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Tattling teeth.

Occurrence of enamel hypoplasia in a 19th century Dutch rural agricultural population.

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Preface/ acknowledgements

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Introduction.

Historical events like epidemics, outbreaks of disease, and wars leave traces, physical and mental, on the people that are involved. These periods of stress are known from written sources, but another important source are the skeletal remains from the individuals who experienced these events. By studying skeletal assemblages, we can obtain data about health, disease and death in past populations. We can learn about the people behind the great stories, the ordinary people who probably were never mentioned in the history books. One way to study the health of past populations is by assessing a defect formed in the teeth called enamel hypoplasia. Enamel hypoplasia is highly correlated with stress episodes and is thus a good indicator of a person's childhood health.

Enamel hypoplasia is a defect that has less than the normal amount of enamel. The missing enamel can results in small dents, grooves, pits, or entire areas of missing enamel. Enamel hypoplasia is seen as a non-specific stress marker, so it is not associated with a specific cause. Many factors can cause enamel hypoplasias, including nutritional deficiencies, infectious disease, metabolic disruption, and trauma (Aufderheide and Rodriguez-Martin 1998; Roberts and Manchester 2010). When the body is under stress, it will focus on the process that are essential for survival and less energy will go to nonvital processes (like enamel formation). Arrests in tooth crown are the skeletal evidence for the survival of these stress events. (Auferdheide and Rodriguez-Martin 1998).

The relationship between enamel hypoplasia and general health makes this an interesting topic. It can provide clues about nutritional status, presence of infectious diseases, trauma, or cultural activities of past populations. The examination of enamel hypoplasia has advantages over other indicators of developmental disruptions. Since enamel does not remodel, it provides a permanent record of developmental disruptions during childhood (Aufderheide and Rodriguez-Martin 1998). The physiological disruptions that cause enamel hypoplasia affect only the portion of the crown that is in the process of forming at that moment. by determining the specific location of the defects, it is possible to

reconstruct the age at which the defect was formed. In this way it is possible to reconstruct chronologies of stressful events in individuals.

Knowledge of the types of disease and dietary deficiencies which can cause dental enamel hypoplasias comes both from experimental work on animals and from studies of human populations. Enamel hypoplasia is seen as a stress-marker, it is difficult to find the specific cause that led to the dental lesion.

After years of research, there are still a lot of uncertainties on this topic.
Some studies (Goodman and Capasso 1992; Enwonwu 1973) have shown a direct association between enamel hypoplasias and socio-economic status. Other researchers (Dummer et al. 1986; Dobney and Goodman 1991; Nation et al. 1987) found the opposite. Several studies found a higher prevalence of LEH in boys than in girls (Palubeckaite et al. 2002; Saunders and Keenleyside 1999; Slaus 2008; Van Gerven et al. 1990) and some of the studies found a higher frequency of LEH in females (Goodman et al. 1987; Slaus 2000). Research has also been done on the tooth types. Some teeth seem to be more susceptible for dental lesions then others (Marks and Rose 1985; Goodman and Rose 1990; Goodman and Armelagos 1985). Stodder (1997) found that individuals who died as subadults (< 21 years) showed a higher frequency of LEH than the ones who survived to adulthood. This is supported by other research (Duray 1996; Goodman et al. 1988; Palubeckaité 2001). Some previous studies (Duray 1996; Slaus 2000; Stodder 1997) demonstrated that individuals with LEH had a significant lower age-at-death than the ones without LEH. There are some studies (Saunders and Keenleyside 1999; Palubeckaité 2001) who also found almost no difference in the mean age-at-death of the individuals with and without LEH. Saunders and Keenleyside (1999) interpreted this as that LEH is a stress marker that does not have an impact on mortality selection.

The age at which the defect was formed can be estimated from the location of the defect. Permanent tooth enamel forms from the age of one to the age of 11 (Hillson 1996). Several schemes (Goodman and Armelagos 1980; Swardstedt 1966; Goodman and Rose 1990) have been published indicating the relationship between the location of a defect and the individuals age at formation. The most

accurate method is published by Reid and Dean (2006) and will be used in this research.
The measurement of the height of the defect can then be correlated to the individuals age to determine the year when the stress occurred. A connection between the records and year can then provide us with further detail about the types of stresses affecting the population during that time.

This research will focus on the occurrence of linear enamel hypoplasia in an early 19th century Dutch rural agricultural population, the Beemster polder. Important research questions are: is the prevalence of linear enamel hypoplasia different in males and females? And what are the causes for this difference? Are there correlations between the presence/absence of linear enamel hypoplasia and the different age-categories? What could have caused these difference? Is there a difference in the mean age-at-death for individuals with and without LEH. At what age does the presence of LEH peak in this population? Is it possible to link these events with a cause. Answering these questions will give an idea about the overall childhood health from this community. Is it possible to use this stress-marker on teeth to link this agricultural population to documented phenomena in the Early Modern Period? Was the population well-buffered from historically documented stress episodes?

The teeth of the Beemster population will be examined macroscopically to determine the presence or absence of enamel hypoplasia. Macroscopic analysis of linear enamel hypoplasia has several advantages. Teeth are well preserved in archaeological contexts which make them ideal to examine. Dental enamel does not remodel, so enamel can be seen as an archive for developmental stress markers, which never fade. Researching linear enamel hypoplasia can be done with only visual observation and sliding calipers. And last of all, this technique is a non-destructive method.

Non-specific stress-markers are the response of human skeletons to environmental stress. These stress-markers can be used to investigate the childhood health of these individuals. This research will focus on a 19th century, Dutch, rural agricultural population. What can we say about the general health of these

individuals? How well was this population buffered against the stress periods from this century?

Chapter 1: Background

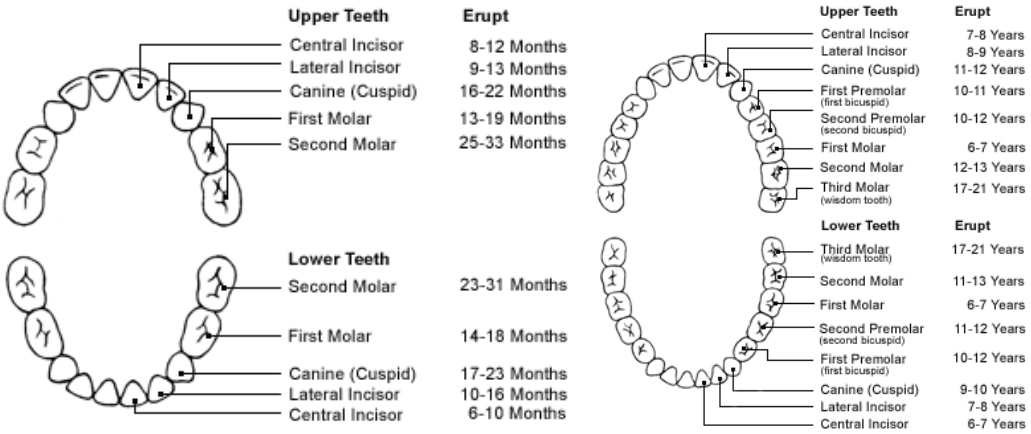
In order to understand the nature of the formation of enamel defects a brief overview of tooth development is provided.

1.1. Teeth

1.1.1. Dentition

Humans have two sets of teeth in their lifetime, deciduous and permanent (Figure 1.1). The deciduous dentition is almost half-formed by birth and erupts during the next two years (Hillson 1996). The deciduous dentition contains eight incisors, four canines and eight molars (Cobourne and DiBiase 2010). The deciduous dentition is gradually replaced by the permanent dentition. The first permanent tooth starts to form just before birth, and the last one is completed at 18 to 25 years of age. The final four permanent teeth that emerge are the third molars, also called "wisdom" teeth. They erupt around age 18 to early twenties. Sometimes these teeth are congenitally absent (Hillson 1996).

Figure 1.1: Deciduous and permanent teeth.



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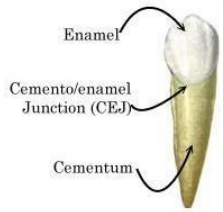
1.1.2. Tissues

Each tooth exists of a crown and root portion (Figure 1.2). The crown and root join at the cementoenamel junction (CEJ). This junction, also called the cervical line is visible on every teeth. The crown is covered with enamel and the root portion is covered with cementum (Hillson 1996).

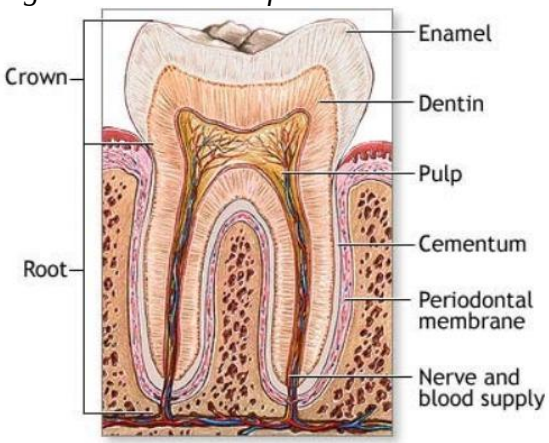
Figure 1.2: Tooth.



http://www.proprofs.com/flashcards/cardshowall.php?title=dental-anatomy-unit-1-2

Human teeth are composed of four tissues: the soft tissue (pulp) and three calcified tissues: dentin, enamel, and cementum (Figure 1.3). The crown consists of two layers: an outer layer of enamel and an inner layer of dentin. Dental enamel is the hardest tissue in the human body. Enamel is the outer structure that covers the tooth crown. Dentin forms the core of the tooth. Dentin lies under the enamel of the crown. Cementum covers the tooth root. (Cobourne and DiBiase 2010; Hillson 1996).

Figure 1.3: Section of a tooth.



http://www.ida.org.in/Information/Stuctureofmonth.aspx?type=stuctureofmonth&name=cell2&name3=cell002&Hname=td2

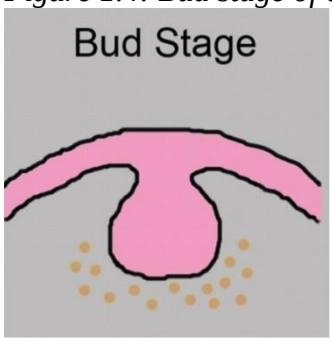
1.1.3. Tooth development

Tooth development is a complex process by which teeth form from embryonic cells. This process is divided in four steps: bud stage, cap stage, bell stage and crown stage or maturation. The different stages are not well defined because they are part of a continuous process (Slootweg 2010).

- Bud stage

When a fetus is around six weeks old, the epithelium of the mandible and maxilla are already thickening in preparation for what is going to happen. The epithelium creates an island of cells, which is the origin of the new tooth. This is the dental lamina (Figure 1.4). The dental lamina is surrounded by ectomesenchymal cells. Ten round epithelial structures, buds, develop at the distal aspect of the dental lamina of the upper and lower jaw. These correspond to the 20 deciduous teeth (Cobourne and DiBiase 2010; Slootweg 2010).

Figure 1.4: Bud stage of a tooth.



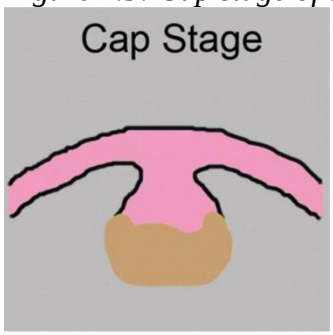
http://radiographics.rsna.org/content/26/6/1751.figures-only

- Cap stage

The tooth bud grows around the ectomesenchymal cells, and will take the form of a cap. This cap is referred to as the enamel organ (Figure 1.5). This enamel organ will produce enamel. Ectomesenchymal cells lying in this cap form the dental papilla and will become the dental pulp, which is the soft tissue core of the teeth. The ectomesenchymal cells that surround the enamel organ will form the dental follicle. This follicle will produce cementum, parts of the alveolar socket and the periodontal ligaments. Three different components can be distinguished now: (1) the inner enamel epithelium, (2) the outer enamel epithelium and (3) and the

epithelium that is called the stellate reticulum (Cobourne and DiBiase 2010; Slootweg 2010).

Figure 1.5: Cap stage of a tooth



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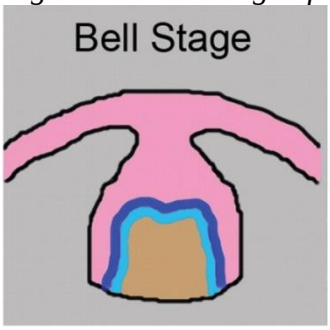
- Bell stage

The cells on the periphery of the enamel organ separate into three important layers. The first layer is the outer enamel epithelium, that is formed by the cells on the periphery of the dental organ. The second layer is the stratum intermedium and the third layer is the inner enamel epithelium, formed by the cells of the enamel organ. (Figure 1.6). The inner enamel epithelium cells will develop into enamel-forming ameloblasts while the dental papilla cells will develop into dentin-producing odontoblasts (Cobourne and DiBiase 2010; Slootweg 2010).

Figure 1.6: Bell stage of a tooth.



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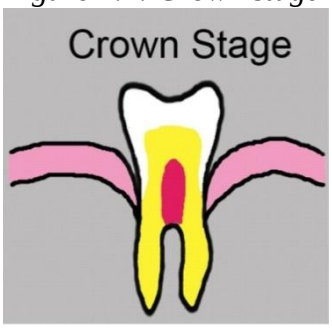
- Crown stage

During this stage hard tissues mineralize. The inner enamel epithelial cells move closer to the stratum intermedium and away from the dental papilla. The layer of cells in the dental papilla increase in size and become the odontoblasts, the cells

that form dentin. These two processes continue to form the tips of the cusps. The odontoblasts start forming dentin, which is an organic matrix. After the formation of dentin begins, the cells of the inner enamel epithelium start forming enamel, also an organic matrix. Enamel formation moves outwards, so adding new enamel to the outer surface of the developing tooth (Figure 1.7) (Cobourne and DiBiase 2010; Slootweg 2010).

