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Linear Enamel Hypoplasias in Hasanlu: a Survey

Abstract

The purpose of this thesis was to investigate the presence of LEH in the Hasanlu and Dinkha Tepe collection, compare the LEH presence between the two collections and between the two areas of the Hasanlu site, High Mound and Lower Mound. The overall questions to be answered were what linear enamel hypoplasia prevalence exists, if there was a difference in linear enamel hypoplasia prevalence between sites and if that difference was significant enough to signal phenomena that shapes systemic health status. Every tooth to each individual in the collection was macroscopically surveyed to look for the presence of linear enamel hypoplasia. If present the LEH lines were measured with a Mitutyo needle nosed caliper and recorded. Only teeth with matching LEH were said to have LEH. The LEH prevalence was found to be 54.2% in High Mound Period IVB, 68.9% in Lower Mound, 80% in High Mound Period IIII, 59.4% in Dinkha Tepe, and 64% overall. The significance of the LEH numbers was determined with a chi-square test, and it was found that there was no statistically significant difference between sites. Statistical significance was found for the Lower Mound age and sex distribution.

Disciplines

Anthropology

LINEAR ENAMEL HYPOPLASIAS IN HASANLU: A SURVEY

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ABSTRACT:

The purpose of this thesis was to investigate the presence of LEH in the Hasanlu and Dinkha Tepe collection, compare the LEH presence between the two collections and between the two areas of the Hasanlu site, High Mound and Lower Mound. The overall questions to be answered were what linear enamel hypoplasia prevalence exists, if there was a difference in linear enamel hypoplasia prevalence between sites and if that difference was significant enough to signal phenomena that shapes systemic health status. Every tooth to each individual in the collection was macroscopically surveyed to look for the presence of linear enamel hypoplasia. If present the LEH lines were measured with a Mitutyo needle nosed caliper and recorded. Only teeth with matching LEH were said to have LEH. The LEH prevalence was found to be 54.2% in High Mound Period IVB, 68.9% in Lower Mound, 80% in High Mound Period IIII, 59.4% in Dinkha Tepe, and 64% overall. The significance of the LEH numbers was determined with a chi-square test, and it was found that there was no statistically significant difference between sites. Statistical significance was found for the Lower Mound age and sex distribution.

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INTRODUCTION:

Examining the teeth of skeletons is helpful when investigating the health of past populations.

First, teeth are usually still preserved with the skeleton, even after a long time. Because the enamel of the teeth is composed mostly of mineral, it is unaffected by the decomposition of organic matter. Enamel is normally one of the best preserved hard tissues (Hillson 2005) even though its solubility may lower over time, poor support may cause it to fracture, and there may be post-mortem pitting and cratering of the surface. Two paths of investigation can be taken with teeth. The number of caries can indicate nutritional information, while the number of enamel defects can indicate a snapshot of overall health of an individual from age 1-6 (Hillson 2005).

This snapshot is possible because enamel formation, or amelogenesis is a process that has a particular rhythm (Hillson 2005). Each ameloblast in amelogenesis undergoes two stages, matrix production and maturation. The ameloblast is first responsible for the matrix formation and seeding of a specific area in the enamel for one year. Then, the cell switches to maturation and removes protein and water to create heavily mineralized mature enamel (Goodman & Rose, 1991). Even after the cells have died, the process of maturation continues in the mouth until about 6 years of age (Hillson 2005). Ameloblasts at the tip of the crown mature first, the cells down the crown sides develop after and the cells at the cervix mature last (Hillson 2005).

Disruption to the maturation of the enamel can leave striations and defects in the teeth.

These defects in enamel quality are generally termed hypoplasias and occur when the ameloblasts cease the matrix production early and the result is underdeveloped portions of enamel (Goodman & Rose 1991). These disruptions to matrix production may be caused by

three conditions, either a hereditary trait, due to a local trauma or by overall stress in the body (Goodman & Rose, 1991). However, these three traits can be distinguished from one another through patterns of defects.

