Zähne. mit besonderer Rücksicht auf dem in Zahnknochen vorkommenden Röhrenbau. (*Müllers*) *Archive of Anatomy and Physiology*, *1837*, 486-566.

Reynard, L. M., & Tuross, N. (2015). The known, the unknown and the unknowable: weaning times from archaeological bones using nitrogen isotope ratios. *Journal of Archaeological Science*, 53, 618-625.

Ripa, L. W., Gwinnett, A. J., & Buonocore, M. G. (1966). The “prismless” outer layer of deciduous and permanent enamel. *Archives of oral biology*, 11(1), 41IN3-48IN5.

Risnes, S. (1986). Enamel apposition rate and the prism periodicity in human teeth. *European Journal of Oral Sciences*, 94(5), 394-404.

Risnes, S. (1990). Structural characteristics of staircase-type retzius lines in human dental enamel analyzed by scanning electron microscopy. *The Anatomical Record*, 226(2), 135-146.

Risnes, S. (1998). Growth tracks in dental enamel. *Journal of Human Evolution*, *35*(4), 331-350.

Robinson, C., Brookes, S. J., Shore, R. C., & Kirkham, J. (1998). The developing enamel matrix: nature and function. *European Journal of Oral Sciences*, 106(1), 282-291.

Robinson, C., Kirkham, J., Brookes, S. J., Bonass, W. A., & Shore, R. C. (2003). The chemistry of enamel development. *International Journal of Developmental Biology*, 39(1), 145-152.

Robinson, J.T. (1956). The dentition of the Australopithecinae. *Annals of the Transvaal Museum*, 9,1-179.

Robson, S. L., & Wood, B. (2008). Hominin life history: reconstruction and evolution. *Journal of Anatomy*, 212(4), 394-425.

Robson, S. L., Van Schaik, C. P., & Hawkes, K. (2006). The derived features of human life history. *The Evolution of Human Life History*, 17-44.

Rönnholm, E. (1962). The amelogenesis of human teeth as revealed by electron microscopy: II. The development of the enamel crystallites. *Journal of Ultrastructure Research*, 6(3-4), 249-303.

Rose, J. C. (1977). Defective enamel histology of prehistoric teeth from Illinois. *American Journal of Physical Anthropology*, 46(3), 439-446.

Rose, J. C., Armelagos, G. J., & Lallo, J. W. (1978). Histological enamel indicator of childhood stress in prehistoric skeletal samples. *American Journal of Physical Anthropology*, 49(4), 511-516.

Rozzi, F. R. (1998). Can enamel microstructure be used to establish the presence of different species of Plio-Pleistocene hominids from Omo, Ethiopia?. *Journal of Human Evolution*, 35(4), 543-576.

Rozzi, F. V. R., & de Castro, J. M. B. (2004). Surprisingly rapid growth in Neanderthals. *Nature*, 428(6986), 936-939.

Rozzi, F. V. R., Bromage, T., & Schrenk, F. (1997). UR 501, the Plio-Pleistocene hominid from Malawi. Analysis of the microanatomy of the enamel. *Comptes Rendus de l'Académie des Sciences-Series IIA-Earth and Planetary Science*, 325(3), 231-234.

Saunders, S. R., Chan, A. H., Kahlon, B., Kluge, H. F., & FitzGerald, C. M. (2007). Sexual dimorphism of the dental tissues in human permanent mandibular canines and third premolars. *American Journal of Physical Anthropology*, 133(1), 735-740.

Scheuer, L., & Black, S. (2004). *The Juvenile Skeleton*. Academic Press.

Scheving, L. E., & Pauly, J. E. (1974). Circadian rhythms: some examples and comments on clinical application. *Chronobiologia*, 1(1), 3.

Schour, I. (1936). The Neonatal Line in the Enamel and Dentin of the Human Deciduous Teeth and First Permanent Molar From the Department of Histology, College of Dentistry, University of Illinois. Presented in the form of a discussion and demonstration before the Ninth International Dental Congress of the FDI, Vienna. *The Journal of the American Dental Association,* 23(10), 1946-1955.

Schour, I., & Hoffman, M. M. (1939a). Studies in Tooth Development: I. the 16 Microns Calcification Rhythm in the Enamel and Dentin from Fish to Man. *Journal of Dental Research*, 18(1), 91-102.

Schour, I., & Hoffman, M. M. (1939b). Studies in tooth development: II. The rate of apposition of enamel and dentin in man and other mammals. *Journal of Dental Research*, 18(2), 161-175.

Schour, I., & Poncher, H. G. (1937). Rate of apposition of enamel and dentin, measured by the effect of acute fluorosis. *American Journal of Diseases of Children*, 54(4), 757-776.

Schultz, A. H. (1960). *Age Changes in Primates and their Modification in Man*. Pergamon press.

Schwartz, G. T. (2000a). Enamel thickness and the helicoidal wear plane in modern human mandibular molars. *Archives of Oral Biology*, 45(5), 401-409.

Schwartz, G. T. (2000b). Taxonomic and functional aspects of the patterning of enamel thickness distribution in extant large-bodied hominoids. *American Journal of Physical Anthropology*, 111(2), 221-244.

Schwartz, G. T., & Dean, C. (2001). Ontogeny of canine dimorphism in extant hominoids. *American Journal of Physical Anthropology*, 115(3), 269-283.

Schwartz, G. T., & Dean, M. C. (2005). Sexual dimorphism in modern human permanent teeth. *American Journal of Physical Anthropology*, 128(2), 312-317.

Schwartz, G. T., Liu, W., & Zheng, L. (2003). Preliminary investigation of dental microstructure in the Yuanmou hominoid (Lufengpithecus hudienensis), Yunnan Province, China. *Journal of Human Evolution*, 44(2), 189-202.

Schwartz, G. T., Mahoney, P., Godfrey, L. R., Cuozzo, F. P., Jungers, W. L., & Randria, G. F. (2005). Dental development in Megaladapis edwardsi (Primates, Lemuriformes): implications for understanding life history variation in subfossil lemurs. *Journal of Human Evolution*, 49(6), 702-721.

Schwartz, G. T., Reid, D. J., & Dean, C. (2001). Developmental aspects of sexual dimorphism in hominoid canines. *International Journal of Primatology*, 22(5), 837-860.