Figure 1.7: Crown stage of a tooth



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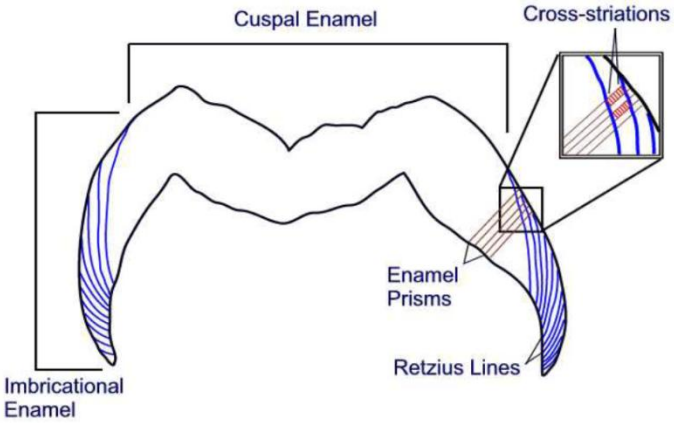
The permanent teeth develop slowly under the deciduous teeth. Once the crown of the permanent teeth is formed, the roots begins to grow. This causes an increase in the pressure on the deciduous tooth and part of the deciduous tooth root is broken down. The tooth will become loose and eventually falls out. The permanent tooth will grow and fills in the place of the deciduous teeth.

Figure 1.8: (Transverse) section of enamel



http://www.eva.mpg.de/evolution/pdf/DentalMicrostructure.pdf

Enamel formation is called amelogenesis and occurs in the crown stage of tooth development. Enamel formation occurs in two stages: the secretory and maturation stage. In the secretory stage, ameloblasts form enamel matrix. Ameloblasts deposit enamel alongside the dentin, at the location of what becomes the cusps of the teeth. Enamel formation then continues outward, away from the center of the tooth. During the maturation stage, the function of ameloblasts changes from enamel production to transportation of substances. This substance is mostly proteins. The enamel is completely mineralized at the end of this stage (Hillson 1996).

Mmicrostructural growth markers of teeth can be grouped into two categories. Short period markers or cross-striations and long period markers or brown striae of Retzius. Laboratory experiments made clear that these cross striations represent a 24 hour rhythm. When examining an enamel section in magnification, a series of lines that follow the pattern of the crown growth is visible. These lines can vary from light brown to almost black and are known as the brown striae of Retzius (Figure 1.8). The lines grow in a rhythm. In the lateral and cervical enamel, these striae of Retzius contact the enamel surface. In this way, they form circumferential rings, known as perikymata. This region is referred to as lateral or imbricational enamel (Slootweg 2010).

1.2. Enamel hypoplasia

Enamel hypoplasia is an example of a non-specific stress marker that is visible on the enamel of a tooth. Dental enamel hypoplasia is a class of developmental defects of enamel. These enamel defects range in appearance. There are single and multiple pits, small furrows or deep and wide troughs of decreased enamel thickness (Goodman and Rose 1990, 64). This research will only focus on horizontal grooves, called linear enamel hypoplasia (LEH). Linear enamel hypoplasia (Figure 1.9) follows the horizontal direction of the perikymata and is the result of variations in perikymata spacing (Hillson 2005).

Figure 1.9: Linear enamel hypoplasia

http://thecontagionist.tumblr.com/

The linear groove that is formed is seen as an arrest of amelogenic growth. This stops the completion of that particular increment. A new increment will be formed after this stressful period, which will leave the previous one as an incomplete layer. From the surface is this seen as a zone of thinner enamel, representing a hypoplastic groove. These defects are recognizable with the naked eye and can be measured. The age at which the LEH was formed can be estimated because it correlates with the chronological development of the tooth enamel. This defect can be seen as a permanent record of developmental disruption since enamel does not remodel after deposition. Which means the groove remains as a permanent feature, a physical record of a stressful physiologic event. (Auferheide and Rodriguez-Martin 1998).

1.2.1. Aetiology LEH

Three main aetiological factors are responsible for the formation of enamel defects: hereditary abnormalities, local trauma, and metabolic stress (Pindborg 1970). Past research (Goodman et al. 1991; Pindborg 1982; Sweeney 1969) linked enamel hypoplastic defects to nutritional deficiencies and disease. Investigation on Guatemalan children by Sweeney (1969) found that individuals with dental enamel hypoplasia showed almost twice the rate of infectious disease than individuals without the defect. Malnourished children from a Mexican village who had been taken nutritional supplements to increase their calorie and protein

levels showed a reduced rate of hypoplasias compared with the children who did not take the supplements (Goodman et al. 1991). Clinical studies linked enamel defects to a variety of diseases such as rubella, tetanus, and syphilis (Pindborg 1982). Goodman et al. (1991) linked enamel defects to a number of systematic and physiological stressors such as malnutrition and micronutrient deficiencies.

1.2.2. Sex difference and LEH

Results of studies examining the correlation between sex and the presence of LEH have different outcomes. Goodman et al. (1987) studied the permanent dentition of 300 rural Mexican children, 5 to 15 years old, from five communities in the Solis Valley. Results showed a higher frequency of LEH in women. They suggested that male children may be obtaining greater access to basic resources such as food, shelter, and health care. Studies from a Late Medieval Croatian population had similar results. Slaus (2000) analyzed 104 individuals from a Late Medieval Croatian population from Nova Raca for LEH. Females had statistically significantly more frequencies of hypoplasias than males. Slaus'study about the Early Medieval Croation period suggested that males and subadults were more susceptible to EH than females. These differences in sex in the two periods were attributed to changing local cultural, political and socio-economic conditions (Slaus 2008). Saunders and Keenleyside (1999) investigated the permanent teeth of 253 adults from the St. Thomas' Anglican Church. The skeletal sample was excavated from a 19th century cemetery in Belleville, Ontario. The prevalence of LEH was significantly higher in males than in females.

Other studies (Palubeckaité 2001) have reported that there is no significant sex difference in the presence of LEH. Palubeckaité's (2001) study about a Lithuanian Iron age population found a small difference: males were slightly more affected than females, but the difference was not statistically significant.

Some authors (Van Gerven et al. 1990; Zhou and Corruccini 1995; Saunders and Keenleyside 1999) suggest that men have more LEH because of their greater biological sensitivity to stress factors. Females are seen as biologically better buffered for physiological stress.

Some authors (Goodman et al. 1987; May et al. 1993; Slaus 2000) interpreted a higher prevalence of LEH in females as evidence of greater parental investment in male children. This includes better access to nutritional and medical resources. This idea, that differential treatment of male and female children can influence the frequency of LEH is supported by studies of living children (May et al. 1993). This preferential treatment of males has been noted in many cultures (Goodman et al. 1987).

1.2.3. Defect width and stress periods

Some researchers (Suckling et al. 1986) have suggested that the depth and breadth of a hypoplastic line can indicate the duration and severity of the event that caused the defect. This evidence suggests that it can be helpful to record this information about the enamel defects. However, Hillson and Bond (1997) concluded that defect width cannot be used to determine the duration of a stress event. The entire width of the tooth defect also includes the period of disruption and recovery. This means there will be an overestimation of the actual period of the stress event. More importantly, the space between perikymata varies along the tooth crown surface. This means that defects that are located in different crown regions, but with similar widths can represent different durations of stress periods. Hubbard et al. (2009) came to the same conclusion. They investigated the Irene skeletal collection. This collection consists of 300 individuals ranging from eight to 50 years of age. The casts that were made from the teeth were investigated microscopically to measure the defect widths. In general, it was not possible to use the widths of the defects as an indication for the duration of a stress episode.

1.2.4. Different tooth types and LEH

Marks and Rose (1985) suggested that some tooth types are more susceptible to growth disruption during crown development. According to Goodman and Rose (1990) anterior teeth are more susceptible to hypoplastic defects, so they recommended that the permanent maxillary central incisors and mandibular canines must be used.

A study on prehistoric Amerindians found that anterior teeth were more susceptible when compared to posterior teeth (Goodman and Armelagos 1985). In

the early stage of tooth crown formation, the maxillary first incisors were more susceptible, while at later periods of development, mandibular canines are more susceptible (Goodman and Armelagos 1985). A study of enamel hypoplasia of a site in Libben showed that canine teeth, in comparison to premolars, have a greater variability of hypoplastic defects (Marks and Rose 1985). Hillier and Craig (1992) suggested that teeth with longer crown development periods could be more susceptible to enamel hypoplasia defects. This is due to their slower rates of enamel deposition.

Enamel formation of the permanent maxillary incisor and the mandibular canine are complete around six years (± 1 year) of age (Hillson 1996). Using only these teeth exclude information about a later age. Enamel formation of the third molar, for example, is completed around12 (± 2 years) (Hillson 1996).
Goodman and Rose (1990) argued that hypoplasias appear when stressors reach a level that causes an ameloblastic disruption. According to them (Goodman and Rose 1990) anterior teeth are more sensitive than posterior teeth and have a lower level of ameloblastic disruption. Building on this research, Wright (1997) states that studies that only focus on the anterior dentition give information about the general prevalence of stress episodes in childhood. Studies of the posterior teeth, on the other hand, provide data on variation in timing of stress (Wright 1997). Stress periods of greater magnitude are thus only recorded on posterior teeth. (Wright 1997).

1.2.5. Socio-economic status and LEH

Some studies (Goodman and Capasso 1992; Enwonwu 1973) have shown a direct association between enamel hypoplasias and socio-economic status. Goodman and Capasso (1992) found that in a rural community, families of low socio-economic status showed higher rates of dental enamel defects than wealthier families. Enwonwu (1973) studied children from different socio-economic background in Nigeria. No enamel hypoplasia was found on any of the well-fed children. Among the 420 less wealthier children, 21 percent of these children showed enamel hypoplasia.

Other studies (Dummer et al. 1986; Dobney and Goodman 1991; Nation et al. 1987) however have failed to find any association between hypoplastic lines and socio-economic status. Investigations by Dummer et al. (1986) from 11- and 12 year old children from Wales found no difference between the different social classes. Also, in the UK, Dobney and Goodmann (1991) found no correlation between enamel defects and social class. Nation et al. (1987) studied the presence of enamel hypoplasia on Californian children and also found no correlation with social class. The latter compared their results with those of Enwonwu (1973) in Nigeria. The US children were of a relatively high socio-economic level, with only a small difference between the different social classes. The socio-economic classes in Nigeria on the other hand, were more different. Nation et al. (1987) therefore suggested that this probably influenced the fact that they did not find a correlation between socio-economic status and enamel hypoplasia.

1.2.6. Age occurrence

The transition from breast milk to food, also known as weaning period, is a harsh period for infants. They lose the passive immunity form their mother and become susceptible to malnutrition and disease (Rodney 1983). This seems very probably due to the relationship between malnutrition and illness. Pour nutrition can decrease an individual's ability to cope with stressful periods. A sample representing a population from a middle 19th century settlement from Florence, Italy was studied to determine the age of occurrence of enamel hypoplasias (Moggi-Cecchi et al. 1994). Individuals between the ages 1.5 and 3.5 years were most affected. Historical sources on weaning habits of 19th century Italian populations indicate children got breastfeeded until 12 to 18 months, with weaning period starting after this age. This is in agreement with the data on enamel defects (Moggi-Cecchi et al. 1994). Katzenberg et al. (1996) suggest that enamel hypoplasia can occur during weaning period, but that it is not necessarily the major cause.

1.2.7. Determination of Age of LEH formation

Research has been done to correlate the formation of LEHs with age. Two main methods are recognizable: macroscopic and microscopic.

The first research done on macroscopic methods was done by Massler et al. (1941) and Schour et al. (1941). The latter examined at the University Of Illinois College Of Dentistry over 1000 teeth of healthy children. They combined the data from Logan and Kronfeld's study (1933) with the histological information from the examined teeth. This resulted in a standard of ages when formation of portions of the tooth crown was developed. Building on this, Swardstedt (1966) developed a method that used the location of LEH's to determine the age at which the defect was formed. He divided each tooth into six-month intervals and made a schematic model to illustrate these increments. This method was revised by Goodman et al. (1980) and Goodman and Rose (1990). The regression equations of Goodman and Rose (1990) were widely used in physical anthropology in the 1990's and 2000's.

Other researchers (Hillson 1992; Hillson and Bond 1997; King et al. 2002, 2005) estimated the ages when LEHs were formed by using microscopic methods. They made coated replicas of teeth and with scanning electron microscopy (SEM) it is possible to count the space between the perikymata. To avoid including enamel defects that could have been caused by local trauma, LEH's are matched across at least two teeth to make sure that the LEH's represent systematic growth disruptions.

To determine the age at which the defect was formed, three times need to be added: (1) the estimated time needed for the cuspal enamel in the tooth to be complete, (2) the age at which the tooth starts growing and (3) the time needed for the lateral enamel formation between the cusp tip and the defect to complete formation.

This approach is not influenced by errors that may arise using the chart method or regression equations. He does not assume that tooth crowns grown at a constant rate. And he includes the time for the formation of cuspal enamel in determining the ages at which LEHs form. He also used more than one reference population. On the other hand, the method is very time consuming and requires use of expensive equipment.

The most recent and accurate method is from Reid and Dean (2006). Four samples of teeth were used. One from southern Africa and one from northern Europe contained all incisors, canines, and molars. The two other samples contained only one tooth type. The Danish sample consisted of only canines and a modern North American sample consisted of only third molars. In total, data was collected on 163 incisors, 189 canines and 326 molars. From every tooth a 200-300 μm thin section was made. Each section was examined under polarized light with a microscope to determine the following: (1) the linear thickness of cuspal enamel; (2) the distance between daily cross striations in the inner, middle and outer portion of the cuspal enamel to estimate the time for the cuspal enamel to complete, (3) in lateral enamel: couting the number of daily enamel cross striations between the striae of Retzius, (4) the number of long-period striae in each tenth of crown length, and (5) the total time for the enamel formation.
Two methods were used to determine the number of days between long-period lines in the lateral enamel of each tooth. The first method uses the count of cross striations between the long-period lines. The second divides the distance between long-period markers by the distance between cross striations. The length of the surface of every tooth was then divided in ten equal sections between the cemento-enamel junction and the incisal edge. Counting the number of long-period striae within each decile that was present at the surface was used to calculate the formation time in days for every decile of the crown length.
In all ten deciles is the total lateral enamel formation time similar to the total count of long-period striae. The sum of the lateral enamel formation time and the cuspal enamel formation time equals the total crown formation time.

Martin et al. (2008) compared the method developed by Swärdstedt (1966) with the Read and Dean (2006) method. They concluded that Reid and Dean are the most accurate method for estimating the age at which LEH was formed because they think about the nonlinear growth and variation in crown formation per tooth type.

The European sample, which will be used in this thesis shows relatively longer enamel formation time than the African sample (Read and Dean 2006).

Chapter 2: Historical background from the Beemster

The Beemster is an area in the Dutch province Noord-Holland. Inside the Beemster are four villages situated: Middenbeemster, Noordbeemster, Westbeemster and Zuidoostbeemster. Middenbeemster is the principal town of De Beemster. The Church and cemetery are located in the south-eastern corner of the town. Every village was supposed to have his own church, but only the one in Middenbeemster was build. All the inhabitants of the Beemster were buried at the Middenbeemster cemetery (Netherlands Department for Conservation 1998).