Hereditary defects are rare and are usually not found (Goodman & Rose, 1991). The distinction between local and system stress as the cause of hypoplasias are determined through examination of all the teeth. System-wide stress causes matrix disruption in many teeth, and is evidenced by the fact that the most prominent lines on enamel can usually be matched with other teeth (Hillson 2005). However, not all teeth can be matched in humans because some teeth do not have much overlap between teeth formation times and molars with thick enamel that form slowly and that may not indicate the LEH that other teeth have (Hillson 2005). These prominent lines on the enamel are associated with systemic developmental defects like those caused by fevers and nutritional deficiencies in humans (Goodman &Rose 1991, Hillson et al, 1990).

Defects across the enamel on several teeth can indicate periods of systemic stress on the body during the enamel maturation (Reid and Dean 2000). Hillson (2005) investigated the time at which certain hypoplasias formed in individuals with known health histories, and the hypoplasia formations seemed to match periods of disease in the individuals. Linear enamel hypoplasia is a furrow form defect that has a clear indented line running through a single plane of a tooth. Because linear enamel hypoplasias run through the enamel, they are better indicators of systemic stress than pit or plane form hypoplasias. While the appearance of LEH is more prominent, each LEH should still be matched between several teeth to confirm that the disruption was systemic.

The defects vary on each tooth according to the specific timing and deposition of enamel formation. It is possible to quantify the age of occurrence of defects since the crown enamel forms earlier than the cervix enamel. However, the age of formation of molar hypoplasias tend to be difficult to estimate, and thus hypoplasias on molars are harder to match with other teeth. Thus most studies match incisors and canines (Hillson 2005, Reid & Dean, 2000). Reid and Dean have given ages limits of the surfaces of the canines and incisors which range from 1-6 years of age.

Based on the frequency of LEH, it may be possible to understand more about the environments of the populations being studied. Developed, industrialized countries tend to have less than ten percent of the population with one or more hypoplasias while the presence of hypoplasias are more common in Third World countries (Goodmand & Rose 1991). This indicates that the frequency of hypoplasias increases with poorer nutritional statuses and conditions. It has been shown in several studies that prehistoric populations have a high frequency of hypoplasias (Goodman & Rose 1991).

Hasanlu Project

Hasanlu is a settlement located in the Solduz valley of northwestern Iran. It has a history that begins from the 2nd millennium B. C. and it has been used extensively throughout the years. When the project began excavating in 1957, the site was a modern village (Muscarella 1964). It had already been scouted and excavated a little by other teams, includine Sir Aureul Stein (Muscarella). Hasanlu's large size and evidence of Iron Age pottery made it a promising site. The excavation was led by Oscar Muscarella and Robert Dyson with the purpose of learning

about the early history of northwestern Iran and to understand the influences of trade, migration and invasions in the pre and protohistoric times.

The physical settlement is the largest of the Gadar/Solduz River valley, and generally consists of two areas. The High mound or "Citadel" is situated on a mound that rises 25m from the plain. The Lower Mound or "Outer Town" is a on mound that rises about 8m from the plain. The Lower Mound has also served as a cemetery (Dyson R, 1989).

Hasanlu has produced extensive information due to the presence of several occupation levels from several time periods on the mounds (Muscarella 1964) (Figure 1, based off of Selinsky 2009)). The level closest to the surface is Period I dating to about 1200-1300 AD. It is a poorly preserved Islamic level (Selinksy 2009). Period II dates to about 300-275 BCE and is marked by the Western and Classic Triangle Ware pottery. Period III dates to about 350-750 and is divided into two parts: IIIA which is closely tied to Period II and involves the same pottery, and Period III B which has distinctly Urartian architecture and wares. The break in IIIA and IIIB signifies a period of abandonment most likely due to the fall of the Urartian reign. These levels are marked mostly by rubble and abandoned settlements.

The level beneath Period III, Period IV 1250-800 BCE, in the High Mound found many remains and artifacts preserved. Period IV is divided into three parts. Period IVA was a time where squatters had taken over the settlement of demolished Hasanlu from the end of Period VIB. Period IVB circa 800 BCE was time that began in 1100 BCE and ended with an invasion and a fire that destroyed the people previously living there. The collapse of the buildings on fire however caused the preservation of many structures. Precious items were found underneath the rubble of a 5 buildings (Muscarella 1989) including a the famous "Golden Bowl". Many people

were also trapped with personal items still on them. The fire and invasion of the Hasanlu site was probably conducted by the Urartians as Period III is dominated by all Urartian structures (Muscarella 1989).