Schwartz, G. T., Reid, D. J., Dean, M. C., & Zihlman, A. L. (2006). A faithful record of stressful life events recorded in the dental developmental record of a juvenile gorilla. *International Journal of Primatology*, 27(4), 1201-1219.

Schwartz, G. T., Samonds, K. E., Godfrey, L. R., Jungers, W. L., & Simons, E. L. (2002). Dental microstructure and life history in subfossil Malagasy lemurs. *Proceedings of the National Academy of Sciences*, 99(9), 6124-6129.

Schwartz, G. T., Thackeray, J. F., Reid, C., & Van Reenan, J. F. (1998). Enamel thickness and the topography of the enamel–dentine junction in South African Plio-Pleistocene hominids with special reference to the Carabelli trait. *Journal of Human Evolution*, 35(4-5), 523-542.

Schwartz, G.T., Reid, D., Dean, C. (2001). Developmental aspects of sexual dimorphism in hominoid canines. *International Journal of Primatology*, 22, 837-860

Schwidetzky, I. (1963). *Die vorspanische Bevölkerung der Kanarischen Inseln: anthropologische Untersuchungen*(Vol. 1). Musterschmidt.

Scull, C. (1993). Archaeology, early Anglo-Saxon society and the origins of Anglo-Saxon kingdoms. *Anglo-Saxon Studies in Archaeology and History*, 6(1), 65-82.

Sealy, J. C., Patrick, M. K., Morris, A. G., & Alder, D. (1992). Diet and dental caries among later stone age inhabitants of the Cape Province, South Africa. *American Journal of Physical Anthropology*, 88(2), 123-134.

Senut, B., Pickford, M., Gommery, D., Mein, P., Cheboi, K., & Coppens, Y. (2001). First hominid from the Miocene (Lukeino formation, Kenya). *Comptes Rendus de l'Académie des Sciences-Series IIA-Earth and Planetary Science*, 332(2), 137-144.

Shellis, R. P. (1984). Relationship between human enamel structure and the formation of caries-like lesions in vitro. *Archives of Oral Biology*, 29(12), 975-981.

Shellis, R. P. (1998). Utilization of periodic markings in enamel to obtain information on tooth growth. *Journal of Human Evolution*, 35(4-5), 387-400.

Shellis, R. P., & Poole, D. F. G. (1977). The calcified dental tissues of primates. *Evolutionary changes to the primate skull and dentition. Springfield: Charles C. Thomas,* pp. 197-279.

Shellis, R. P., Beynon, A. D., Reid, D. J., & Hiiemae, K. M. (1998). Variations in molar enamel thickness among primates. *Journal of Human Evolution*, 35(4), 507-522.

Shinoda, H. (1984). Faithful records of biological rhythms in dental hard tissues. *Chemistry Today*, 162, 34-40.

Silverstone, L. M., & Poole, D. F. G. (1968). The effect of saliva and calcifying solutions upon the histological appearance of enamel caries. *Caries Research*, 2(1), 87-96.

Simmer, J. P., & Fincham, A. G. (1995). Molecular mechanisms of dental enamel formation. *Critical Reviews in Oral Biology & Medicine*, 6(2), 84-108.

Simmer, J. P., Papagerakis, P., Smith, C. E., Fisher, D. C., Rountrey, A. N., Zheng, L., & Hu, J. C. (2010). Regulation of dental enamel shape and hardness. *Journal of Dental Research*, 89(10), 1024-1038.

Simons, E. L, & Pilbeam, DR (1972) Hominoid paleoprimatology. In: *The Functional and Evolutionary Biology of Primates*. Chicago: Aldine.

Simons, E. L. (1976). The nature of the transition in the dental mechanism from pongids to hominids. *Journal of Human Evolution*, 5(5), 511-528.

Simons. E. L. (1961). The phyletic position of Ramapithecus. *Postilla (Yale Peabody Museum)*, 57, 1-9

Skinner, M. F., & Hung, J. T. W. (1989). Social and biological correlates of localized enamel hypoplasia of the human deciduous canine tooth. *American Journal of Physical Anthropology*, 79(2), 159-175.

Skinner, M. M., Alemseged, Z., Gaunitz, C., & Hublin, J. J. (2015). Enamel thickness trends in Plio-Pleistocene hominin mandibular molars. *Journal of Human Evolution*, 85, 35-45.

Skinner, M. M., Evans, A., Smith, T., Jernvall, J., Tafforeau, P., Kupczik, K., & Toussaint, M. (2010). Brief communication: Contributions of enamel-dentine junction shape and enamel deposition to primate molar crown complexity. *American Journal of Physical Anthropology*, 142(1), 157-163.

Skinner, M., & Anderson, G. S. (1991). Individualization and enamel histology: a case report in forensic anthropology. *Journal of Forensic Science*, 36(3), 939-948.

Skobe, Z. (1976). The secretory stage of amelogenesis in rat mandibular incisor teeth observed by scanning electron microscopy. *Calcified Tissue Research*, 21(1), 83-103.

Skobe, Z., Stern, D. N., & Prostak, K. S. (2017). The cell biology of amelogenesis. In *Dental Enamel Formation to Destruction*, pp. 23-57, CRC Press.

Smith, B. H. (1984). Patterns of molar wear in hunter–gatherers and agriculturalists. *American Journal of Physical Anthropology*, 63(1), 39-56.

Smith, B. H. (1989a). Dental development as a measure of life history in primates. *Evolution*, 43(3), 683-688.

Smith, B. H. (1991). Dental development and the evolution of life history in Hominidae. *American Journal of Physical Anthropology*, 86(2), 157-174.

Smith, B. H. (1994). Patterns of dental development in Homo, Australopithecus, Pan, and Gorilla. *American Journal of Physical Anthropology*, 94(3), 307-325.

Smith, B. H., Crummett, T. L., & Brandt, K. L. (1994). Ages of eruption of primate teeth: a compendium for aging individuals and comparing life histories. *American Journal of Physical Anthropology*, 37(S19), 177-231.

Smith, C. E. (1998). Cellular and chemical events during enamel maturation. *Critical Reviews in Oral Biology & Medicine*, 9(2), 128-161.

Smith, C. E., & Nanci, A. (2003). Overview of morphological changes in enamel organ cells associated with major events in amelogenesis. *International Journal of Developmental Biology*, 39(1), 153-161.

Smith, P., & Zilberman, U. (1994). Thin enamel and other tooth components in Neanderthals and other hominids. *American Journal of Physical Anthropology*, 95(1), 85-87.