2.1. Living in the Beemster.

Site

In the summer of 2011, the Laboratory for Human Osteoarchaeology and the archaeological company Hollandia conducted an excavation on the former cemetery of the Beemster, which is located next to the church of the Beemster (Griffioen 2011). The excavation was necessary because of planned construction works next to the church. The cemetery was in use from 1617 to 1866, although, according to historical documents, the majority of the excavated graves can be dated between 1829 and 1866. (personal communication Dr M. Hoogland)

Middenbeemster is located in the Beemsterpolder. This polder used to be a lake, but was dried out at the beginning of the 17th century. Around 1605 private investors started to drain the Beemster lake. The lake was almost completely drained in 1610, but because of a break in the dikes, the lake refilled. They decided to make the dike a meter above the surrounding country. The polder was dry in 1612 and the land was divided among the investors. The entire Beemster polder has been UNESCO world heritage singe 1999 (Netherlands Department for Conservation 1998). In 1618 they started with the building of the Middenbeemster church. The cemetery around the church was in use until 1866 (Griffioen 2011). Most of the excavated skeletons are dated between 1829 and 1866 (personal communication Dr. M. Hoogland).

Agriculture

The land was originally used for cereal production. Because of the height of the water table and soil conditions, the land was not suitable for arable farming, so the ground turned into land for cattle. Until the 1880s, the land was primarily used for cattle. After the introduction of a pumping station, it was possible to drain more deeply and the land turned into horticulture. Nowadays the land is a mixture of arable land, greenhouse horticulture and fruit farming (Netherlands Department for Conservation 1998).

2.2. Epidemics and diseases in 19th century Netherlands.

The 19th century was ruled by three diseases: smallpox, cholera and malaria. Cholera was also known as the blue death, because patients turned blue. This disease reached Europe in 1830. Seven outbreaks in the Netherlands in the 19th century are known: 1832-1833; 1848-1849; 1853; 1854; 1855; 1859 and 1866-1867. Symptoms of cholera are diarrhea and dehydration. The disease is transmitted by contaminated water or food. Eating fish that lived in contaminated water or vegetables washed with contaminated water can transmit the bacteria (Vis 1991).

Another disease from the 19th century are the smallpox. There are two big epidemics known form the smallpox in the Netherlands: 1813 and 1817. Smallpox are an infectious disease, caused by either of two virus variants: Variola major and Variola minor. Transmission of this disease occurs through face-to-face contact with an infected person, inhalation of airborne variola virus. It can also spread through direct contact with infected body fluid or contaminated objects. Symptoms of smallpox are fever, muscle pain, headache, nausea and vomiting (Vis 1991).

Malaria is an infectious disease, caused by the parasite Plasmodium. The parasite is transmitted from one person to another by mosquitos. Symptoms of malaria include fever, shivering, joint pain, vomiting, hemolytic anemia and retinal damage. Consequences of severe malaria include coma and, if untreated, death

(Vis 1991). A big malaria epidemic is known from 1846. The winter before was very soft, which lead to the survival of the mosquitos. And because of the following dry, hot summer which is ideal for the parasites. Other big epidemics happened in 1808 and 1826 (De Meere et al. 1982).

Potatoes became the major crop in farming in the first half of the 19th century and would remain so until 1845. It was a more reliable crop than grains, it was cheap to produce, easy to cultivate and fed both farmers and farm animals. Between 1845 and 1847 the potato blight hit the Dutch agriculture and destroyed most of the crops. In the same period were some other diseases present. The long, hot summer of 1845 caused the wheat crops to be only two third of the normal amount. The amount of rye in 1846 was very low and there was a mice plague in the same year (Bieleman 2010).

The Beemster population was more focused on cattle than on crops. There are also two known epidemics of rinderpest, which would have influenced the population. The first plague was in 1851-1852 and the second one was from 1865-1887 (De Meere et al. 1982). This infectious viral disease causes fever, diarrhea and high mortality. The rinderpest had a big influence on humans because they lost animals because of high mortality and also loss of production.

Economy of the citizens of the Beemster polder was more focused on cattle instead of crops. The rinderpest influenced the individuals directly while the potato blight had more to do whit the trade with neighboring villages and had probably less influence. All the epidemics from this century hade probably more influence on the health situation of the population of the Beemster.

Chapter 3: Materials and methods.

3.1. Materials

3.1.1. Sample

The Beemster collection is housed in the Laboratory for Human Osteoarchaeology of Leiden University. Osetobiological information is based on the skeletal analysis performed in the lab by Osteoarchaeology Msc students under the supervision of Dr Andrea Waters-Rist.

The excavated sample is composed of approximately 450 individuals and 50 of them are analyzed in this thesis with an approximately even sex distribution. The sample consist of 18 females (36%), 21 males (42%), and 11 (22%) subadults. Individuals with relatively complete dentition were chosen.
Age-at-death of the subadults is in Table 3.1. There are three juveniles and eight adolescents.

Table 3.1: Age-at-death of the subadults

ID	Age range	Age group
S044V0027	12 ± 1year	juvenile
S269V1065	10 ± 2 years	juvenile
S480V1042	9 ± 1 year	juvenile
S229V0324	17 ± 1 year	adolescent
S275V0526	15 ± 1 year	adolescent
S350V0844	16 ± 1 year	adolescent
S446V0944	16,5 ± 1 year	adolescent
S340V0724	17 ± 1 year	adolescent
S454V0963	17 ± 1year	adolescent
S460V0971	16,5 ± 3 years	adolescent
S507V1093	15 ± 1 year	adolescent

Age-at-death for adults is divided in four groups (Table 3.2.). The sample consists of 13 early young adults (18-25 years), 15 late young adults (26-35 years), ten middle adults (36-49 years) and one old adults (50+).

Table 3.2: Adult age category number

Age range	N=	%
18-25 years	13	33
26-35 years	15	38
36-49 years	10	26
50+	1	3

The 50 individuals of this sample had a total of 891 teeth in total. Almost half of the molars were missing. This is due to the fact that some of the third molars were unerupted (because of congenital absence or uneruption). Another reason is that from every individual two teeth were extracted for DNA, which were mostly molars. The incisors and canines sample was almost complete.

Of the 891 permanent teeth, 334 incisors, 184 canines and 373 molars were observed (Table 3.3). Molars were the most frequently observed type of tooth (42 %), followed by incisors (37,5%). Canines were the least frequently observed tooth type (20,5 %), which could be expected because there are 12 molars but only 4 canines in the permanent dentition.

Table 3.3: Number of teeth per tooth type

Tooth Type	N=	%
I	334	37,5
C	184	20,5
M	373	42

The total number of permanent teeth observed by tooth type and jaw is presented in Table 3.4.

Table 3.4: Number of teeth per tooth type and per jaw

Mandible	N=	%	Maxilla	N=	%
I_1	78	8,6	I^1	86	9,6
I_2	88	9,7	I^2	82	9,1
C	94	10,4	C	90	10
M_1	69	7,7	M^1	74	8,2
M_2	69	7,7	M^2	60	6,7
M_3	53	5,9	M^3	58	6,4

3.1.2. Historical information on the Beemster

The written historical documentation from the Beemster make this an interesting population to investigate.

Information about the individuals is included in a ground plan of the cemetery from the 19th century. This ground plan was divided in sections and the date of the burial and name of the individual was noted for each of these sections. A separate book had records of individuals for which the church bell was rung. People had to pay to have the church bell ringed. This could indicate the socio-economic status of an individual.

The ground plans of the archaeological excavation, made for every horizontal layer, contain all the excavated burials. Combining the available documents with the skeletal analysis enables us to identify the individuals buried in the Beemster cemetery.

Some difficulties arise during the study of the historical and archaeological documents. The 19th century plan from the cemetery indicates that there are 12 rows. On the excavation plans there are sometimes 12 and sometimes 13 rows present, which means there was probably a discrepancy between the official number of people buried and the actual number. The cemetery was also partially cleared out in 1829. Consequently, some people, though not all, buried before 1829 have been removed from the cemetery. Thus, one-to-one identification is very difficult and is an ongoing project.

3.2. Methodology

3.2.1. Skeletal analysis

Age and sex estimations are established for every skeleton. Adult age was estimated using methods for the auricular surface (Buckberry and Chamberlain 2002), pubic symphysis (Suchey and Brooks 1990), cranial suture closure (Meindl and Lovejoy 1985), dental attrition (Maat 2001) and sternal rib ends (Iscan and Loth 1986).

Adult sex estimation was based on pelvic and cranial morphological features (Buikstra and Ubelaker 1994). Metrics were used to supplement the sex estimation (White and Folkens 2005).

Subadult age was determined using long bone length (Maresh 1955; Black and Scheuer 1996 and Young 1957), epiphyseal fusion (Schaefer et al. 2008), dental formation (Moorrees et al. 1963) and dental eruption (Moorrees et al. 1963; Ubelaker 1989).
Because sexual dimorphism in subadults is less than in adults, it is not possible to accurately estimate sex of subadults.

3.2.2. Enamel Hypoplasia: Data Collection
Every aspect of this research is non-destructive. A total of 881 teeth were macroscopically examined for linear enamel hypoplasia. Data collection focused on the presence or absence of LEH from all available teeth of each individual. The sample was visually inspected under natural light with the naked eye and a small hand lens of 10x magnification.

The labial crown height is measured using a pair of dental calipers accurate to 0.001 mm (Hillson Fritzgerald 2005). The total crown height of incisors and canines is measured on the labial surface from the occlusal surface to the cemento-enamel junction. In molars, crown height is defined as the distance between the tip of the mesiobuccal cusp to the cemento-enamel junction, measured along a line parallel to the long axis of the tooth. LEH's present on incisors and canines are measured from the midpoint of the CEJ along a line parallel to the long axis to the middle of the groove. LEH's present on the molars are measured from the cemento-enamel junction to the middle of the groove on the most mesial cusp.

Age estimations for LEH's on complete teeth can be directly calculated using the chronological ages from Read and Dean (2006). For worn teeth, the original total crown height needs to be calculated. The original total crown length of the worn teeth is computed by using the complete teeth. Mean and standard deviation are

calculated for the complete teeth, with a distinction between tooth type, left and right and maxilla and mandible. The results are seen in Table 3.5.

Table 3.5: Mean and standard deviation per tooth type.

Right						Left					
Tooth	N	Mean ± SD	Tooth	N	Mean ± SD	Tooth	N	Mean ± SD	Tooth	N	Mean ± SD
I^1	12	11,077 ± 1,05	I_1	8	9,318 ± 1,15	I^1	8	11,219 ± 1,63	I_1	8	9,584 ± 1,04
I^2	11	9,580 ± 0,73	I_2	10	9,594 ± 0,90	I^2	12	9,795 ± 1,10	I_2	11	9,453 ± 1,21
C	18	10,400 ± 1,35	C	17	11,068 ± 1,27	C	13	10,241 ± 1,19	C	15	10,825 ± 1,55
M^1	12	7,183 ± 0,75	M_1	10	6,492 ± 0,57	M^1	11	7,0282 ± 0,61	M_1	11	6,590 ± 0,54
M^2	16	6,951 ± 0,45	M_2	15	7,3723 ± 0,72	M^2	14	7,195 ± 0,66	M_2	15	7,139 ± 0,66
M^3	10	6,370 ± 0,60	M_3	10	6,551 ± 0,70	M^3	3	6,330 ± 0,23	M_3	6	7,228 ± 0,28

* N= number of teeth used to calculate the mean crown length; SD= standard deviation

If the cemento-enamel junction was not visible, mostly due to calculus, teeth were excluded from this study. When the crown was completely worn off, teeth were also excluded. The latter explains why only one old adult (50+) was included in the sample.

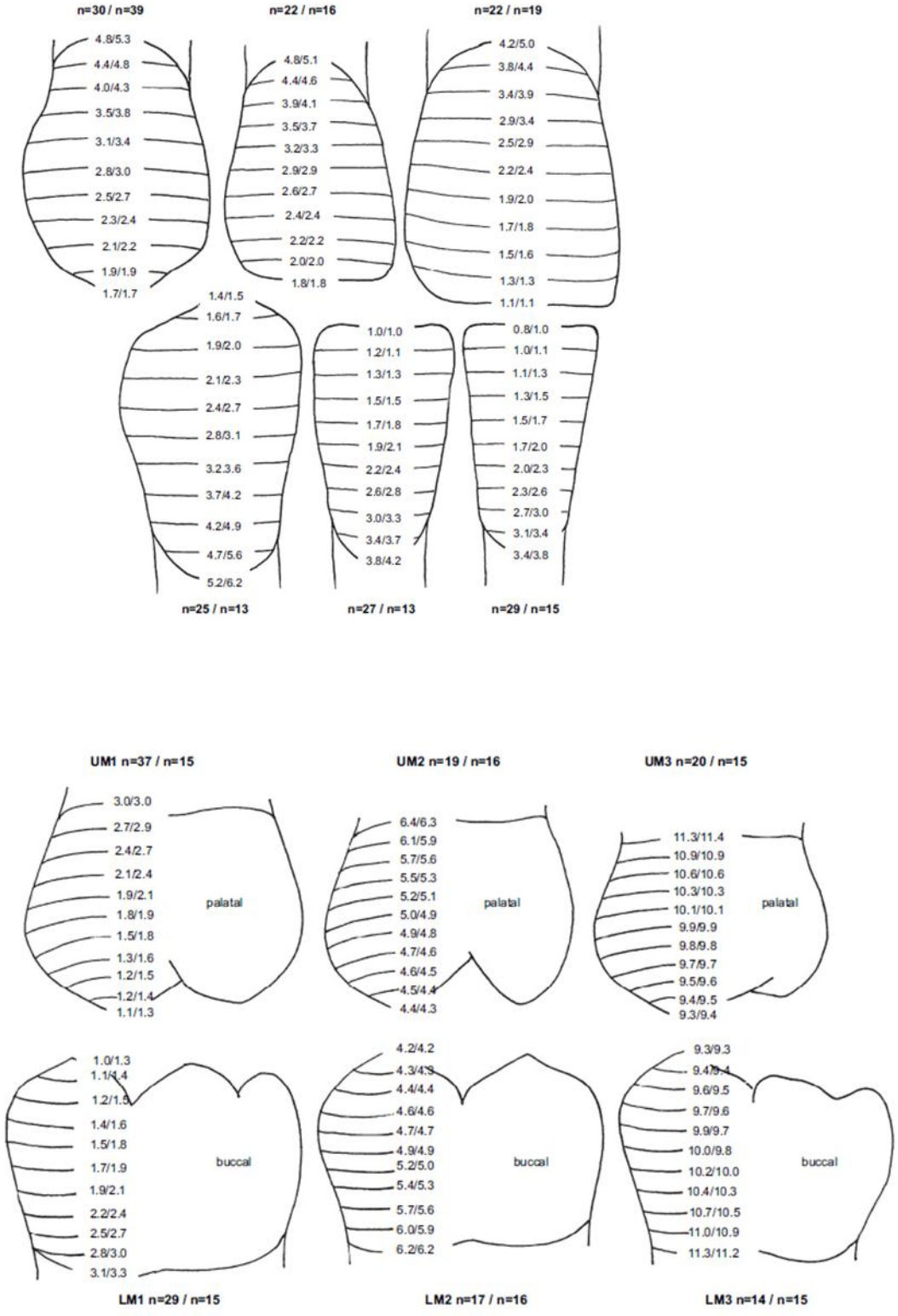
3.2.3. Enamel Hypoplasia: Age of occurrence

Ages for the enamel hypoplasia episodes were calculated using regression equations from Reid and Dean (2006). The mean estimates for chronological ages for enamel formation according to Reid and Dean (2006) in incisors, canines and molars is presented in Figure 3.1. Calculating the relative height from every LEH compared to the total crown length results in an age estimation for that LEH. To exclude grooves occurred by local trauma, an event needs to be present on at least two different teeth. The consideration of nonlinear growth and the variation in crown formation per tooth type makes the Reid and Dean method the most accurate for estimating age of LEH formation (Martin et al. 2008).