Period IVC was marked by a period of construction and expansion. It ended however, with the burning of several buildings. While there is evidence of fire, the fire is not unequivocally intentional like it was in Period IVB. Thus, it is safe to assume that there was continuous settlement of Hasanlu between Period IVB and Period IVC.

The culture found from the Period IV level seems to continue into Period V as the pottery and material culture appear the same. Because there are difficulties with the chronology of when Period V ended and Period IV began (Muscarella 2006), it is likely that the populations were from the same line of people who settled in the area (Muscarella 2006).

Period V (c. 1450 - c. 1250 BCE) and Period VI do have distinct differences that signal a replacement of the population in the settlement around 1450 BCE. The pottery styles are markedly different between Period IV and V. Period IV has a distinctive monochrome grey in contrast to Period VI's painted vessels (Muscarella 2006).

Period VI is referred to as "Khabur Ware" that is evidenced by a wall and a burial with pottery vessels and dates to a Late Bronze Age Period c.1600 - c. 1450 BCE. Period VII, from Late 4m cent. - c. 1600 BCE, is labeled the "Earlier Bronze Age" and is evidenced by the presence of a paint-decorated vessel and a burial.

The excavation at Hasanlu has produced one of the largest skeletal collections of the Near East (Selinsky 2009). Approximately 246 skeletons of people were found, most of whom had died in the fire. The rest were victims of violence during the simultaneous invasion (Muscarella

1989). The violent deaths are evidenced through the presence of head traumas, limb disarticulation and bone damage on the skeletons (Muscarella 1989). It is possible that some of the invaders also fell at the site and were comingled with the bodies of the victims. However, it is unlikely that many invaders died because of the lack of weaponry on the skeletons and evidence of violent deaths (Smay-Toebbe 2005).

The invasion and fire came to the people in Period IVB of Hasanlu during the late summer or early fall since evidence of charred harvested crops was clearly abound in the site (Muscarella 1989). Because of the unique time of the invasion, it was possible to see what kind of food crops was eaten. Emmer wheat, bread wheat, barley and broomcorn millet were popular cultivated crops that were found in the buildings (Harris 1989). Evidence of chickpeas, lamb, grapes and figs around the site signal their use in the diet (Harris 1989). The food crops provide a context for the Hasanlu settlement's nutritional status and explanations for some dental pathologies.

While unfortunate, these untimely deaths at the High Mound in the Hasanlu site provide a cohort of healthy people from 800 BCE Iran to study. These people would not have died naturally as the skeletons were of a wide age range with men, women and children. Many people were unprepared to die as their skeletons still bore jewelry and held valuables (Muscarella 1964). Thus, this population seems to provide a healthy cross section of the population. Because the cohort displays traits shown by healthy individuals, examination the linear enamel hypoplasia presence is an excellent opportunity to understand the childhood health of the living population at the time.

In contrast to the constant transitions of High Mound, Lower Mound is more peaceful. In the north, there is a cemetery. The dead were deliberately folded in simple inhumations with grave goods. The deaths of the individuals in Lower Mound were singular and occurred over gradually. These burials seem to date to Period II to Period V (Smay-Toebbe 2005).

Because the both remains are from similar time ranges but with deaths of different causes, it is possible that the prevalence of linear enamel hypoplasia (LEH) is different between the Lower Mound group and the High Mound group. The cause of death in Lower mound may have been due to strong developmental defects and deficiencies, so the LEH patterns may possibly reflect that by having more or less LEH than the High Mound group.

Dinkha Tepe was excavated in conjunction with Hasanlu in the 1966. It was located about 15 miles west of Hasanlu (Muscarella 1974) separated by ridges but without any serious barriers to stop travelers from going from one site to another (Muscarella 1974). Dinkha Tepe was a *tepe* or mound about 66 ft. tall and 1,330 feet wide in diameter. At the site, a major cemetery was discovered with 94 burials (Figure 2 from Muscarella 1974). The contents of the burials were similar to the objects found in Hasanlu Periods IV (1000-800 BCE) and V (1300-1000 BCE) (Muscarella 1974). Unlike Hasanlu, Dinkha Tepe does not show evidence of a later Iron III period. The burials dated to two Dinkha periods, sixty eight in Dinkha II (1000-800 BCE) and twenty six in Dinkha III (1300-1000 BCE). The graves were very deliberate with 3 distinct types and were lined with numerous grave goods usually in the form of pottery vessels. Some burials had food remaining. Sheep and goat bones were also found.