Smith, T. M. (2016). Dental development in living and fossil orangutans. *Journal of Human Evolution*, 94, 92-105.

Smith, T. M., Bacon, A. M., Demeter, F., Kullmer, O., Nguyen, K. T., De Vos, J., & Zhao, L. (2011). Dental tissue proportions in fossil orangutans from mainland Asia and Indonesia. *Human Origins Research*, 1(1), 1.

Smith, T. M., Harvati, K., Olejniczak, A. J., Reid, D. J., Hublin, J. J., & Panagopoulou, E. (2009). Brief communication: dental development and enamel thickness in the Lakonis Neanderthal molar. *American Journal of Physical Anthropology*, 138(1), 112-118.

Smith, T. M., Kupczik, K., Machanda, Z., Skinner, M. M., & Zermeno, J. P. (2012). Enamel thickness in Bornean and Sumatran orangutan dentitions. *American Journal of Physical Anthropology*, 147(3), 417-426.

Smith, T. M., Martin, L. B., & Leakey, M. G. (2003). Enamel thickness, microstructure and development in Afropithecus turkanensis. *Journal of Human Evolution*, 44(3), 283-306.

Smith, T. M., Martin, L. B., Reid, D. J., de Bonis, L., & Koufos, G. D. (2004). An examination of dental development in Graecopithecus freybergi (= Ouranopithecus macedoniensis). *Journal of Human Evolution*, 46(5), 551-577.

Smith, T. M., Olejniczak, A. J., Kupczik, K., Lazzari, V., De Vos, J., Kullmer, O., & Tafforeau, P. (2009). Taxonomic assessment of the Trinil molars using non-destructive 3D structural and development analysis. *Paleoanthropology*, *2009*, 117-129.

Smith, T. M., Olejniczak, A. J., Martin, L. B., & Reid, D. J. (2005). Variation in hominoid molar enamel thickness. *Journal of Human Evolution*, 48(6), 575-592.

Smith, T. M., Olejniczak, A. J., Reh, S., Reid, D. J., & Hublin, J. J. (2008). Brief communication: enamel thickness trends in the dental arcade of humans and chimpanzees. *American Journal of Physical Anthropology,*136(2), 237-241.

Smith, T. M., Olejniczak, A. J., Reid, D. J., Ferrell, R. J., & Hublin, J. J. (2006a). Modern human molar enamel thickness and enamel–dentine junction shape. *Archives of Oral Biology*, 51(11), 974-995

Smith, T. M., Olejniczak, A. J., Tafforeau, P., Reid, D. J., Grine, F. E., & Hublin, J. J. (2006b). Molar crown thickness, volume, and development in South African Middle Stone Age humans. *South African Journal of Science*, 102(11-12), 513-517.

Smith, T. M., Olejniczak, A. J., Zermeno, J. P., Tafforeau, P., Skinner, M. M., Hoffmann, A., & Moggi-Cecchi, J. (2012). Variation in enamel thickness within the genus Homo. *Journal of Human Evolution*, 62(3), 395-411.

Smith, T. M., Reid, D. J., Dean, M. C., Olejniczak, A. J., & Martin, L. B. (2007a). Molar development in common chimpanzees (Pan troglodytes). *Journal of Human Evolution*, 52(2), 201-216.

Smith, T. M., Reid, D. J., Dean, M. C., Olejniczak, A. J., & Martin, L. B. (2007b). New perspectives on chimpanzee and human molar crown development. In: *Dental perspectives on human evolution: state of the art research in dental paleoanthropology*. Springer, Dordrecht, pp. 177-192.

Smith, T. M., & Tafforeau, P. (2008). New visions of dental tissue research: tooth development, chemistry, and structure. *Evolutionary Anthropology: Issues, News, and Reviews*, *17*(5), 213-226.

Smith, T. M., Tafforeau, P., Le Cabec, A., Bonnin, A., Houssaye, A., Pouech, J., & Menter, C. G. (2015). Dental ontogeny in pliocene and early pleistocene hominins. *PLoS One*, 10(2), e0118118.

Smith, T. M., Tafforeau, P., Reid, D. J., Grün, R., Eggins, S., Boutakiout, M., & Hublin, J. J. (2007a). Earliest evidence of modern human life history in North African early Homo sapiens. *Proceedings of the National Academy of Sciences*, 104(15), 6128-6133.

Smith, T. M., Tafforeau, P., Reid, D. J., Pouech, J., Lazzari, V., Zermeno, J. P., & Makaremi, M. (2010). Dental evidence for ontogenetic differences between modern humans and Neanderthals. *Proceedings of the National Academy of Sciences*, 107(49), 20923-20928.

Smith, T. M., Toussaint, M., Reid, D. J., Olejniczak, A. J., & Hublin, J. J. (2007c). Rapid dental development in a middle Paleolithic Belgian Neanderthal. *Proceedings of the National Academy of Sciences*, 104(51), 20220-20225.

Sorenti, M., Martinón-Torres, M., Martín-Francés, L., & Perea-Pérez, B. (2019). Sexual dimorphism of dental tissues in modern human mandibular molars. *American Journal of Physical Anthropology*.

Sponheimer, M., Passey, B. H., De Ruiter, D. J., Guatelli-Steinberg, D., Cerling, T. E., & Lee-Thorp, J. A. (2006). Isotopic evidence for dietary variability in the early hominin Paranthropus robustus. *Science*, 314(5801), 980-982.

Stoodley, N. (2000). From the cradle to the grave: age organization and the early Anglo-Saxon burial rite. *World Archaeology*, 31(3), 456-472.

Strait, D. S., Constantino, P., Lucas, P. W., Richmond, B. G., Spencer, M. A., Dechow, P. C., & Weber, G. W. (2013). Viewpoints: diet and dietary adaptations in early hominins: the hard food perspective. *American Journal of Physical Anthropology*, 151(3), 339-355.

Strait, D. S., Weber, G. W., Neubauer, S., Chalk, J., Richmond, B. G., Lucas, P. W., & Grosse, I. R. (2009). The feeding biomechanics and dietary ecology of Australopithecus africanus. *Proceedings of the National Academy of Sciences*, 106(7), 2124-2129.

Stroud, J. L., Buschang, P. H., & Goaz, P. W. (1994). Sexual dimorphism in mesiodistal dentin and enamel thickness. *Dentomaxillofacial Radiology*, 23(3), 169-171.