3.2.4. Statistical analysis

The statistical package used to analyze the date presented in the following chapter was SPSS 19. Figures and tables are made in excel 2010.

Figure 3.1: Mean estimates for chronological ages of enamel formation for incisors, canines and molars (From Reid and Dean 2006: Pp 343 and 344).



Chapter 4: Results

Of the 50 analyzed individuals, 18 (36 %) had no LEH. Thirty-two individuals (64%) had at least one hypoplastic events. Seventy-six different events were observed. A total of 236 linear enamel hypoplastic defects were observed on a total of 891 teeth. An average of 4.72 enamel hypoplastic defects were observed per individual.

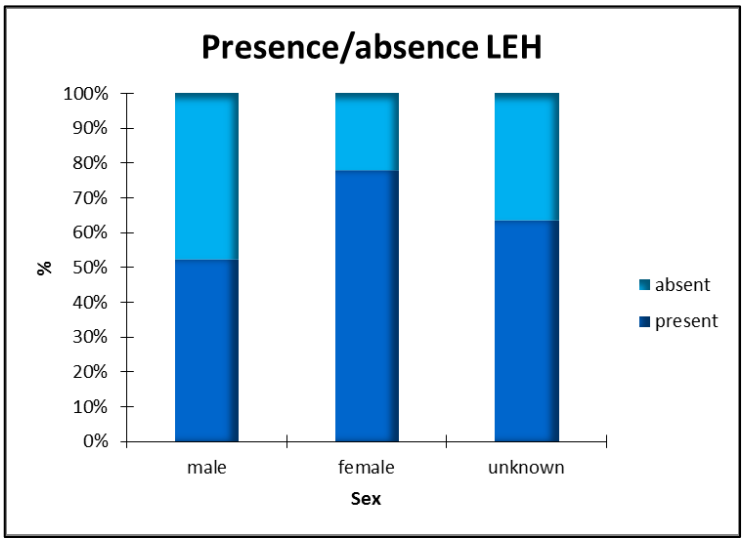
4.1. LEH by Sex

As seen in Table 4.1 and Figure 4.1 in this sample of 50 individuals, 11 males (52%), 14 females (78%) and 7 subadults (63%) suffered from LEH.

Table 4.1: Presence/absence LEH in the sample, divided by sex

		LEH				Total
		Present		Absent		
Sex	Male	11	52%	10	48%	21
	Female	14	78%	4	22%	18
	Subadult	7	64%	4	36%	11
Total		32	64%	18	36%	50

Figure 4.1.:Absolute % of presence/absence of LEH in the sample



In this sample of 50 individuals, females (78%) are more affected than subadults (63%) and males (52%). To determine if the difference for the presence or absence of LEH is statistically significant between males, females and subadults, a chi-square test was conducted. The result showed that the difference is not statistically significant ($X^2 = 2{,}714$; $p = 0{,}257$). As well, the chi-square test indicates, the difference for the presence or absence of LEH for males and females is not statistically significant ($X^2 = 2{,}717$; $p = 0{,}099$).

Table 4.2: Number of individuals and the number of events for males, females and subadults

		Events							
		0	1	2	3	4	5	6	Total
Sex	Male	10	4	4	3	0	0	0	21
	Female	4	7	3	2	1	0	1	18
	Unknown	4	0	2	1	1	3	0	11
Total		18	11	9	6	2	3	1	50

Table 4.2 gives the number of individuals and the number of events for males, females and subadults. The number of events ranges from zero to six. The most severe case is a female with LEHs from six different events. Almost half of the subadults (unknown sex) had LEH from three or more events. The number of people with events goes down when the number of events increases. Only a small number of individuals have a high number of LEH, while a high number of individuals have a low number of LEH. Men have an average of 1.00 events per person, females have an average of 1.61 events per person, and subadults have an average of 2.36 events per person. The latter can partly be explained because three subadults have LEH from five different events.

The data for the total number of events per individual is not normally distributed ($D = 0.206$; $p = 0.000$), so a non-parametric test was performed. To determine if there is a significant difference between males, females and subadults, the data were subjected to a Kruskal-Wallis test ($H = 3.362$; $p = 0.186$). The difference is not statistically significant. By excluding the subadults from the sample, we can compare the males and females. To determine if there is a significant difference

between males and females, the data were subjected to a Mann-Whitney U test (U= 145.000; p = 0.197). The difference is not statistically significant. There is also no statistically significant difference between subadults and adults in the number of hypoplastic events (H= 1.872; p = 0.171). However, comparing only adolescents with adults gives another result. There is statistically significant difference in the number of events for adolescents and adults (H= 4.477; p = 0.034).

Table 4.3 gives the number of individuals and the number of hypoplastic defects for males, females and subadults. The number of hypoplastic defects ranges from zero to 22. The most severe case is a subadult (unknown sex) with 22 hypoplastic defects. Men have an average of 2.9 defects per person, females have an average of 4.8 defects per person, and subadults have an average of 10.7 defects per person.

Table 4.3: Number of individuals and number of defects for males, females and subadults

	Defects															
	0	2	3	4	6	7	8	9	10	11	12	13	16	21	22	Total
Male	10	3	0	3	1	1	1	0	1	0	1	0	0	0	0	21
Female	4	4	2	2	0	2	0	0	1	0	1	1	1	0	0	18
Unknown	4	0	0	0	1	1	0	1	1	1	0	0	0	1	1	11
Total	18	7	2	5	2	4	1	1	3	1	2	1	1	1	1	50

The data for the number of hypoplastic defects per individual is not normally distributed (D = 0.199; p = 0.000), so a non-parametric test was performed. To determine if there is a significant difference in the number of defects for males, females and subadults, the data were subjected to a Kruskal-Wallis test (H= 3.418; p = 0.181). The difference is not statistically significant. To determine if there is a significant difference in the number of hypoplastic defects for males and females, the data were subjected to a Mann-Whitney U test (U= 140.000; p = 0.156). There is no statistically significant difference in the number of hypoplastic defects between males and females.

To determine if there is a significant difference in the number of hypoplastic defects between subadults and adults, the data were subjected to a Mann-Whitney U test (U= 213.500; p = 0.978). There is no statistically significant difference between the number of hypoplastic defects for subadults and adults.

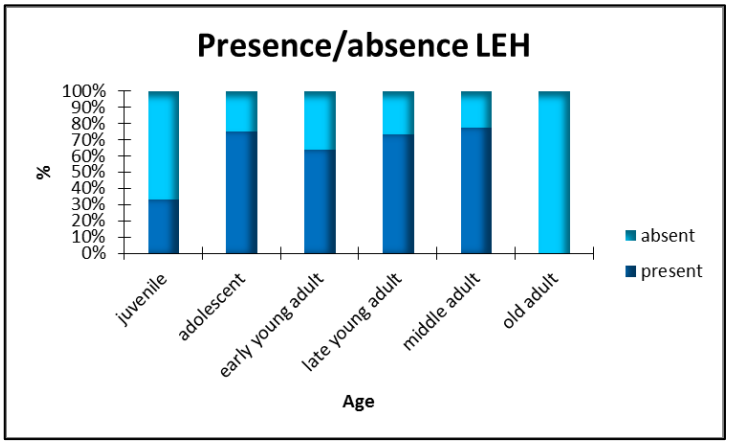
4.2. LEH by Age Category

Table 4.4: Presence/absence LEH in every age category

		LEH				Total
		present		absent		
Age	juvenile	1	33%	2	67%	3
	adolescent	6	75%	2	25%	8
	early young adult	9	70%	4	30%	13
	late young adult	10	66%	5	34%	15
	middle adult	6	60%	4	40%	10
	old adult	0	0%	1	100%	1
Total		32	64%	18	36%	50

In this sample the adolescents (75%) and the early young adults (69%) are the most affected with LEH, closely followed by the late young adults (66%) and middle adults (64%) (Table 4.4 and Figure 4.2). The least affected group is the juveniles (33%), although the sample size is only three. The sample consists of only one old adult, without LEH. To determine if the difference for the presence of absence of LEH is statistically significant between the different age-categories, a chi-square test was conducted. The result showed that the difference is not statistically significant (X^2 = 3,693; p = 0,594).

Figure 4.2: Absolute % of presence/absence LEH in every age category



The number of events ranges from zero to six. The more events, the less persons are affected. Most of the persons have three or fewer events (Table 4.5).

Among the juveniles, of which one has LEH, five teeth with enamel defects were identified on a total of 48 observable teeth. This gives an average of 0.10 affected teeth with LEH in this group. A total of two different events were identified in this subgroup, with an average of 0.66 events per individual. Among the adolescents, of which six had LEH, 41 teeth with enamel defects were identified out of a total of 154 observable teeth. This gives an average of 0.26 teeth with an enamel defect per individual. A total of 24 different events were identified in this subgroup, with an average of three events per individual. Among the early young adults, of which nine have LEH, 37 teeth with enamel defects were identified out of a total of 237 observable teeth. This gives an average of 0.15 teeth with an enamel defect per individual. Nineteen different events were identified in this subgroup, with an average of 1.46 events per individual. Among the late young adults, of which 10 have LEH 41 teeth with enamel defects were identified out of a total of 272 observable teeth. This gives an average of 0.15 teeth with an enamel defect per individual. Eighteen different events were identified in this subgroup, with an average of 1.2 events per individual. Among the middle adults, of which seven have LEH, 19 teeth with enamel defects were identified out of a total of 161 observable teeth. This gives an average of .011 teeth with an enamel defect per individual. Thirteen different events were identified in this subgroup, with an average of 1.3 events per individual.

Table 4.5: Number of individuals and number of events per age category

		Events							Total
		0	1	2	3	4	5	6	
Age	juvenile	2	0	1	0	0	0	0	3
	adolescent	2	0	1	1	1	3	0	8
	early young adult	4	3	3	2	1	0	0	13
	late young adult	5	5	2	3	0	0	0	15
	middle adult	4	3	2	0	0	0	1	10
	old adult	1	0	0	0	0	0	0	1
Total		18	11	9	6	2	3	1	50

The data for the total number of hypoplastic events per individual is not normally distributed (D=0.222; p = 0.00). To determine if there is a significant difference between juveniles, adolescents, early young adults, late young adults, middle adults and old adults, the data were subjected to a Kruskal Wallis test (H= 6,808 ; p = 0.235). The difference is not statistically significant. There is also no statistically significant difference between subadults and adults in the number of hypoplastic events (H= 1.872; p = 0.171). However, comparing only adolescents with adults gives another result. Than a statistically significant difference in the number of events is visible (H= 4.477; p = 0.034).

Table 4.6 gives the number of individuals and the number of hypoplastic defects for every age category. The number of hypoplastic defects ranges from zero to 22. The two most severe cases are two adolescents with 21 and 22 defects. Juveniles have an average of 3 defects per person, adolescents have an average of 9.6 defects per person, early young adults have an average of 4.7 defects per person, late young adults have an average of 3.6 defects per person and middle adults have an average of 3.3 defects per person.

Table 4.6: The number of individuals and hypoplastic defects for every age category.

	Defects															Total
	0	2	3	4	6	7	8	9	10	11	12	13	16	21	22	
juvenile	2	0	0	0	0	0	0	1	0	0	0	0	0	0	0	3
adolescent	2	0	0	0	1	1	0	0	1	1	0	0	0	1	1	8
early young	4	2	0	2	0	2	0	0	1	0	1	1	0	0	0	13
late young	5	2	2	2	1	0	1	0	1	0	1	0	0	0	0	15
middle adult	4	3	0	1	0	1	0	0	0	0	0	0	1	0	0	10
old adult	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Total	18	7	2	5	2	4	1	1	3	1	2	1	1	1	1	50

The data for the number of hypoplastic defects per individual is not normally distributed (D = 0.199; p = 0.000), so a non-parametric test was performed. To determine if there is a significant difference in the number of defects for the age categories, the data were subjected to a Kruskal-Wallis test (H = 5.704; p = 0.336). The difference between the age categories for the number of hypoplastic defects is not statistically significant.

4.3. LEH by tooth type

Of the observed teeth, canines (31%) are the most affected tooth type, followed by the incisors (22.5%). Molars were the least affected tooth type (3%) (Table 4.7).

Table 4.7: Number of affected and not affected teeth per tooth type

	Affected		Not affected	
Tooth type	**N=**	**%**	**N=**	**%**
I	75	22,5	259	77,5
C	57	31	127	67
M	11	3	362	97

Incisors showed a slightly higher average number of defects per tooth type (1.69 events per tooth), closely followed by canines with 1.68 events per tooth. Molars had the lowest average number of defects at 1.09 defects per tooth.

Table 4.8: Number of affected teeth for maxilla and mandible

	Maxilla				Mandible			
	Affected	**%**	**Not Affected**	**%**	**Affected**	**%**	**Not Affected**	**%**
I	37	18	168	82	38	18	166	82
C	27	23	90	77	30	24	94	76
M	7	4	182	96	4	2	191	98

The difference between the affected teeth of the maxilla and mandible are seen in Table 4.8. The difference between left and right is shown in Table 4.9.