Both Hasanlu and Dinkha Tepe burials involve a flexed skeleton position, and similar types of pottery, jewelry and animals for food. However Hasanlu burials were typically simple

inhumations while Dinkha Tepe burial had some mudbrick and stone tombs that are not found in Hasanlu.

The purpose of this investigation is to survey the linear enamel hypoplasia in the Hasanlu collection and also the skeletons from Dinkha Tepe. There have been imperfect surveys of teeth because of the different locations of the skeletons in the museum and the numerous organization systems.

The percent of linear enamel hypoplasias present will add to data on the population health of Hasanlu during the time in which it was found. The percentage of LEH can then be compared with other populations to see how the teeth defects vary.

The specific objectives are to record the amount of linear enamel hypoplasias, analyze the prevalence of LEH and to see if any comparisons can be made between the amount of LEH in High Mound, Lower Mound and Dinkha Tepe populations.

While the objectives of this study are extensive, even with perfect methods, there are limitations to the knowledge that can be gleaned. First, the collection is only part of what was excavated in Hasanlu since some of the skeletons were too delicate to recover (Selinsky 2009). Also, the skeletons found in the site are only a portion of the population living at the time. There is no way to determine how representative of the overall population the skeletons in Hasanlu are. Only a little more than 50% of the skeletons from the Hasanlu collection have mandibles or maxillas with teeth. Of those with teeth parts, some skeletons have teeth that are very worn, with enamel that is missing or with the teeth so scattered determination of individuals is difficult. Even skeletons with mandibles and maxillas that contain intact teeth may miss some teeth. Thus a survey of LEH will not be able to be confirmed across all teeth types. Also, an inventory of the

Dinkha Tepe skeletons was not available and thus, it was difficult to ascertain the demographics of the burials.

BACKGROUND TO THE RESEARCH PROBLEM:

Physical anthropologists and archaeologists both work by speculating and hypothesizing in their fields. Artifacts and skeletons that are thousands of years old can no longer respond to the questions asked by researchers. Thus, both kinds of anthropologists examine and interpret the evidence available in order to draw conclusions. One of the overlaps between archaeology and physical anthropology is the use of skeletons for clues. For the archaeologists, where and when the individual was found and with what belongings give hints to the overall picture of life at the time. For biological anthropologists, detailed examinations and comparisons of specific body parts reveal the extent of variation in the population and the environment may have affected the individuals in their specific context and history.

In the past, teeth were not as interesting to physical anthropologists were more intent on viewing other differences between people like hair color (Scott & Turner 2008). Yet from the 1920's onwards, the study of teeth grew more popular. The connection between teeth and human genetics gave way to the first casting study in the 1940's by Dahlberg (Scott & Turner 2008). In the late 1950's compilations of teeth size, morphology, pathology and wear began to be published. At first, these studies were used to compare different teeth forms and measurements. Researchers attempted to find sex differences in teeth size and greater variation such as shoveled incisors or population specific identifiers (Scott & Turner 2008). However, it seems as though teeth are relatively uniform among humans or at the very least, teeth morphology and dimensions vary more within populations than between them.

While population specific morphology was not found, experimental studies on enamel defects in rats, mice and other animals showed a connection between stressors and the development of an enamel defect. This finding tied to the idea that stressors caused by the environment could lead to a stress response in the body and result in a stress marker (Figure 3). This linear cause and effect idea ushered in thoughts and experiments about how teeth could reveal clues to the past. For example through analysis dental morphology or pathology or growth defects, there could arise greater understanding of genetics, diets or systemic stress on an individual.

Study of teeth defects has grown quite popular, and investigations are not limited to the teeth of human populations. Guatelli-Steinberg (2004) has published research on the teeth of ancient hominins and well comparisons between humans and Neandertal populations. Lukacs (2009) has examined the markers of physiological stress on bonobos in the form of dental hypoplasias. All of the studies have been done in the attempt to understand how the environment and health of the populations can be learned through teeth.