Suga, S., & Gustafson, G. (1962). Studies on the development of rat enamel by means of histochemistry, microradiography and polarized light microscopy. *Archives of Oral Biology*, 7, 223-244.

Sunderland, E. P., Smith, C. J., & Sunderland, R. (1987). A histological study of the chronology of initial mineralization in the human deciduous dentition. *Archives of Oral Biology*, 32(3), 167-174.

Suwa, G., & Kono, R. T. (2005). A micro-CT based study of linear enamel thickness in the mesial cusp section of human molars: reevaluation of methodology and assessment of within-tooth, serial, and individual variation. *Anthropological Science*, 113(3), 273-289.

Swales, D. L. M. (2012). *Life Stress: A Bio-cultural Investigation into the Later Anglo-Saxon Population of the Black Gate Cemetery* (Doctoral dissertation, University of Sheffield).

Swindler, D. R. (1961). Calcification of the permanent first mandibular molar in rhesus monkeys. *Science*, 134(3478), 566-566.

Swindler, D. R., & Beynon, A. D. (2005). 13 The development and microstructure of the dentition of Theropithecus. *Theropithecus: The Rise and Fall of a Primate Genus*, 351.

Sykes, N. (2000). *The Norman Conquest: A Zooarchaeological Perspective* (Doctoral dissertation, University of Southampton).

Tagiguchi, H. (1966). Chronologic relationship of human tooth crown formation. *Nihon University Dental Journal*, 40, 391-397.

Teaford, M. F., & Ungar, P. S. (2000). Diet and the evolution of the earliest human ancestors. *Proceedings of the National Academy of Sciences*, 97(25), 13506-13511.

Thylstrup, A., Fejerskov, O., & Larsen, M. J. (1976). Polarized light microscopy of enamel structure in incisors from newborn infants. *European Journal of Oral Sciences*, 84(5), 243-254.

Ulhaas, L., Henke, W., & Rothe, H. (1999). Variation in molar enamel thicknss of the gener “cercopithecus” and “colobus”. *Anthropologie (1962-)*, 265-271.

Ungar, P. S., & Hlusko, L. J. (2016). The evolutionary path of least resistance. *Science*, 353(6294), 29-30.

Ungar, P. S., & Sponheimer, M. (2011). The diets of early hominins. *Science*, 334(6053), 190-193.

Vangala, M. R., Rudraraju, A., & Subramanyam, R. V. (2016). Mounting ground sections of teeth: Cyanoacrylate adhesive versus Canada balsam. *Journal of Oral and Maxillofacial Pathology*, 20(1), 20.

Viciano, J., López-Lázaro, S., & Alemán, I. (2013). Sex estimation based on deciduous and permanent dentition in a contemporary Spanish population. *American Journal of Physical Anthropology*, 152(1), 31-43.

Vogel, E. R., van Woerden, J. T., Lucas, P. W., Atmoko, S. S. U., van Schaik, C. P., & Dominy, N. J. (2008). Functional ecology and evolution of hominoid molar enamel thickness: Pan troglodytes schweinfurthii and Pongo pygmaeus wurmbii. *Journal of Human Evolution*, 55(1), 60-74.

Walker, P. L., & Erlandson, J. M. (1986). Dental evidence for prehistoric dietary change on the northern Channel Islands, California. *American Antiquity*, 51(2), 375-383.

Weatherell, J. A., Robinson, C., & Hallsworth, A. S. (1974). Variations in the chemical composition of human enamel. *Journal of Dental Research*, 53(2), 180-192.

Weber, D. F., & Ashrafi, S. H. (1979). Structure of retzius lines in partially demineralized human enamel. *The Anatomical Record*, 194(4), 563-569.

Weber, D. F., & Glick, P. L. (1975). Correlative microscopy of enamel prism orientation. *Developmental Dynamics*, 144(4), 407-419.

White, T. D., Black, M. T., & Folkens, P. A. (2011). *Human Osteology*. Academic press.

Williams, H. (2004). Potted histories–cremation, ceramics and social memory in early Roman Britain. *Oxford journal of archaeology*, *23*(4), 417-427.

Wilson, D. F., & Shroff, F. R. (1970). The nature of the striae of Retzius as seen with the optical microscope. *Australian Dental Journal*, 15(3), 162-171.

Wilson, P. R., & Beynon, A. D. (1989). Mineralization differences between human deciduous and permanent enamel measured by quantitative microradiography. *Archives of Oral Biology*, 34(2), 85-88.

Witzel, C., Kierdorf, U., Schultz, M., & Kierdorf, H. (2008). Insights from the inside: histological analysis of abnormal enamel microstructure associated with hypoplastic enamel defects in human teeth. *American Journal of Physical Anthropology*, 136(4), 400-414.

Wright, J. T., Carrion, I. A., & Morris, C. (2015). The molecular basis of hereditary enamel defects in humans. *Journal of Dental Research*, 94(1), 52-61.

Wright, L. E., & Schwarcz, H. P. (1998). Stable carbon and oxygen isotopes in human tooth enamel: identifying breastfeeding and weaning in prehistory. *American Journal of Physical Anthropology*, 106(1), 1-18.

Xing, S., Guatelli-Steinberg, D., O'Hara, M., Wu, X., Liu, W., & Reid, D. J. (2015). Perikymata distribution in Homo with special reference to the Xujiayao juvenile. *American Journal of Physical Anthropology*, 157(4), 684-693.

Zanolli, C., Dean, C., Rook, L., Bondioli, L., Mazurier, A., & Macchiarelli, R. (2016). Enamel thickness and enamel growth in Oreopithecus: Combining microtomographic and histological evidence. *Comptes Rendus Palevol*, 15(1-2), 209-226.

Zheng, J., Li, Y., Shi, M. Y., Zhang, Y. F., Qian, L. M., & Zhou, Z. R. (2013). Microtribological behaviour of human tooth enamel and artificial hydroxyapatite. *Tribology International*, 63, 177-185.

Zihlman, A., Bolter, D., & Boesch, C. (2004). Wild chimpanzee dentition and its implications for assessing life history in immature hominin fossils. *Proceedings of the National Academy of Sciences*, 101(29), 10541-10543.

Zilberman, U., & Smith, P. (1992). A comparison of tooth structure in Neanderthals and early Homo sapiens sapiens: a radiographic study. *Journal of Anatomy*, 180(3), 387.