Table 4.9: Number of affected teeth for left and right

	Right				Left			
	Affected	**%**	**Not Affected**	**%**	**Affected**	**%**	**Not Affected**	**%**
I	41	20	168	80	44	21	166	79
C	29	23	95	77	28	24	89	76
M	5	2	190	98	6	3	183	97

The numbers show that there is no difference between the maxilla and mandible and the left and right jaw.

4.4. Age-Specific Occurrence of LEH

A histogram for all the ages when hypoplastic events occurred in the 32 affected males, females and subadults with LEH is given in Figure 4.3. The earliest forming of LEH occurs at 1.5 years of age, while the latest age at which LEH occurs is 5.3 years of age. There is a peak at 3.4 years of age.

Figure 4.4 gives the distribution of the ages when LEH was formed for the males, females and subadults separately. The Kolmogorov Smirnov test shows that the age at which hypoplasias occur follows a normal distribution ($D= 0.079$; $p = 0.200$). The one way ANOVA test ($F= 0.067$; $p = 0.935$) shows that there is not a statistically significant difference between males, females and subadults in the specific age occurrence of LEH.

The occurrence of LEH for females is between 2.2 and 4.8 years of age, with three peaks: 2.5; 2.9 and 3.4 years of age. The occurrence of LEH for males start at a younger age. The youngest LEH occurrence for males is at 1.7 years of age and the latest is at 4.8 years of age. Four peaks visible: 2.6; 3.2; 3.4 and 3.6 years of age. The longest period when LEH occurred is for the subadults. LEH occurs from 1.5 to 5.3 years of age. For the subadults are also four peaks visible: 1.8; 2.3; 2.8 and 3.6 years of age.

If we leave the subadults out of the sample, we can compare males and females for every age at which LEH was formed. The Kolmogorov Smirnov test shows that the age at which hypoplasias occurs follows a normal distribution ($D= 0.079$; $p = 0.200$). The results of the independent t-test ($t= -0.347$; $p = 0.730$) shows that there is no statistically significant difference between males and females in the age when LEH was formed. The greatest peak is visible at 3.4 years of age.

Figure 4.3: Spreading of the age occurrence of LEH for all affected individuals. The x-axis are the ages when LEH occurred while the y-axis presents the number of people who had LEH at that age.

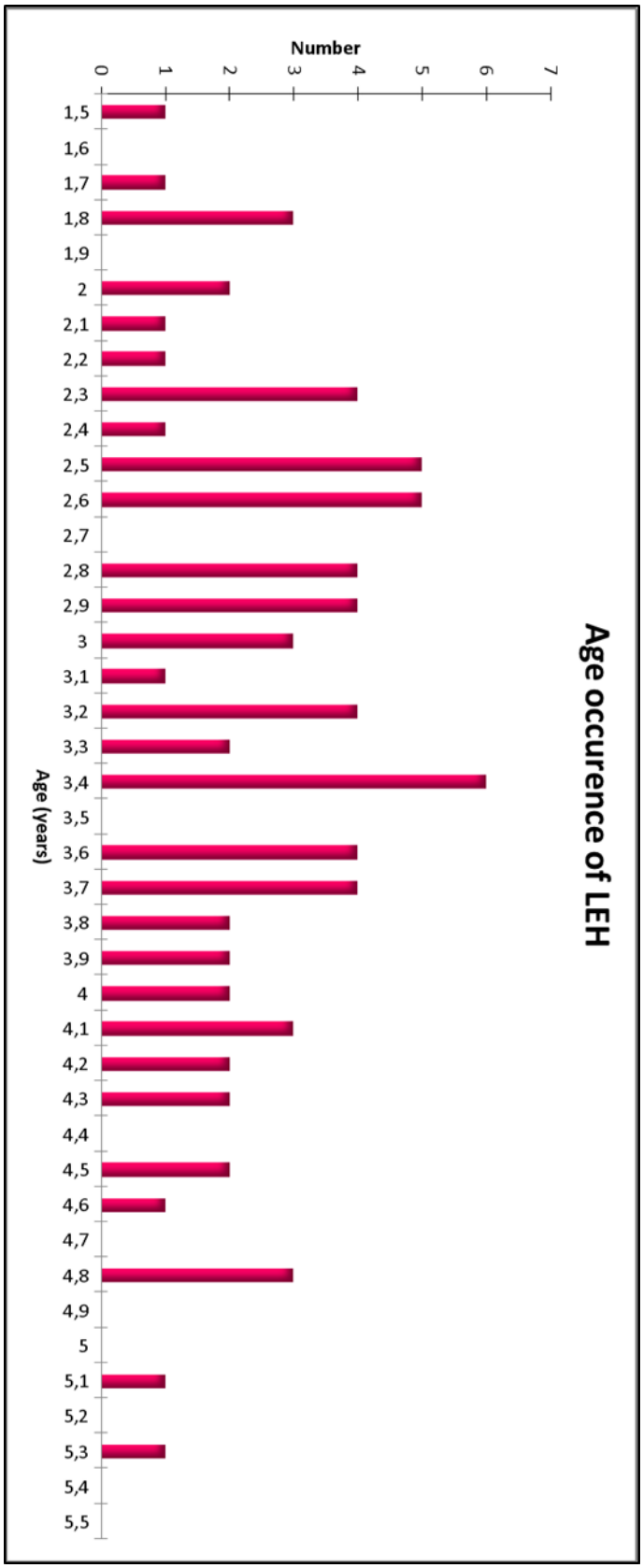
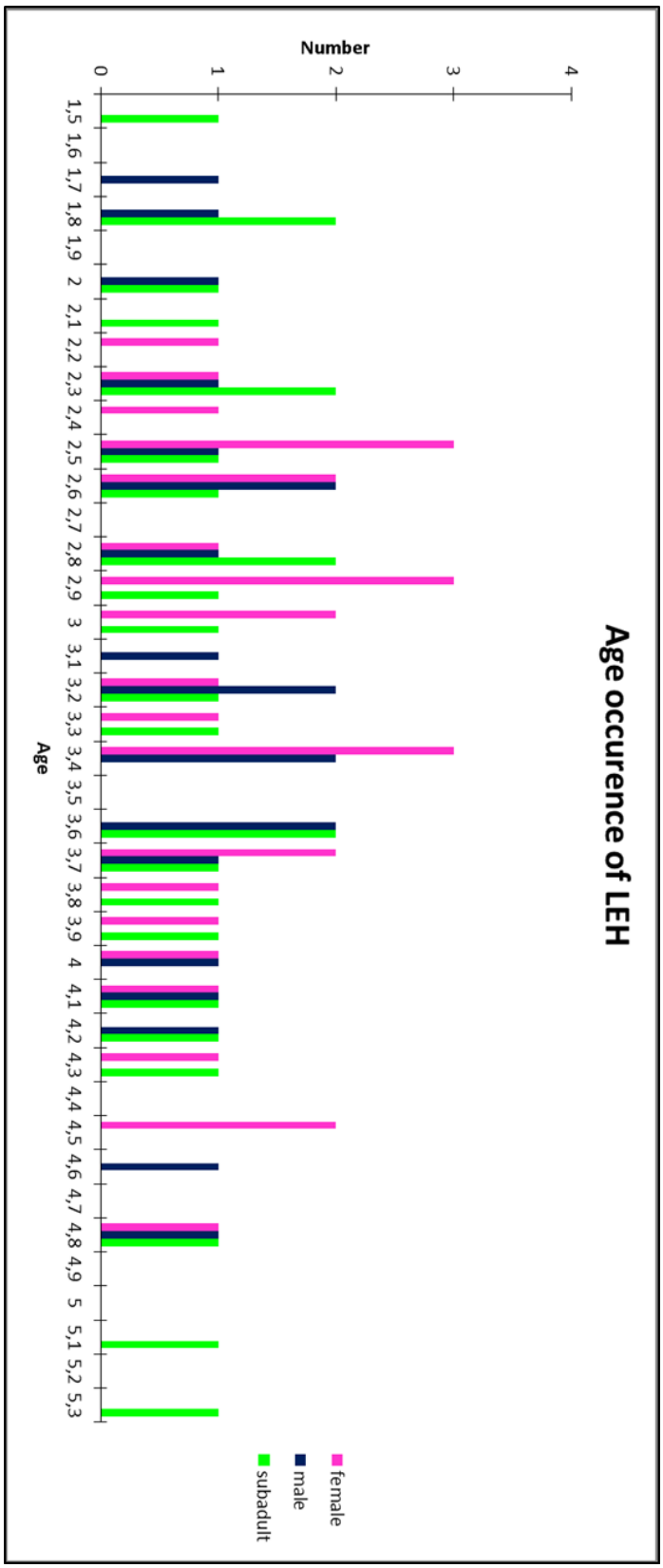


Figure 4.4: Spreading of the age occurrence of LEH for all affected males, females and subadults. The x-axis presents the specific age when LEH occurred. The y-axis shows the number of males, females and subadults who formed LEH at that age.



To have a better overview of the affected groups, every half year was taken together (Figure 4.5 and 4.6). The earliest affected age category is 1.5 – 1.9 years of age, while the oldest affected age category 5.0 – 2.4 years of age is. The peak for the occurrence of LEH is clearly between 2.5 and 2.9 years of age. After that period, the number of LEH's decreases smoothly.

Figure 4.5: Age occurrence of LEH for males, females and subadults together for the six-months age categories. The x-axis gives the six-months age categories while the y-axis presents the number of the individuals who had LEH at that age.

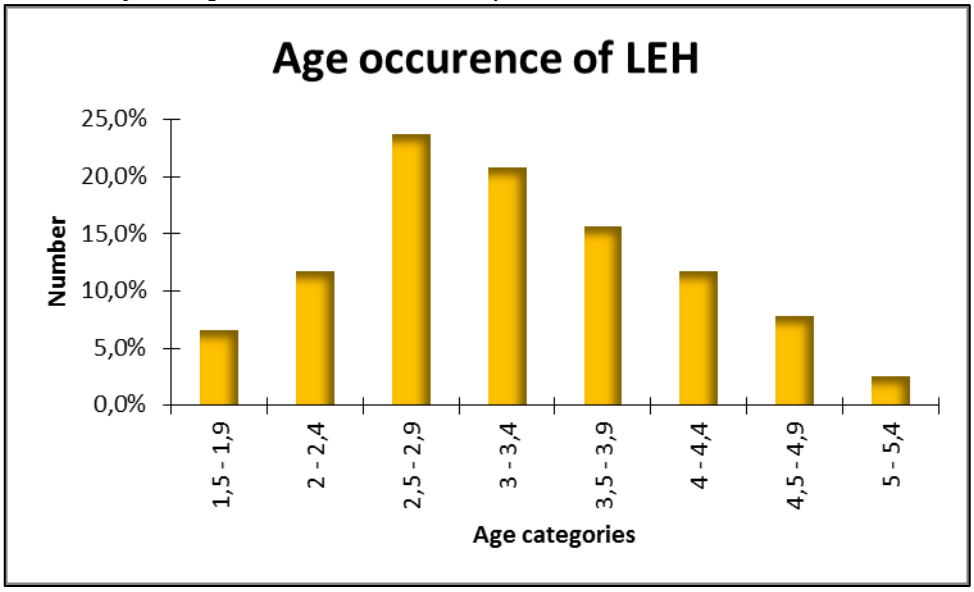


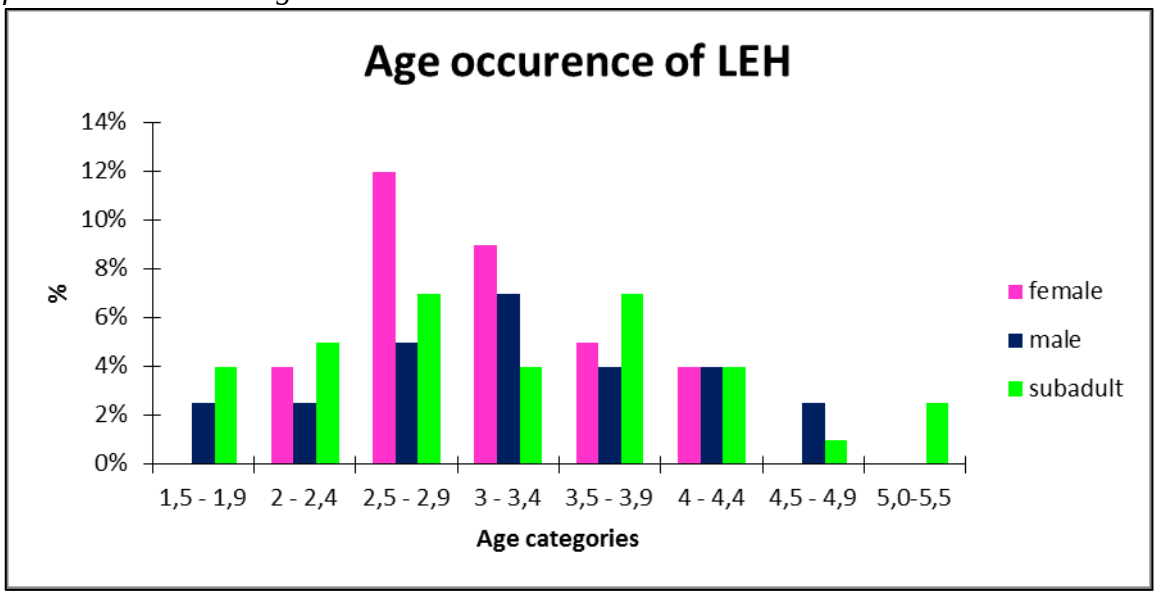
Figure 4.6. presents the number of LEH's from the males, females and subadults. A Kolmogorov Smirnov test (D = 0.144; p = 0.000) proved that this data is not normally distributed. According to a Kruskall-Wallis test (H= 0.188; p = 0.886) there is no significant difference between the specific age occurrence of LEH between males, females and subadults.

The peak for females is in the age category 2.5 – 2.9 years of age, while the peak for males is between 3.0 – 3.4 years of age. Females have for almost every age category more LEH's than males. It is also clear that the LEH's for females are present in a shorter period, between 2.0 – 2.4 and 4.0 – 4.4 years of age, while LEH's in males are present in a longer period, between 1.5 – 1.9 and 4.5 – 4.9 years of age. LEH's in subadults are present in every age category.