Linear enamel hypoplasias are of particular interest in dental anthropology because teeth have the potential to signal clues about the overall health and stress from ages 1-6 of the populations studied. Usually, the amount of LEH can be a proxy for overall stress in that population's environment and also a measure to compare health between populations and even species (Guatelli-Steinberg 2004).

Several investigators have also researched the timing of enamel formation so that the period of development of hypoplasias can be scored (Hillson 2005, Reid & Dean 2000, 2006). Different teeth measurements have also been examined in an attempt to understand how teeth

features like crown or cervical length are associated with each other so that they can be used in cases where one is not available (Hillson 2005). Investigations on the teeth formation times in several populations have compared enamel formation times between humans (Goodman &Rose 1991, Reid et al 2000, 2006).

Because of the many studies have been conducted on the formation of teeth, a variety of techniques and methods exist for the identification and recording of enamel hypoplasias. First, identification of enamel hypoplasias can be done on a macroscopic or microscopic scale. The macroscopic method identifies the defect with the naked eye (Hassett 2012). The microscopic methods identify the defect through an examination of the individual perikymata under 100 x magnifications (Hassett 2012). The length of a linear enamel defect is typically associated with the period of stress to the system. Each perikymata represents a period of growth from 7-10 days and varies between individuals. Disruptions to the perikymata can cause them to be abnormally spaced and the depth of the defects could signal a time of trauma.

Some researchers like Hillson highly recommend the perikymata counting method for there has been research that shows that the amount of perikymata matches closely with age (Hillson 2005). The microscopic method gives a more detailed picture of the tooth surface and catches minute changes. Perikymata can also give an accurate estimation of age at death for young individuals (King et al. 2005) and time of when the health disruptions occurred (Hillson 2005). However, several limitations exist on the microscopic method. First, it is time consuming and difficult. The teeth used need to be specially prepared or expensive casts need to be molded. The measuring microscope is often needed to record proper measurements. Also, while the microscopic method is more detailed, methods to match the spacing and growth of perikymata between teeth are not held constant between investigators.

The less detailed macroscopic method, which involves the naked eye, tangential light and needle nosed calipers is more accessible for researchers and is easy to employ in the field. Also, several growth charts exist to aid in the determination of enamel development (Reid & Dean 2006). Many researchers have led investigations of LEH with the macroscopic method, and it is possible to compare frequencies between populations. Also, from the investigation of Reid et.al in 2006, variation in enamel formation times between populations. This finding is a positive one for this study because it is easier to compare LEH development between human populations if the conditions for teeth growth are relatively universal. However, macroscopic surveys of LEH's do not always identify all cases of abnormal perikymata spacing and may not recognize all the individuals affected by enamel growth disruptions.

Many studies have worked with the presence of linear enamel hypoplasias in both living and deceased populations, and several hypotheses have been developed. First, because linear enamel hypoplasias tend to have a high prevalence in societies with nutritional or health stressors, there is an idea that with the prevalence of LEH can measure the biological toll of an environment. Thus, many studies have compared the contemporary skeletons from different locations to assess the toll of the environment (Garcin et al. 2010, Palubeckaité 2001).

One hypothesis exists that individuals with poorer health will have a greater amount of LEH present on the teeth. This "representationalist" position directly ties signs of stress on the body to stress in real life and the non-existence of stress on the body to no stress in life. King et al. (2005) have found in an investigation of crypt skeletons that individuals who died younger in life had an earlier age of first enamel hypoplasia occurrence than those who survived to adulthood. In another study, LEH prevalence has been associated with cohort specific stressor events (Littleton and Townsend 2005). Other researchers accept this hypothesis when analyzing

their data, and assume that the population with more LEH was more stressed and tended to die earlier.

While the hypothesis that more LEH means more disease and earlier death, an opposite hypothesis exists as well. That is the one of the "osteological paradox" (Wood 1992) which challenges the idea that no sign of stress on the body means a healthier individual. There is the idea that individuals that survive to old age will have more signs of linear enamel hypoplasia and other stressors on their body because they have been able to survive it. Individuals with less stress on their skeletons may have been too weak to survive the trauma and died, therefore missing out on the opportunity to have the stress written on their bodies. This hypothesis is supported by studies as well. In a piece of research by Klaus and Tam (2009) examining the systemic stress in a population late fifteen century Peru, it was found that the physically stressful colonial period with more stress and disease actually had less LEH present. It was possible that the reason for such a low prevalence was because the minor survivable health insults that caused past LEH had been exchanged for high mortality diseases that killed the individual instead.