Zilberman, U., Skinner, M., & Smith, P. (1992). Tooth components of mandibular deciduous molars of Homo sapiens sapiens and Homo sapiens neanderthalensis: a radiographic study. *American Journal of Physical Anthropology*, 87(3), 255-262.

Zuckerman, S. (1928). Age-changes in the Chimpanzee, with special reference to Growth of Brain, Eruption of Teeth, and Estimation of Age; with a Note on the Taungs Ape. *Journal of Zoology*, 98(1), 1-42.

APPENDIX A: GLOSSARY

A.1 Glossary of relevant terms

A.1.1 Directional and surface definitions

Dental and histological analysis involves the observation of both two and three dimensional features. Directional terms are therefore a key part of their analysis. The following terms are those most commonly used within dental histology (see Fig A.1; Beek, 1983):

Lingual – this refers to the surface or edge of a tooth facing the inwards within the jaw, towards the tongue.

Labial – the labial surface is that opposite to the lingual surface facing outwards. Only anterior teeth (incisors and premolars) have a labial surface.

Buccal – the buccal surface of the tooth is descriptively the same as the labial surface, but refers only to the posterior premolar and molar teeth.

Mesial – all teeth have a mesial surface which faces towards the median plane passing equidistant between the first incisors.

Distal – the distal surface of a tooth lies directly opposite to the mesial surface away from the median line.

Occlusal – the occlusal surface is that which makes contact between maxillary and mandibular teeth during mastication. In incisors (and occasionally canines) this is often also referred to as the incisal surface.

Apex – the furthest point from the dental cervix, usable in reference to both enamel and dentine.

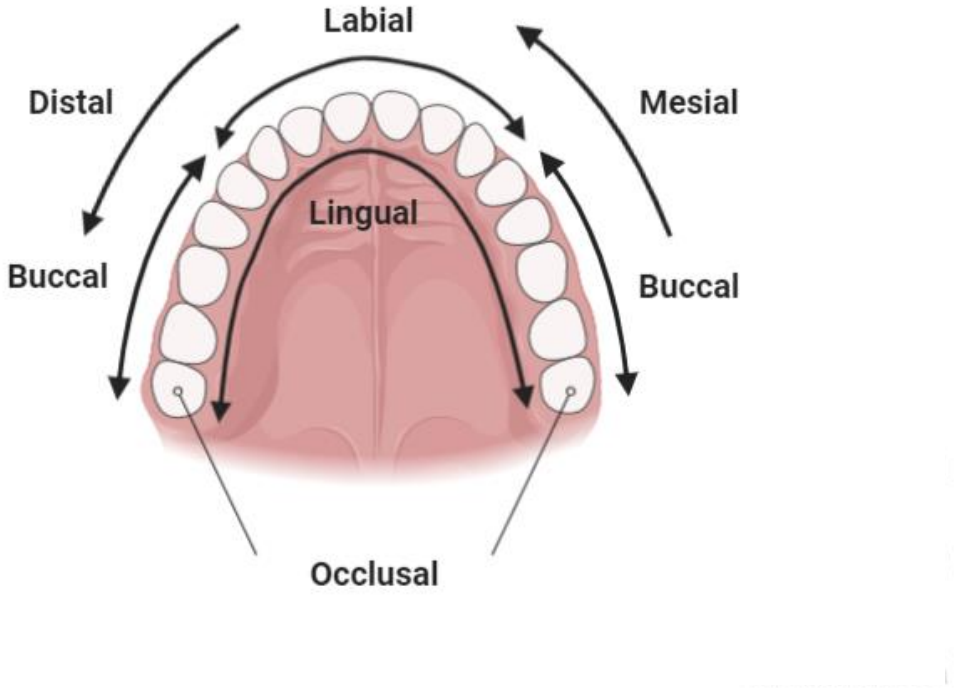

Fig. A.1. *Figure of directional and surface terms used in this project. Image produced using the BioRender tool set (*https://biorender.com/*).*

A.1.2 Numbering and descriptive systems

Within text tooth type will be referenced using Latin abbreviations (Beek, 1983:6), tooth side via use of "L" and "R", and tooth position (upper/lower) and number (where needed) though the use subscript and superscript (see Table A.1).

Table A.1. *Acronyms and devices used to refer to teeth in the shorthand.*

Tooth Types	
Tooth name	**Abbreviation**
Incisor	I
Canine	C
Molar	M
Tooth Siding	
Tooth side	**Abbreviation**
Left	L
Right	R
Tooth Position	
Tooth position	**Abbreviation**
First	1
Second	2
Third	3
Upper (for molars and incisors)	Superscript of number
Lower (for molars and incisors)	Subscript of number
Upper (for canines)	U
Lower (for canines)	L

Example: RM^2 would stand for upper right second molar

Example: ULC would stand for upper left canine

A.1.3 Enamel specific abbreviations and acronyms

EDJ – Enamel dentine junction: the visible line where the enamel cap adheres to the outermost dentine.

DSR – Daily enamel secretion rate: refers to the quantified aspect of measuring the daily rate of enamel matrix secretion.

RP – Retzius periodicity: the measure, in days, of the circa-septum rhythm producing long period lines within dental enamel.

EER – Enamel extension rate: the rate of differentiation among ameloblasts initiating as the dentine horn and culminating at the dental cervix.

iEER – Initial enamel extension rate: a preliminary calculation displaying the rate at which the earliest ameloblasts differentiate along the EDJ.

AET – Average enamel thickness: refer to the average thickness of enamel observable and calculated using a two-dimensional dental cross section.

RET – Relative enamel thickness: the relative measure of AET calculated when accounting for the area of dentine encapsulated by the enamel cap.

CT – Cuspal thickness: the linear thickness of enamel between the dentine horn and the occlusal surface of the same cusp tip.

BCT – Buccal cuspal thickness: a molar specific variant of CT measured at a buccal cusp.

BLT – Buccal lateral thickness: the maximum linear thickness measurable within the buccal-lateral imbricational enamel between the EDJ and the enamel occlusal surface

CEFT – Cuspal enamel formation time: the time taken for the morphology of appositional enamel to have formed, measured in days.

LEFT – Lateral enamel formation time: the time taken for the morphology of imbricational enamel to have formed, measured in days.

CFT – Crown formation time: the time taken for the morphology of a dental crown to have formed, measured in days. The sum of CEFT and LEFT.