Leaving out the subadults gives us an idea about the difference in age occurrence of LEH between males and females for every six-month interval. The result from the Kolmogorov Smirnov test (D= 0.158; p = 0.005) shows that the data is not normally distributed. A Kruskal-Wallis test (H= 1.086; p = 0.189) showed that the specific age occurrence for males and females is not statistically different.
the peak for the age of occurrence of LEH for adults is between 2.5 – 2.9 years of age. The second peak is between 3.0 – 3.4 years of age.

In Figure 4.6 are the adults divided by sex. The peak for females in the presence of LEH is between 2.5 – 2.9 years of age, while the peak for males is between 3.0 – 3.4 years of age.

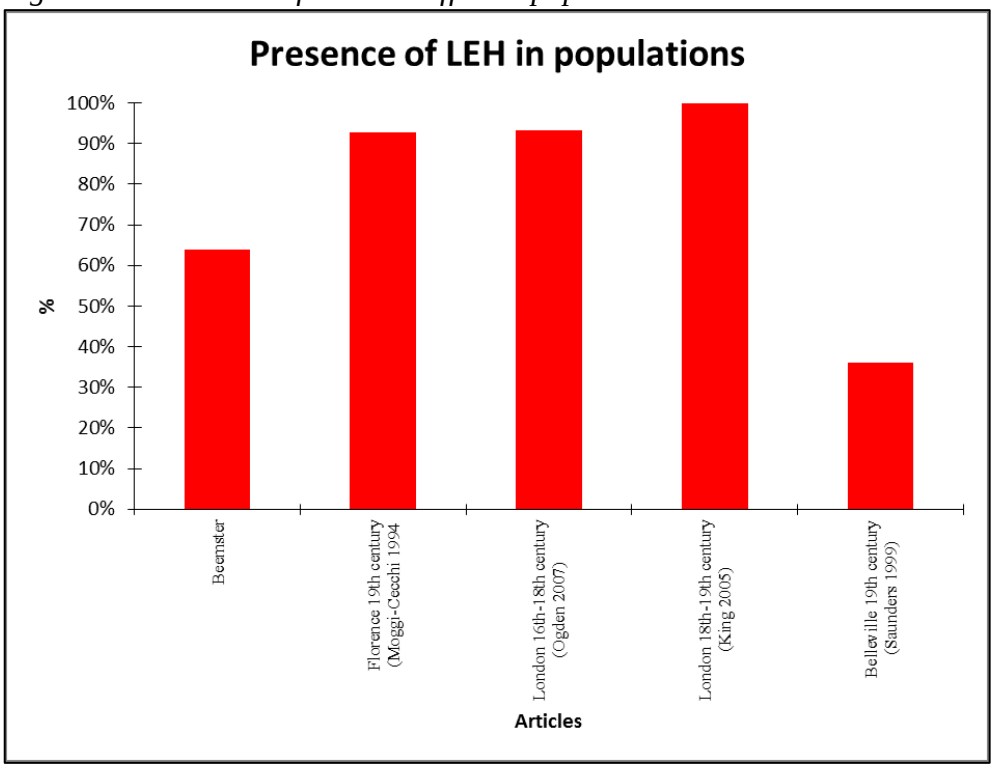
Figure 4.6: Age occurrence of LEH for males, females and subadults separately for the six-months age categories. The x-axis presents the specific age when LEH occurred. The y-axis shows the number of males, females and subadults who formed LEH at that age.



Chapter 5: Discussion

In this study, 64% of the individuals suffered from LEH. Figure 5.1 shows the % of LEH from the Beemster in comparison to other 19th century populations. In comparison to these populations, the % of individuals with LEH is rather low. The only population with less % of individuals with LEH is from the skeletal sample in Belleville, Ontario (Saunders and Keenleyside 1999). The sample consisted of individuals buried between 1821 and 1874 (Saunders and Keenleyside 1999, 515). The low level of LEH was explained that the community where there was enough food, chronic disease levesl were low, but acute infectious diseases were common (Saunders and Keenleyside 1999, 519). The other three populations from Florence (Moggi-Cecchi et al. 1994), London (Ogden 2007) and London (King et al. 2005) all show a higher % of LEH in the population.

Figure 5.1: Presence of LEH in different populations.



5.1. The osteological paradox

According to Wood et al. (1992, 344) interpretation about health status and/or demography can be more difficult than assumed. They outlined three problems that complicate the reconstruction of health status and demography of past populations: (1) demographic nonstationarity, (2) selective mortality and (3) hidden heterogeneity in risks.

Demographic nonstationarity of a population. This means there is no migration, constant age-specific fertility and mortality, zero growth rate, and an equal age distribution. Changes in fertility, in contrary to changes in mortality, have a big influence on the age-at-death distribution. If a population is not stationary, a small variation in fertility has large effects on the age-at-death distribution. A second problem is selective mortality. We never have the complete population at a certain age. The sample consists only of the ones who died at that age. The only 25-year olds we have in the sample are the ones who died at that age. But there is no information from the ones who died at an older age when they were 25 years old. The last problem is the hidden heterogeneity in risks. The population exists of an unknown mixture of individuals who varied in their susceptibility to disease and death. Factors that contribute to this heterogeneity are genetics, socioeconomic differentials, micro environmental variation, and temporal trends in health (Wood et al 1992, 344-345).

This is not completely true for LEH. Dental enamel defects provide a record of childhood stress experience. This can be compared to morbidity and mortality at later ages (Wright and Yoder 2003). In contrary to what Wood et al. (1992) says about selective mortality, LEH gives us the opportunity to collect information for every individual at for example two years of age. We can see for every individual that became older than two if they formed LEH at the age of two or not.

Individuals with LEH are not necessarily the ‘weakest’ members of the population. We know that they were strong enough to survive whatever event caused them stress. Those who died did not live long enough to form a lesion. However, some studies (Stodder 1997; Slaus 2000; Duray 1996) comparing age-

at-death of population noticed that individuals with enamel hypoplasia died younger.

5.2. LEH by sex

Past research has failed to find a consistent pattern in the correlation between sex and LEH. In this study, the difference between males and females in the presence or absence of LEH is not statistically significant. This seems to suggest that both sexes were equally exposed to similar levels of stress during childhood. However, from the numbers it is obvious that more females (78 %) than males (52 %) suffered from LEH. The number of events per individual is also not statistically different when it comes to males and females. Females have an average of 1.61 events per individual while males have an average of 1.00 events per individual. The number of events per tooth show the same pattern. There is no statistically significant difference, but females have 4.8 hypoplastic defects per individuals while, males have only 2.9 hypoplastic defects per individual. The number of affected permanent teeth for males and females also is not statistically different, but the average number of affected teeth shows again that females have 3.05 teeth with LEH and males only 2.00 teeth with LEH. In general, there is no statistically difference between males and females in the frequency of LEH, but females are slightly more affected in every category. The smaller the sample size, the less likely a difference between groups will be found to be statistically significant. This is way larger sample sizes need to be analyzed.

Several studies found a higher prevalence of LEH in boys than in girls (Palubeckaite 2001, 84; Van Gerven et al. 1990, 418) and some of the studies also found a higher frequency of LEH in females (Goodman et al. 1987, 13; Slaus 2000, 201). Goodman et al. (1987) conducted a study of 300 children from an agricultural population from the Solis Valley in Mexico. They noted a higher frequency of LEH in females than in males. Goodman et al. (1987, 18) suggested that male children may have greater access to basic resources such as food, shelter, and health care.

The same trend is visible in a study done by Slaus (2000). He conducted research on the human skeletal remains of 104 individuals from the late medieval Nova Raca cemetery in Croatia. Historical records showed that the population was agricultural and was very dependent on the yearly crops. Significant sex differences are present in the frequency of LEH. The sample was also investigated for the presence of cribra orbitalia. The frequency of cribra orbitalia showed a similar pattern. Both data together suggested that females in Nova Raca experienced greater childhood stress than males. Slaus (2000, 206) suggested that this difference in stress periods between males and females was due to the different access to food.

The higher prevalence of LEH in females can be interpreted in two ways. Some researchers (Goodman et al. 1987; Gurri et al. 1996; May et al. 1993) interpreted the higher frequency of LEH in females as evidence of parental preference for male children which lead to daughter neglect. Because of this less favorable treatment, females may have been more exposed to childhood stress. An alternative of this hypothesis is that both males and females are exposed to the same level of childhood stress but that females are better buffered against stress periods. It is suggested that females are biologically better buffered against environmental stress (Guatelli-Steinberg and Lukacs 1999) . This would mean that during these stress periods, females form LEH and males die. This results in a higher mortality under males and a higher frequency of LEH under females, who survived (King et al. 2005, 556; Guatelli-Steinberg and Lukacs 1999, 95).

The archival information from the 19th century Beemster population may shed light on this problem. When all the individuals are identified, information is available about the number of males and females who died at a certain age and with that information, it is possible to calculate the mortality rate for males and females. If the mortality rate for girls is higher than that for boys, it could be an indicator that there was a slight preference for sons. If, on the other hand, the mortality rate for boys is higher than for girls, this could indicate that there was not really a ‘daughter neglect’ in the Beemster population and that females indeed are biologically better buffered against stress periods.

5.3. LEH by age category

This sample consists of only one old adult, without LEH. Teeth of old adults are normally more worn, that is why there is only one old adult in the sample.

There is no statistically significant difference in the presence of LEH between the age categories. However, the numbers show that adolescents are much more affected than the other age categories. Off all the adolescents in this sample, 75% of them suffered from LEH. The least affected category are the juveniles. Only 33% of this category was affected with LEH.

A higher average number of defects are observed in the adolescent category (0.5 defects per tooth). In contrast, the other age categories have an average of 0.2 defects per tooth. Adolescents have the highest average in every category: average number of defects per tooth, average number of affected teeth and average number of events per individual (summery in Table 5.1)

Table 5.1: Summary of the information by age-category

	defects/ tooth	number of affected teeth	events/ individual
juvenile	0,22	0,10	0,66
adolescent	0,50	0,26	3,00
early young	0,25	0,16	1,46
late young	0,20	0,15	1,20
middle	0,20	0,11	1,30

Especially the number of events per individual is quite interesting. There is no statistically significant difference in the comparison of all the age categories for the number of events. There is also no statistically significant difference when comparing subadults with adults for the number of events. But there is a statistically significant difference in comparing the adolescent group with the adults. The number of affected teeth is also the highest in the adolescent category. This could suggest that individuals who suffered more stress periods are more susceptible to diseases at a later age and thus die at a younger age in comparison to individuals who suffered fewer stress periods in their childhood. These

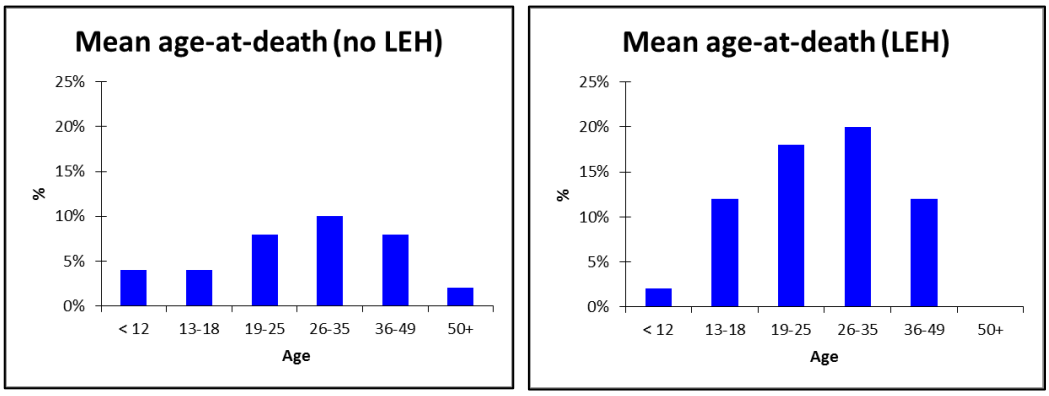
individuals have a permanent weakening of their biological response to physiological stress.

These results are consistent with some previous work. Stodder (1997, 376) found that individuals who died as subadults (< 21 years) showed a higher frequency of LEH than the ones who survived to adulthood. This is supported by research from Duray (1996). He examined a prehistoric population form Ottowa County Ohio for enamel defects. Subadults showed a higher frequency of LEH than the adults. The early mortality of these individuals is, according to Duray (1996, 283) due to biological damage to the immune system during childhood. A similar idea was made by Goodman et al. (1988, 181). They suggest that individuals who are exposed to stress during childhood are biologically damaged. This will reduce their ability to cope with stress later on in their life. The high frequency of LEH can indicate that these individuals had a hard childhood and were exposed to greater stress during childhood. Or, it could mean that these individuals lived a more wealthy life and were able to survive the stress periods during childhood (Palubeckaité et al. 2002, 195).

Palubeckaité (2001) found the same results. He found that the number of stresses has influence on an individuals' longevity. Individuals with multiple stress markers died at a younger age. Palubeckaité (2001, 83) suggest that multiple stress periods in childhood can affect the immune system, so these individuals are less resistant to stress in adulthood.

The individuals in this sample from the Beemster were able to cope with the stress period and survive. But, the individuals who exhibited more and severe stress periods died at a younger age. This could mean that repeated stress periods had an impact on the immune system of the Beemster population. It becomes even more interesting when we compare the age-at-death of the individuals with and without LEH (Figure 5.2).

Figure 5.2: The left graph gives the age-at-death for the individuals without LEH, while the right graph shows the age-at-death of those who died with LEH.



Some previous studies (Duray 1996, 279; Slaus 2000, 204; Stodder 1997) demonstrated that individuals with LEH had a significant lower age-at-death than the ones without LEH. Figure 5.2 gives the mean age-at-death of the individuals of this sample from the Beemster. The mean age-at-death for the individuals with LEH is 26.7 years of age, while the mean age-at-death for the individuals without LEH 27.8 years of age is. The difference is very small. There are some studies (Saunders and Keenleyside 1999, 517; Palubeckaité 2001, 82) who also found almost no difference in the mean age-at-death of the individuals with and without LEH. Saunders and Keenleyside (1999, 521) interpreted this as that LEH is a stress marker that does not have an impact on mortality selection. The individuals with LEH were strong enough to survive these stress periods during childhood, which makes them 'stronger' than the ones without LEH. There is then no reason why they would die at a younger age.

In this sample there is no difference between the mean age-at-death of people with and without LEH. This could mean that LEH had little influence on mortality at a later age. But the individuals with the most number of events and the most affected teeth, died at a younger age. This could suggest that their immune system is weaker because of all the childhood stress periods, which makes it harder to cope with stress in adulthood. The data suggest there may be a mortality effect. Future research involving the entire sample will give a clearer image.