An investigation based on the "Osteological Paradox" has been performed at the University of Pennsylvania as well. In 2005, Diana Smay-Toebbe researched the presence of inclusion bias in the Lower Mound cemetery sample of the Hasanlu collection and compared the findings with samples from the warfare population in High Mound Hasanlu to see if there was the inclusion bias that Wood described in his paper. Her findings found no statistical difference in the amount of linear enamel defects between the High Mound invasion population and the cemetery sample. Unfortunately, her results are complicated by the fact the she unwittingly included samples that belonged to Dinkha Tepe, another collection, in her analysis of Hasanlu.

Several other investigations on Hasanlu have taken place over the years. Ted Rathbun completed several theses and a book on Hasanlu. His studies covered morphological and paleopathological findings in order to connect the population at Hasanlu with other Iranian populations. Other research has been done on interpersonal violence and trauma (McCarthy, Perlin and Monge), craniofacial morphology differences for purposes of identification (Dulik) and aging and paleodemography (Selinsky 2009).

Both Page Selinsky (2009) and Diana Smay-Toebbe (2005) have been written dissertations on these collections, and both have surveyed the dental enamel hypoplasia to some degree. At the time of her dissertation, Smay-Toebbe (2005) may have unknowingly included samples from the neighboring Dinkha Tepe collection with her Hasanlu survey. Selinsky (2009) did not include the Dinkha Tepe collection. Thus, this survey of linear enamel hypoplasias from skeletons of both Dinkha Tepe and Hasanlu will allow for a better assessment of linear enamel hypoplasias in the entire skeletal collection from the site.

RESEARCH DESIGN/METHODOLOGY:

Study Population:

The skeletal samples studied belong to the Hasanlu site in northwestern Iran. The collection consisted of 263 individuals excavated from both High Mound and Lower Mound sites. These individuals vary from Period VII to Period III levels which cover a date range of 1400 BCE to 450 BCE. The bones were housed in boxes in the University of Pennsylvania Museum of Anthropology and Archaeology.

Unearthed from Iran during 1957 to 1977, the Hasanlu samples were shipped to several places. The early skeletons were first sent to the University of Kansas (Selinsky 2009) and then

split between the University of Tennessee and the University of South Carolina. The later samples (1965-1977) were sent to the University of Pennsylvania. In the late 1980's, all of the outside Hasanlu skeletons were sent to the University of Pennsylvania Museum where the remains were reunited.

Given the multiple homes of the collection and the multiple uses of the collection, the state of the skeletons and teeth vary greatly. Part of the collection has marked lines where supposed linear enamel hypoplasias are found. Some individuals have teeth whose enamel has been damaged or sampled. Also, postmortem wear and teeth loss has occurred. For the most part however, the teeth in the collection are relatively well preserved and available for study.

From the collection of 263 people, only 140 individuals had some mandible or maxilla parts present. The mandible/maxilla parts were examined for teeth type and wear. Teeth with less than half their original crown length or missing all of the canines and incisors were excluded. 72 samples fit the criteria for scoring and were consequently observed and measured. 3 deciduous samples were tested but did not show hypoplasias, thus the data collected was on permanent teeth only.

The Dinkha Tepe sample originally consisted of individuals from 94 burials. However, only 43 individuals were found in the collection and only 27 met the criteria for scoring.

Study Methods:

The entire skeletal collection was reviewed for the presence or absence of teeth. Photos of each skeleton that presented a mandible and/or a maxilla and/or isolated teeth were taken with a Nikon D40 with an AF-S Nikkor 18-135mm 1:3.5-5.6 ED lens. The photographs (Figure 4) were reviewed to examine which tooth types were present.

While the skeletons were labeled and an inventory of skeletal parts was available, the investigator was "blind" to which skeletons belonged to which burial. The measurements were taken first and then assigned their burial location.