A.2 Dental anatomy

The clinical anatomy of any tooth is comprised of a number of organic and inorganic components:

Crown – the crown is the most naturally visible, portion of the tooth which is directly involved in mastication (see Fig A.2; Berkovitz et al., 2002; White et al., 2011). Within the crown exists the enamel-dentine junction (EDJ), the region where the internal enamel borders the primary dentine (Berkovitz et al., 2002). Externally the crown possesses the cemento-enamel junction (CEJ), where the surface of the enamel meets the cementum surrounding the root dentine of the tooth (White et al., 2011:104).

Root – the root is the portion of the tooth which acts to anchor the tooth within the alveolar bones of the jaw (see Fig A.2; Berkovitz et al., 2002; White et al., 2011). Typically a tooth will have between one and three roots according to its type and location within the jaw (White et al., 2011:104). Within a dental root is found the root canal, secondary dentine, and cementum (Berkovitz et al., 2002; White et al., 2011:104).

Dental Pulp – a soft tissue which supports the dentinal structures of a tooth comprised of nerves and blood vessels which provide a blood supply (Berkovitz et al., 2002; White et al., 2011). Dental pulp is unique within the tooth as it can actively produce dentine in reaction to toxins (Berkovitz et al., 2002). The majority of the dental pulp is contained within the inner area of the crown and is referred to as the pulp cavity (see Fig A.2; Berkovitz et al., 2002). The remaining dental pulp compromises the root canals, where the nerve and blood flow enters the root through the apical foramina and travel to the pulp cavity (Berkovitz et al., 2002; White et al., 2011:106).

Cervix (or neck) – the cervical region of the tooth is the constricted region where the crown and root(s) meet (see Fig A.2; Berkovitz et al., 2002; White et al., 2011:104). Regardless of the number of roots a tooth only ever has a single cervix. The majority of the cervix is defined by the external CEJ (White et al., 2011:104). This is the line which encircles the cervix at the border between the enamel of the crown and the cementum of the root (White et al., 2011:104).

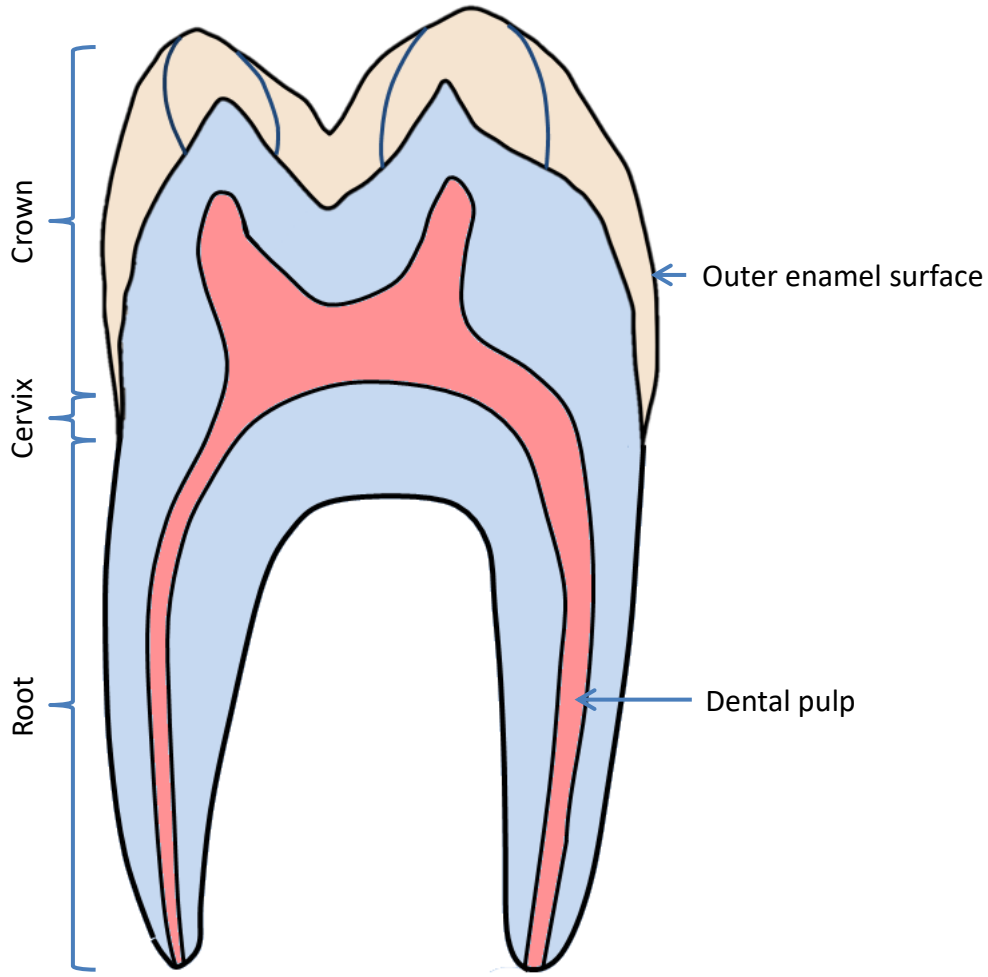

Fig. A.2. *Two-dimensional cross-sectional diagram of a human permanent molar, displaying all major anatomical features compromising the internal structures of dentition. Beige areas denote enamel, blue denotes dentine, and red denotes dental pulp. The outer black line of the enamel represents the outer enamel surface.*

Enamel – a mineralised tissue which creates a thick layer over the primary dentine (see Fig A.3; Berkovitz et al., 2002). Although brittle, enamel is the hardest biological substance which comprises the human body, acting as the primary substance involved in mastication (its specific function varying between tooth types; Simmer and Fincham, 1995; Berkovitz et al., 2002; Wright et al., 2015). It is formed of between 90-92% hydroxyapatite crystallites and between 10-8% organic protein material and water (Berkovitz et al., 2002).

Appositional enamel – the earlier forming region of enamel, referred to as cuspal enamel, which is observed to have formed before the first Retzius line to develop in contact with both the EDJ and outer enamel surface (see Fig A.3; Berkovitz et al., 2002). Appositional enamel is found at the cusp tips of dental enamel.