5.4. LEH by tooth type

Of all the present teeth, 44% of the canines and 29% of the incisors were affected with LEH. From the molars, only 3% showed LEH. The average number of events per tooth show a little different image. Incisors have an average of 1.69 events per tooth and canines had 1.68 evens per tooth. These numbers are almost the same. Molars have an average of 1.09 events per tooth. There are no differences between left and right and maxilla and mandible. There are some differences in affected tooth type between males and females. Thirty-three % of the female incisors are affected with LEH, while only 12% of the male incisors has LEH. Canines and molars are similarly affected for males and females. Another difference is that female incisors are more affected than the female canines. While for males, the canines are more affected with LEH than the incisors. The same pattern is visible when we look at the number of events per tooth type. Female incisors have an average of 0.52 events per tooth while male incisors have only an average of 0.17 events per tooth. The average number of events for female incisors (0.52) is higher than that for canines (0.37). For males it is the other way around. The average number of events for male canines (0.46) is higher than the average number of the incisors (0.17).

It is clear that canines are the most affected tooth type, followed by incisors. The high prevalence of LEH in canines and incisors is in line with other research (Hillson 1996; Goodman and Rose 1990; Brook and Smith 2006; Krenz-Niedbala and Kozlowski 2011; Lovell and Whyte 1999). According to Hillson (1996) canines have a slower rate of enamel deposition. They develop over a longer period than other tooth types. Goodman and Rose (1990) concluded that incisors and canines are more often affected with LEH in comparison with premolars and molars. These results are consistent with previous research. Knowledge of intratooth variations in susceptibility are important for future research. Comparing studies based on different tooth types can be problematic, tooth specific rates must always be reported to avoid these problems.

There is no statistically significant difference in the number of affected teeth between the maxilla and mandible. Goodman and Armelagos (1985, 482) stated that maxillary central incisors and mandibular canines are the teeth with the highest frequency of LEH. This trend has been seen by other researchers (Moggi-Cecchi 1994; Goodman and Rose 1990; Goodman et al. 1987) and is also visible in this research (Table 4.7).

Hillson (1996) suggested that there is no systematic difference in the crown formation times between the maxillary and mandibular dentition. The mean estimates for chronological ages of enamel formation of Reid and Dean (2006) can maybe help with understanding this trend. It is clear that there are some differences between the ages of crown formation of the maxillary and mandibular dentition (Table 5.2). The crown formation of maxillary canines begins around 1.7 years of age and completes at approximately 5.3 years of age, while the crown formation of mandibular canines begins at around 1.5 years of age and completes at approximately 6.2 years of age. The crown formation of maxillary first incisors begins at around 1.1 years of age and completes at approximately 5.0 years of age, while the crown formation of mandibular first incisors begins at around 1.0 years of age and completes at approximately 3.8 years of age (Reid and Dean 2006, 343-344).

Table 5.2: Beginning and completion ages for tooth crown formation (according to Reid and Dean 2006).

	maxilla	mandible
I^1	1.1 - 5.0	1.0 - 3.8
I^2	1.8 - 5.1	1.0 - 4.2
C	1.7 - 5.3	1.5 - 6.2

The teeth with the longest tooth crown formation, according to Reid and Dean (2006), are the central maxillary incisor and the mandibular canine (Table 5.2). these are also the teeth with the highest LEH frequency.

5.5. LEH age-specific occurrence

The age distribution of individual LEH for males, females, and subadults demonstrates peaks at 2.5, 2.6, and 3.4 years of age. Between the age of one and two, the presence of LEH is very low, while the presence of LEH is very high in the next three years of life. If we divide the year into six-month intervals, a peak is seen in the 2.5 – 2.9 age category. After and before this peak, the number of LEH decreases gradually.

If we leave out the subadults from the sample, we can see if there is a difference in age-occurrence of LEH between males and females. The age distribution of individual LEH for adults demonstrates a peak at 2.5, 2.6 and 3.4 years of age. The presence of LEH is low in the first year and increases in the second, third and beginning of the fourth year. If we divide the year in six-month intervals, a peak is seen in the 2.5-2.9 age category. So if we keep out the subadults we have the same results. If we compare males and females, some differences are visible. The presence of LEH for females peaks at 2.5-2.9 years of age, while for males the peak is at 3.0-3.4 years of age. The presence of LEH for females is also in a shorter period, from 2.2 until 4.8 years of age. The first presence of LEH for males is at 1.7 years of age and ends at 4.8 years of age. The presence of LEH for subadults is from 1.5-5.3 years of age.

It must be kept in mind that these results are reached using the mean chronological age estimations from Reid and Dean (2006, 343-344). This article is very recent. Research (Goodman et al. 1987) before this was done using other equations. This can give some problems when comparing to other studies that used these equations. Ritzman et al. (2008) made a comparison between the microscopic study from Reid and Dean and existing studies. Ritzman et al. (2008) showed that there are significant differences between LEH age estimations using previous methods and those from Reid and Dean. The results found that ages estimated from the Reid and Dean method are older by one to four months (Ritzman et al. 2008). Therefore, comparisons with studies using other methods of LEH age estimation must be made with this in mind.

In this research we see that LEHs are formed from the second half of the first year until the first half of the fourth year of life. These results are also found in previous research (Goodman et al. 1987; King et al. 2005; Malville 1997; Moggi-Cecchi et al. 1996; Palubeckaité 2001; Palubeckaité et al. 2002; Saunders and Keenleyside 1999). The peak of this sample is in the 2.5-2.9 and 3.0-3.4 age category. The peak in other studies is similar to this result. Malville (1997) found in his study on Puebloan populations from Southwestern Colorado that the peak for LEH was between 3.0-3.5 years of age. Goodman et al. (1987) found a peak in the second and third year of life for Mexican children.

Some researchers (Lanphear 1990; Moggi-Cecchi et al. 1994; Goodman et al. 1987) have linked the presence of LEH in this period of an individuals' life to the process of weaning. Weaning is the transition from breast milk to non-breast milk food. If this transition occurs too early, it tends to be associated with diarrhea and food allergies. This cannot be seen as a single event, but more as a process where the child needs to adapt to 'new' food (Saunders and Keenleyside 1999). Saunders (1999) combined historical data and nitrogen isotope analysis of skeletal remains from Belleville to see if there is a correlation between LEH and weaning. These data suggest that the people from Belleville were nursed for about 14 months, and were introduced to food at around 5 months of age. The results do not correspond with the LEH peaks occurring at two years and 3.5 years of age (Saunders and Keenleyside 1999).

There is no information available about breastfeeding children in the Beemster area in the Netherlands in the 19th century. However, more general information about nursing in the Netherlands suggests that children were breastfeeded until approximately one year of age (Engelen and Ying-Hui 2007).

Archival information has showed that a lot of the women from the Beemster were housewives (personal communication Dr. M. Hoogland). This could indicate they had time to breastfeed their children longer, in contrast to wives who worked outside the home. However, these housewives may have been involved in daily farm activities. As well, parents had a lot of children (personal communication Dr.

M. Hoogland). If these children followed each other quickly, the mother would not have had the time to give breastmilk for a long period.

Only five LEHs are formed in the two year of age range in the Beemster sample. If we assume that the weaning period for the Beemster population falls between one and two years of age, most of the LEHs cannot be explained using the transition from mother's milk to non-mother's milk food. Either children from the Beemster are breastfeed longer, so the weaning period falls between 2.5 and 4 years of age, or the formation of LEHs have other causes. Further stable isotope research will be able to determine the length of breastfeeding and weaning in the Beemster sample.

LEH can be produced by a lot of stressors. It is likely that some of the LEHs in this sample are due to sickness as a consequence of weaning, but this is not the only reason of the formation of these LEHs. Other factors are possible causes of the LEHs, which are discussed in the next section.

5.6. Causes of LEH

The specific aetiology of LEH is unknown. LEH is seen as a general stress indicator for childhood. The biggest cause of LEH is metabolic stress.

Nutrition plays an important role in enamel defect formation. Goodman et al. (1991) studied the effect of nutritional intake during childhood in Mexican adolescents. Individuals who took a nutrient supplement had a significant lower rate of LEH than a group without supplements. These results are supported by a study of May et al. (1993) on 63 Guatemalan children. Children who received a daily intake of nutritional supplements had less LEH than children who did not received the supplements. Sweeney (1969) found that 75% of Guatemalan children who were in hospital for malnutrition had enamel defects.

The relationship between nutrition and LEH is further supported by a study by Zhou and Corruccini (1998). They investigated the relationship between LEH and the nutritional stress causes by a historical famine in China (1959-1961). A significantly higher prevalence of LEH was detected in individuals whose teeth were developing in the period of that famine than in the individuals whose teeth were not forming in that period. The rural population had significantly higher LEH frequency than the urban population, probably due to poorer nutritional and living conditions in rural areas.

Previous research has also focused on the relation between diseases and enamel defects. A lot of diseases are linked to the formation of LEH, but there is no one-to-one relation. If two individuals have the same infectious disease, it does not necessarily mean that they both form LEH.

The relatively high amount of individuals with LEH suggest these individuals had a stressful childhood. Finding one specific cause for the presence of LEH in the Beemster population is not possible. But it is possible to give some options.

The 19th century was a harsh period for the Netherlands when it comes to diseases. It was ruled by three diseases: smallpox, cholera and malaria. Seven outbreaks of cholera are known from this period: 1832-1833; 1848-1849; 1853; 1854; 1855; 1859 and 1866-1867. More than 8000 people died from these epidemics in Noord-Holland (Vis 1991). Two big epidemics of the smallpox are known from this century. Almost every year, a few hundred people died from this disease (Vis 1991). The last big epidemic disease is malaria. Approximately 200.000 people died in the 19th century in the Netherlands from malaria and his consequences (Vis 1991).

All these epidemics probably had a big influence on the general health of the population of the Beemster polder. LEH is a non-specific stress marker, it is therefore not possible to link it to one specific disease. It is clear that many individuals died during these epidemics. A lot more people probably suffered from one of these diseases and survived. These individuals possibly formed LEH

and are in this sample. Unfortunately, it is not possible to link every hypoplastic event from every individual to a specific cause.

The biggest famine in the 19th century in the Netherlands result from the potato blight in 1845-1847. In these years the potato crop failed. Which led to a lower availability of food per individual (De Meere et al. 1982). In the same year was a failure of the rye crop because of a mice plague and the wheat crops were only two third of the normal amount (Bieleman 2010). The agriculture in the Beemster was focused on cattle and less on cereal production. This could mean the impact of the potato blight was less severe than in other areas. There were some rinderpest epidemics in the 19th century, but it is not clear to with extend they reached the Beemster.

Many LEH events in this sample are probably due to the epidemics of infectious diseases from this period and the famine caused by the potato blight and the rinderpest.

Conclusion

Even if there is no statistically significant difference between males and females in the presence of LEH, females (78%) are more affected that males (52%). This can be explained in two ways. Either parents favored sons, so males survived stress periods in childhood with fewer problems, while females formed LEH. Or females are biologically better buffered against childhood stress, which would indicate that females form LEH, but survive these stress periods in childhood, while males die. Information about the mortality rate for boys and girls can give a direction for this discussion.

There is no statistically significant difference in the presence of LEH between the different age categories and the mean age-at-death for individuals with and without LEH is almost the same. But the adolescent category (13-18 year) has the highest number of events per individual, the highest number of affected teeth and the highest average of events per tooth. If we compare the adolescents with the adults (the rest of the age categories combined) we see there is a statistically significant difference in the number of events per individual.
This could indicate that the formation of LEH in childhood has no influence on mortality in adulthood. Except maybe for the most severe cases, who have multiple LEHs and have more affected teeth. This could indicate that multiple 'attacks' on the immune system in childhood leads to a weaker immune system in adulthood.

The most affected tooth type are the incisors and canines. The most affected teeth are the central maxillary incisor and the mandibular canine. This is in agreement with previous studies. This makes sense as these teeth have the longest tooth crown formation times which means they have more time to form LEH.

LEHs in this study are present between 1.7 and 5.3 years of age: there are three peaks in this sample, at 2.5, 2.6 and 3.4 years of age. The peak for females is between 2.5-2.9 years while the peak for males is slightly later, between 3.0-3.4 years of age. Overall however there is no significant difference between age of defect formation in males and females.

There are a lot of factors that play a role in the formation of enamel defects. Discussing the different options, it seems that weaning can be an explanation, but is probably not the main cause. It is generally assumed weaning is a cause for LEH. In the Beemster, the weaning period probably occurred between one and two years of age. Almost no LEHs are formed in this period, and the peaks for LEH are definitely at a later age. Another cause of LEH is malnutrition. In 19th century Netherlands a few famines are known, mostly from crop failures. The most famous is the potato blight from 1845-1847. But the Beemster population was more focused on cattle than on crops, which means the influence of these events was probably lower than in other areas. One of the other main causes of LEH are diseases. The 19th century is known for his epidemics: cholera, smallpox, malaria and a lot of other smaller diseases. The most LEHs formed in the population of the Beemster are probably due to malnutrition and epidemics.

Future research

Future research for this sample will provide more information about the childhood health of the 19th century Beemster population. For example, stable isotope research will help determining the weaning period for this population. This research can help determining the impact of weaning on the formation of LEH in this population.

Once the archival information is available, some topics can be studied more intensively and specifically.

For this study age estimations from the skeletons instead of specific ages were used. Once the specific age-at-deaths are available from the archives, it is possible to be more specific about the difference in age-at-death for individuals with and without LEH. In this sample, females were more affected than males. Sex of the subadults can not determined in this study, but will be available in the archive. This information can be used to determine if this trend also occurs in the subadults. LEH was seen by researcher as a disease of the 'poor'. The archival information about the socio-economic status of every individual can be used to see if there is a correlation between LEH and status in the Beemster population.

And maybe the most interesting study will be the correlation between these individuals and known historical events. With the archival information, it is possible to determine in which year exactly the hypoplastic events were formed. These events can then be linked to known historical events like an episode of cholera or the potato blight. This can also help us understand why some individuals had LEH and some did not.

Abstract

Linear enamel hypoplasias are growth disruptions in enamel thickness, formed during childhood and, since enamel does not remodel, stay forever. These defects are seen as stress markers and widely used to interpreted childhood health. This study examined the permanent dentition of a sample of the 19th century rural Beemster population. The teeth are examined macroscopically and systematic disturbances are identified by matching hypoplasias among different teeth. The highest frequency of LEHs are on the central maxillary incisor and mandibular canine. The sample consists of 11 subadults and 39 adults. From all individuals, 64% showed linear enamel hypoplasia. A higher prevalence of LEH was found in the females, although there was no statistically significant difference between the sexes. Adolescents exhibit a significantly higher number of events in comparison to the adults, and have more affected teeth per individual. There is no difference in the mean age-at-death between individuals with and without LEH. Thus, the presence of LEH does not affect mortality in this sample, but the individuals with the highest number of hypoplastic events die at a younger age.