Imbricational enamel – the later forming enamel, referred to as lateral enamel, which is observed to have formed after the first Retzius line to develop in contact with both the EDJ and outer enamel surface (see Fig A.3; Berkovitz et al., 2002). Imbricational enamel also include cervical enamel which is found in near proximity to the dental cervix (Berkovitz et al., 2002).

Dentine – a hard but elastic tissue which makes up the highest percentage mass of any tooth (see Fig A.3 and/or Antoine et al., 2018). The tooth is comprised of two types of dentine: primary dentine which forms deep to the enamel of the tooth crown, and secondary dentine which forms the roots (Berkovitz et al., 2002; White et al., 2011:104; Antoine et al., 2018). Dentine is relatively soft compared to enamel, formed of between 47-50% hydroxyapatite and 50-53% organic material and water (Berkovitz et al., 2002; White et al., 2011:104; Antoine et al., 2018).

Cementum – a mineralised tissue which thinly coats the roots of teeth (see Fig A.3). It acts to adhere the tooth within the alveolar bones of the jaw (Berkovitz et al., 2002; White et al., 2011:106). Softer than dentine, cementum is comprised of between 45-50% inorganic substances and 50-55% organic material and water (Berkovitz et al., 2002; White et al., 2011:106).

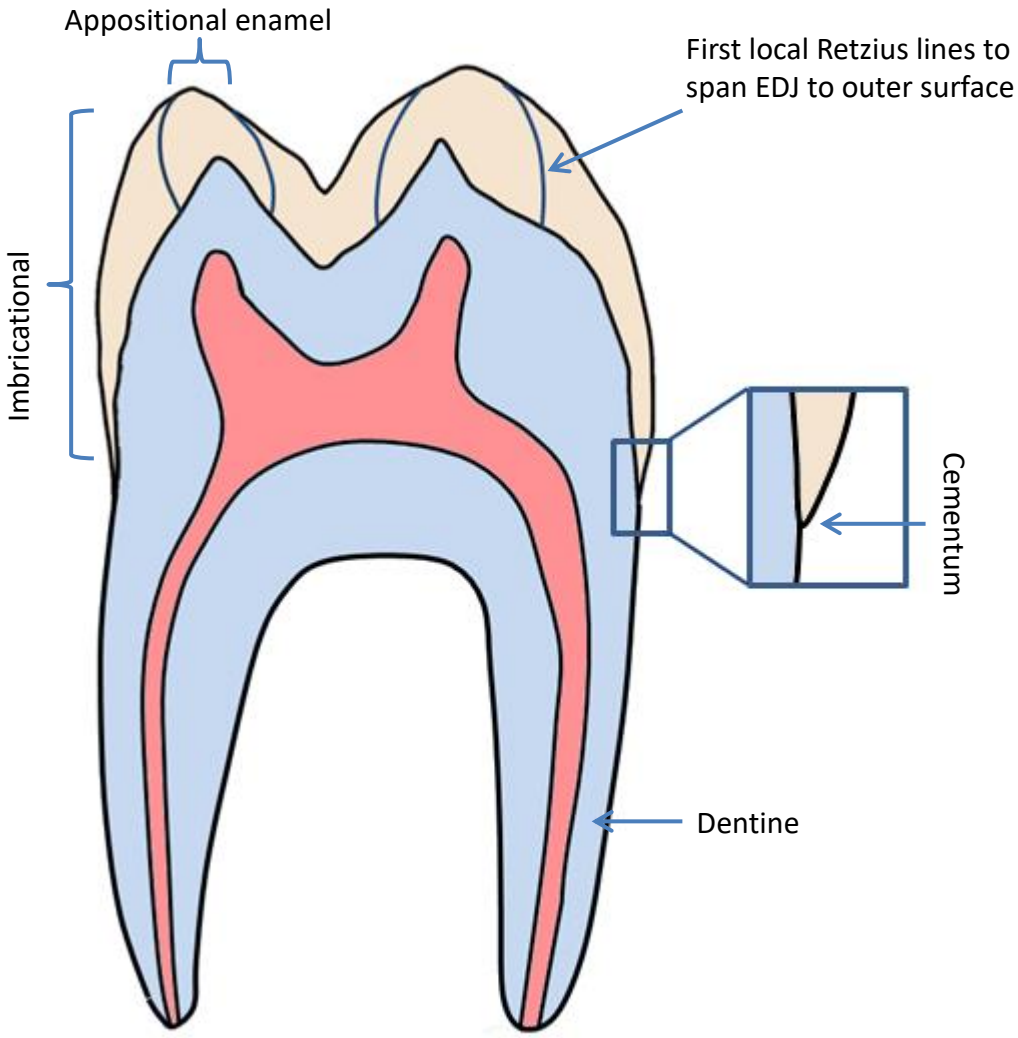

Fig. A.3. *Two-dimensional cross-sectional diagram of a human permanent molar, displaying relevant secondary anatomical features associated with the structures of dentition. Within the beige areas dark blue lines represent the Reztius lines used to differentiate appositional and Imbricational enamel, blue denotes dentine, and red denotes dental pulp. The right superimposition highlights the location of dental cementum.*

A.3 Tooth types

Morphologically, adult human dentition starting at the midline, moving distally, is divided into four tooth types: incisors, canines, premolars, and molars (see Fig A.4; White et al., 2011:104). Each of these has a different function in the process of mastication and has dimorphism to accommodate for its role (Beek, 1983; White et al., 2011). It should be noted that all descriptions here refer to permanent dentition.

Incisors – the incisors are the most medial teeth within the jaw developing (Beek, 1983; White et al., 2011:110). Each adult human possesses eight permanent incisors (four upper and four lower). Maxillary incisors tend to be bigger than their mandibular counterparts, with the first incisors also being larger than their second (or lateral) neighbours (Beek, 1983; White et al., 2011:110). This is particularly evident in the maxillary teeth. Incisors have a shovel-like appearance (see Fig A.4), allowing them to cut and slice food during mastication (White et al., 2011:110). Incisors are also typical of a tapering root and a flat crown that widens on the mesio-distal plane. This widening is more exaggerated in the maxillary teeth (Beek, 1983; White et al., 2011:110).

Canines – the canines are the third tooth (observed laterally) developing distally to the second incisor (Beek, 1983; White et al., 2011:111). Each adult human possesses four permanent canines (two upper and two lower). Maxillary canines tend to be larger than the mandibular but not to such an extreme degree as incisors (Beek, 1983). Canines have an exaggerated incisive edge on either side which forms a single pointed cusp; the lateral incisal edge is more prominent than the mesial (see Fig A.4; Beek, 1983; White et al., 2011:110). This cusp acts to grip and tear food during mastication and develops an extremely long root to provide anchorage within the jaw (White et al., 2011:110).