The specific age occurrences of LEH fall between 1.5 and 5.3 years of age, with the highest number of LEH between 2.5 and 3.5 years of age. These peaks do no correspond to historically suggested weaning age. The combination of malnutrition and epidemics are suggested to be responsible for the majority of LEH forming events.

Bibliography

Aufderheide, A.C. and C. Rodriguez-Martin, 1998. *The Cambridge Encyclopedia of Human Paleopathology*. Cambridge: Cambridge University Press.

Bieleman, J., 2010. *Five centuries of farming: a short history of Dutch agriculture 1500-2000*. Wageningen: Wageningen Academic Publishers.

Black, S. and L. Scheuer, 1996. Age changes in the Clavicle: From the Early Neonatal Period to Skeletal Maturity. *International Journal of Osteoarchaeology* 6 (5), 425-435.

Brook, A.H. and J.M. Smith, 2006. Hypoplastic enamel defects and environmental stress in a homogeneous Romano-British population. *European Journal of Oral Science* 114, 370-374.

Buckberry, J.L. and A.T. Chamberlain, 2002. Age estimation from the Auricular Surface of the Ilium: A revised Method. *American Journal of Physical Anthropology* 119, 231-239.

Buikstra, J.E. and D.H. Ubelaker (eds), 1994. *Standards for data collection from human skeletal remains*. Arkansas Archaeological Survey Research Series No. 44, Fayetteville: Arkansas Archaeological Society.

Cobourne, M.T. and A.T. DiBiase, 2010. *Handbook of orthodontics*. Edinburgh: Elsevier.

De Meere, J.M.M., 2010. *Economische ontwikkeling en levensstandaard in Nederland gedurende de eerst helft van de negentiende eeuw*. 's Gravenhage: Nijhoff.

Dobney, K. and A.H. Goodman, 1991. Epidemiological studies of dental enamel hypoplasias in Mexico and Bradford; their relevance to archaeological skeletal

material. In: H. Bush and M. Zvelebil (eds), *Health in past societies; biocultural interpretations of human skeletal remains in archaeological contexts*. British archaeological reports international series 567. Oxford, tempus repartum, 81-100.

Dummer, P.M.H., A. Kingdon and R. Kingdon, 1986. Prevalence of enamel developmental defects in a group of 11- and 12-year old children in South Wales. *Community Dentistry and Oral Epidemiology* 14 (2), 119-122.

Duray, S.M., 1996. Dental indicators of stress and reduced age at death in prehistoric Native Americans. *American Journal of Physical Anthropology* 99 (2), 275-286.

Engelen, T. and H. Ying-Hui, 2007. *Two cities, One life: Marriage and Fertility in Lugang and Nijmegen*. Nijmegen: Valkhof Press.

Enwonwu, C.O., 1973. Influence of socio-economic conditions on dental development in Nigerian children. *Archaeology Oral Biology* 18, 95-108.

Goodman, A.H. and G.J. Armelagos, 1980. Enamel hypoplasias as indicators of stress in three prehistoric populations from Illinois. *Human Biology* 52, 515-528.

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

Goodman, A.H., L.H. Allen, G.P. Hernandez, A. Amador, L.V. Arriola, A. Chavez, and G.H. Pelto, 1987. Prevalence and age at development of enamel hypoplasias in Mexican children. *American Journal of Physical Anthropology* 72, 7-19.

Goodman, A.H., R.B. Thomas, A.C. Swedlund and G.J. Armelagos, 1988. Biocultural Perspectives on Stress in Prehistoric, Historical and Contemporary Population Research. *Yearbook of Physical Anthropology* 31, 169-202.

Goodman, A.H. and J.C. Rose, 1990. Assessment of systematic physiological perturbation from dental enamel hypoplasias and associated histological structures. *Yearbook of Physical Anthropology* 33, 59-110.

Goodman, A.H., C. Martinez and A. Chavez, 1991. Nutritional supplementation and the development of linear enamel hypoplasia in children from Tezonteopan Mexico. *American Journal of Clinical Nutrition* 53, 773-781.

Goodman, A.H. and L. Capasso, 1992. Recent contribution to the study of enamel development defects. *Journal of Paleopathology Monographic Publications 2,* 151-170.

Griffioen, A., 2011. *Inventariserend veldonderzoek te Middenbeemster, Middenweg* 148. Hollandia reeks 346. Zaandijk.

Guatelli-Steinberg, D. and J.R. Lukacs, 1999. Interpreting sex differences in enamel hypoplasia in human and non-human primates: developmental, environmental, and cultural considerations. *Yearbook of Physical anthropology* 42, 73-126.

Gurri, F.D., G. Balam, and E.F. Moran, 1996. Sex differences in the frequency and distribution of linear enamel hypoplasias among the Yucatec Maya. *American Journal of Physical Anthropology*, Supplement 22, 117.

Hillier, R.J. and G.T. Craig, 1992. Human dental enamel in the determination of health patterns in children. *Journal of Paleopathology* 2, 381-390.

Hillson, S.W., 1992. Impression and Replica Methods for Studying Hypoplasia and Perikymata on Human Tooth Crown Surfaces from Archaeological Sites. *International Journal of Osteoarchaeology* 2 (1), 65-78.

Hillson, S., 1996. *Dental Anthropology*. Cambridge: University Press.

Hillson, S., 2005. *Teeth* (2nd edition). Cambridge University Press.

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

Hillson, S., C. FitzGerald and H. Flinn, 2005. Alternative Dental Measurements: Proposals and Relationships With Other Measurements. *American Journal of Physical Anthropology* 126, 413-426.

Hubbard, A., D. Guatelli-Steinberg and P.W. Sciulli, 2009. Under Restrictive Conditions, Can the Widths of Linear Enamel Hypoplasias Be Used as Relative Indicators of Stress Episode Duration. *American Journal of Physical Anthropology* 138 (2), 177-189.

Iscan, M.Y. and S.R. Loth, 1986. Estimation of age and determination of sex from the sternal rib. In: K. J. Reichs (ed), *Forensic Osteology: Advances in the Identification of human remains*. Springfield: Illionois, 68-89.

Katzenberg, M.A., D.A. Herring and S.R. Saunders, 1996. Weaning and Infant Mortality: Evaluating the Skeletal Evidence. *Yearbook of Physical Anthropology* 39, 177-199.

King, T., S. W. Hillson and L.T. Humphrey, 2002. A detailed study of enamel hypoplasia in a post-medieval adolescent of known age and sex. *Archives of Oral Biology* 47(1), 29-39.

King, T., L.T. Humphrey and S. Hillson, 2005. Linear enamel hypoplasia as indicators of systematic physiological stress: evidence from two known age-at-death and sex populations from postmedieval London. *American Journal of Physical Anthropology* 128, 547-559.

Krenz-Niedbala, M. and T. Kozlowski, 2011. Comparing the chronological distribution of enamel hypoplasia in Rogowo, Poland (2nd century AD) using two methods of defect timing estimation. *International Journal of Osteoarchaeology* (unpublished article).

Lanphear, K.M., 1990. Frequency and distribution of enamel hypoplasias in a historic skeletal sample. *American Journal of Physical Anthropology* 81, 35-43.

Logan, W. and R. Kronfeld, 1933. Development of the human jaws and surrounding structures from birth to the age of fifteen years. *Journal of American Dental Association* 20, 379-427.

Lovell, N. and I. Whyte, 1999. Patterns of Dental Enamel Defects at Ancient Mendes, Egypt. *American Journal of Physical Anthropology* 110, 69-80.

Maat, G.J., 2001. Diet and age-at-death determinations from molar attrition. A review related to the low countries. *Journal of Forensic Odontostomatology* 19, 18–21.

Malville, N.J., 1997. Enamel hypoplasia in ancestral Puebloan populations from southwestern Colorado: I. Permanent dentition. *American Journal of Physical Anthropology* 102, 351-367.

Maresh, M.M., 1955. Linear growth of long bones of extremities from infancy through adolescence. *American Journal of Diseases of Children* 89, 725-742.

Marks, M.K. and J.C. Rose, 1985. Scanning electron microscopy of Wilson bands and enamel hypoplasias. *American Journal of Physical Anthropology* 66, 202-220.

Martin, S.A., D. Guatelli-Steinberg, P.W. Sciulli and P.L. Walker, 2008. Brief communication: Comparison of Methods for Estimating Chronological Age at Linear Enamel Formation on Anterior Dentition. *American Journal of Physical Anthropology* 135, 362-365.

Massler, M., I. Schour, and H. Poncher, 1941. Developmental pattern of the child as reflected in the calcification pattern of the teeth. *American Journal of Diseases of Children* 62, 33-67.

May, R.L., A.H. Goodman and R.S. Meindl, 1993, Response of bone and enamel formation to nutritional supplementation and morbidity among malnourished Guatemalan children. *American Journal of Physical Anthropology* 92, 37-51.

Meindl, R.S. and C.O. Lovejoy, 1985. Ectocranial Suture Closure: A Revised Method for the Dtermination of Skeletal Age at Death Based on the Lateral-Anterior Sutures. *American Journal of Physical Anthropology* 68, 57-66.

Moggi-Cecchi, J., E. Pacciani and J. Pinto-Cistemas, 1994. Enamel hypoplasia and age at weaning in 19th century Florence. *American Journal of Physical Anthropology* 93, 299-306.

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

Nation, W.A., L. Matsson and J.E. Peterson, 1987. Developmental enamel defects of the primary dentition in a group of Californian children. *Journal of Dental Children* 54, 330-334.

Netherlands Department for Conservation, 1998. *Droogmakerij de Beemster*. Netherlands: UNESCO.

Palubeckaité, Z., 2001. Patterns of linear enamel hypoplasia in Lithuanian Iron age population. *Variability and Evolution* 9, 75-87.

Palubeckaité, Z., R. Jankauskas and J. Boldsen, 2002. Enamel hypoplasias in Danish and Lithuanian Late Medieval/Early Modern Samples: A possible reflection of child morbidity and mortality patterns. *International Journal of Osteoarchaeology* 12, 189-201.

Pindborg, J.J., 1970. *Pathology of the dental hard tissues*. Philadelphia: Saunders.

Pindborg, J.J., 1982. Aetiology of developmental enamel defects not related to fluorosis. *International Dental Journal* 32, 123-134.

Reid, D. and M. Dean, 2006. Variation in modern human enamel formation times. *Journal of Human Evolution* 50, 329-346.

Ritzman, T.B., B.J. Baker and G.T. Schwartz, 2008. A Fine Line: A comparison of methods for estimating ages of linear enamel hypoplasia formation. *American Journal of Physical Anthropology* 135, 348-361.

Roberts, C. and K. Manchester, 2010. *The archaeology of disease*. Stroud: Cornell University Press.

Rodney, J.D., 1983. The age related distribution of dental indicators of growth disturbance in ancient lower Nubia: an ethological model from the ethnographic record. *Journal of Human Evolution* 12, 535-543.

Saunders, R.S. and A. Keenleyside, 1999. Enamel hypoplasia in a Canadian Historic Sample. *American Journal of Human Biology* 11, 513-524.

Schaefer, M., S. Black and L. Scheuer, 2008. *Juvenile Osteology. A laboratory and field manual*. London: Elsevier.

Slaus, M., 2000. Biocultural analysis of sex differences in mortality profiles and stress levels in the late medieval population from Nova Raca, Croatia. *American journal of Physical anthropology* 111(2), 193-209.

Slaus, M., 2008. Osteological and dental markers of health in the transistion form the Late Antique to the Early Medieval period in Croatia. *American Journal of Physical Anthropology* 136(4), 455-469.

Slootweg, P.J., 2010. *Dental pathology. A practical introduction*. Nijmegen: Springer.

Stodder, A.L.W., 1997. Subadult stress, morbidity, and longevity in Latte Period populattions on Guam, Mariana Islands. *American Journal of Physcial Anthropology* 104(3), 363-380.

Suchey, J. M. and S. Brooks, 1990. Skeletal age determination based on the os pubis: A comparison of the Acsádi-Nemeskéri and Suchey-Brooks methods. *Human evolution* 5(3), 227-238.

Suckling, G., D.C. Elliott and D.C. Thurley, 1986. The macroscopic appearance and associated histological changes in the enamel organ of hypoplastic lesions of sheep incisor teeth resulting from induced parasitism. *Archives of Oral Biology* 31(7), 427-439.

Swärdstedt, T., 1966. *Odontological aspects of a Medieval population in the province of Jämtland/Mid Sweden*. Stockholm: Tide-Barnagen.

Sweeney, E.A., 1969. Factors associated with linear hypoplasia of human deciduous incisors. *Journal of Dental Research* 48(6), 1275-1279.

Ubelaker, D.H., 1989. *Human skeletal remains: excavation, analysis, interpretation*, 2nd edition. Washington: Taraxacum.

Van Gerven, D. P., R. Beck, and J.R. Hummert, 1990. Patterns of enamel hypoplasias in two medieval populations form Nubia's Batn el Hajar. *American Journal of Physical Anthropology* 82, 413-420.

Vis, G.N.M. 1991. *650 jaar ziekenzorg in Alkmaar*. 1341-1991. Alkmaar: Uitgeverij Verloren.

White, T.D. and P.A. Folkens, 2005. *The Human Bone Manual*. Amsterdam: Elsevier Academic Press.

Wright, L. E., 1997. Intertooth Patterns of Hypoplasia Expression: Implications for Childhood Health in the Classic Maya Collapse. *American Journal of Physical*

Anthropology 102(2), 233-247.

Wright, L.E. and C.J. Yoder, 2003. Recent progress in bioarchaeology: approaches to the osteological paradox. *Journal of Archaeological Research* 11(1), 43-70.

Wood, J.W., G.R. Milner, H.C. Harpending and K.M. Weiss, 1992. The osteological paradox. Problems of inferring prehistoric health from skeletal samples. *Current Anthropology* 33(4), 343-370.

Young, R., 1957. Postnatal growth of the frontal and parietal bones in white males. *American Journal of Physical Anthropology* 15, 367-386.

Zhou, L. and R.S. Corruccini, 1998. Enamel hypoplasias related to famine stress in living Chinese. *American Journal of Human Biology* 10(6), 723-733.

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