Molars – permanent molars have the largest occlusal surfaces of any tooth type (Beek, 1983). First molars, within both the mandible and maxilla, develop the largest occlusal surface, with decreasing surface areas moving distally along the jaw in subsequent second and third molars (Beek, 1983; White et al., 2011:119-120). The first maxillary molar possesses four cusps (two buccal and two lingual) and three roots (two buccal and one lingual) (see Fig A.4); the mandibular equivalent develops five cusps (two buccal, two lingual and one distal) and two roots (one buccal and one lingual) (Beek, 1983; White et al., 2011:119-120).

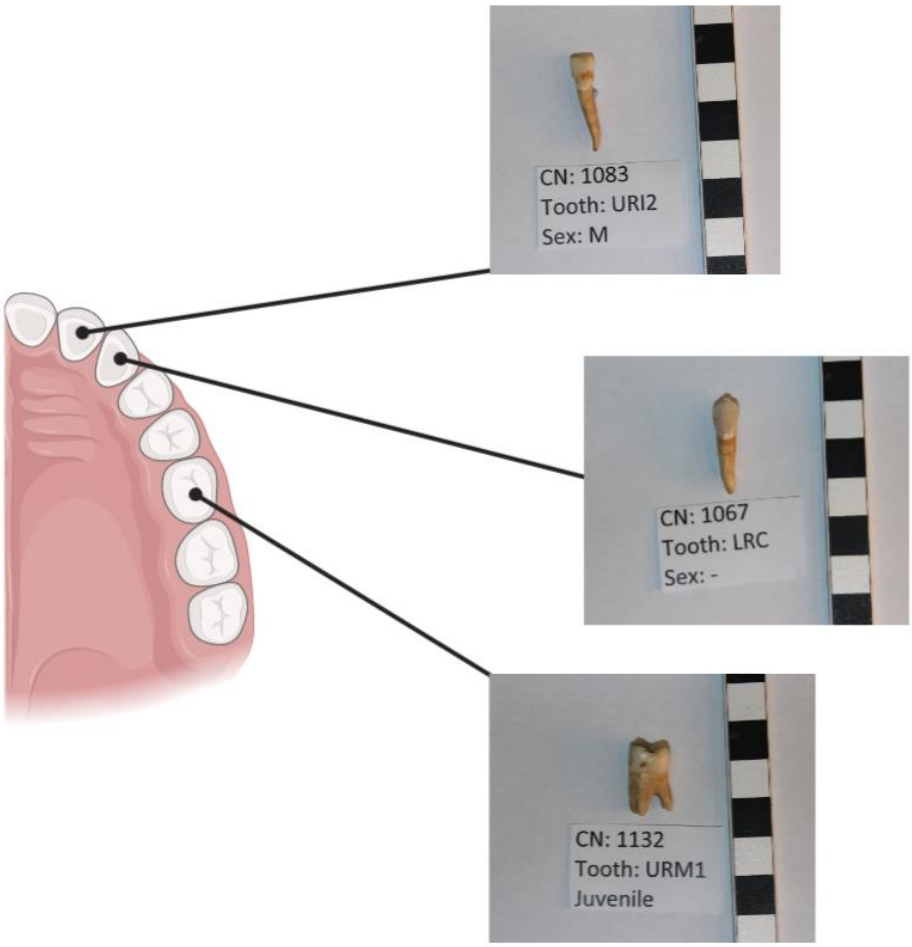

Fig. A.4. *Figure of the location and appearance of the tooth types researched in the project: incisor (top right), canine (mid right), and molar (bottom right). Photographic images taken from teeth collected from the Fishergate House Medieval collection (see Chapter 5: Materials and Methods). Digital imagery produced using the BioRender tool set (*https://biorender.com/*).*

APPENDIX B: SEX DETERMINATION SHEET

Table B.1 *Sheet template for sex determination from features of the pelvis. Each feature was given a score of between 1-5 (1 = definitely female; 2 = likely female; 3 = indeterminate; 4 likely male; 5 = definitely male).*

Morphological Characteristic	Pelvis	
	Right	Left
Overall Structure		
Overall Shape (anterior aspect)		
Pelvic Inlet		
Iliac Crest (vertical aspect)		
Iliac Blade (anterior aspect)		
Iliac Tuberosity		
Greater Sciatic Notch		
Auricular Surface		
Pereauricular Sulcus		
Postauricular Space		
Pubic Symphysis Height		
Pubic Rami		
Sub-pubic Angle		
Pubic Tubercle		
Inferior Pubic Ramus		
Ventral Arc		
Sub-pubic Concavity		
Medial Ischio-pubic Ridge		
Obturator Foramen		
Ischial Tuberosity		
Ischial Spine		
Anterior Sacral Curvature		
Sacral Auricular Surface		
Overall Sex (side)		
Overall Sex (total)		

Table B.2 *Sheet template for sex determination from features of the skull. Each feature was given a score of between 1-5 (1 = definitely female; 2 = likely female; 3 = indeterminate; 4 likely male; 5 = definitely male).*

Morphological Characteristic	Skull	
	Right	Left
Frontal Tuberosities		
Zygomatic Process of Frontal		
Temporal Ridges		
Suprameatal Crests		
Mastoid Process		
Nuchal Area		
External Occipital Protuberance		
Occipital Condyles		
Canine Emminence		
Mandibular Ramus (anterior-posterior)		
Mandibular Ramus		
Depth from Incisors to Mentum		
Mental Protuberance		
Lower Margin of Mandibular Corpus		
Angle of Mandible		
Overall Sex		

APPENDIX A: PUBLISHED ARTICLES

The two articles presented in the subsequent pages represent the published versions of Chapter 5 and Chapter 6 of this thesis. The citations for these articles, in order, are:

Aris, C., Mahoney, P., & Deter, C. (2020a). Enamel thickness and growth rates in modern human permanent first molars over a 2000 year period in Britain. *American Journal of Physical Anthropology*.

Aris, C., Mahoney, P., O'Hara, M. C., & Deter, C. (2020b). Enamel growth rates of anterior teeth in males and females from modern and ancient British populations. *American Journal of Physical Anthropology*.

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