Linear enamel hypoplasias were assessed macroscopically using the field method (Hassett 2012). The teeth were observed by both visual inspection and with 5x magnification under a light spot. For each set of teeth, the crown height and location of any linear enamel hypoplasias on all teeth types were recorded. The lengths were measured from the cement enamel junction (CEJ) to the target area using digital Mitutoyo sliding needle nosed calipers, and the measurement was recorded to the nearest tenth of a millimeter. The measurements for each individual were written on Ubelaker and Buikstra's Enamel Defects Recording form which uses the F.D. I Developmental Defects of Enamel (DDE) Index.

Sample Criteria

Only teeth with enamel were measured and recorded. Teeth with less than half of their crown length or with missing enamel were excluded. Samples with only molars and premolars were excluded due to the less demarcated defects on those teeth and the difficulty matching developmental times with molar hypoplasias. Samples with canines and incisors were favored due to the clarity of the linear enamel hypoplasias (Goodman & Rose 1991). All the linear enamel hypoplasias were found on permanent teeth due to the clearer demarcations on permanent dentition (Hillson 2005).

Scoring LEH and analyzing prevalence

Linear enamel hypoplasias were scored by one investigator. Because the linear enamel hypoplasia lines differ in severity, and the different lines were identified with the range of

identification criteria used by Guatelli-Steinberg (2004). The lower limit to identifying any LEH was the presence of any visible groove or line that was larger than the neighboring ones under a light with gentle magnification. The upper limit to LEH identification was prominent grooves or lines that are clearly visible without magnification (Guatelli-Steinberg et al, 2004). Although defects can be categorized (Cucina 2002), due to the tendency for LEH to be missed macroscopically, all defects were considered (Hassett 2012).

In order to calculate the overall presence of linear enamel hypoplasias (LEH) regardless of cause, all the samples with a LEH on at least two teeth were included.

For the LEH to be associated with systemic stress and deemed significant to overall health, the LEH timing had match in several teeth. More specifically the timing of the linear enamel hypoplasias should match between three anterior teeth or between a set of antimeres.

Usually, the strict criteria for the presence of systemic LEH is for the hypoplasia to match in all sets of teeth or at least a set of antimeres and one other tooth type (Goodman & Rose 1991).

However, since some samples miss all but 2 or 3 anterior teeth, the criteria were changed to a match in only one set of antimeres.

The time of development of LEH was estimated for these cases by dividing the location of the LEH by crown height and matching the resulting proportion with the enamel formation schedule by Reid and colleagues (Reid 2006). The enamel formation times chart used for the Hasanlu population was for Northern Europeans. No formal enamel formation chart exists for the Iranian population. It is likely that the population at Hasanlu is more similar to the Northern European population than the southern African one. While the timing of growth can vary up to a year between the two populations, human enamel formation times seem to be relatively similar

(Reid & Dean 2006). Thus, even with slightly different charts, it is still possible to roughly correspond which hypoplasias belong to which growth disruption. However, the age estimates are only rough estimations since the complete crown height may or may not have been present at the time. Also, because growth depends on the environment and genetics, it is population specific which means that it will not match perfectly to the Northern European sample times.

The hypoplasias were recorded for each individual and then checked with an inventory (Selinsky 2009) for demographic information such as sex and age category.

Data Analysis

The number of hypoplasias was compared between High Mound, Lower Mound and Dinkha Tepe. How the prevalence of LEH distributed among age groups and sex in High and Lower Mound was also calculated. The number of individuals with hypoplasias from High Mound Period IVB, the war invasion, and the number of individuals with hypoplasias from Lower Mound Period IV and earlier were compared. Chi-square tests were run to determine the significance of the differences between populations. Chi-square tests were also run for age and sex categories in the High and Lower Mounds of Hasanlu but not for Dinkha Tepe because no demographic data on Dinkha Tepe was available. The test was considered significant if the p-value < 0.05.

RESULTS

All but three samples in the Hasanlu collection demonstrated some degree of enamel defect. However, not all of the hypoplasia could be classified as linear enamel hypoplasia (LEH). The resulting LEH prevalence was 54.2% in High Mound Period IVB, 68.9% in Lower Mound, 80% in High Mound Period IIII, 59.4% in Dinkha Tepe, and 64% overall (Table 1,

Figure 5, Figure 6). When the chi square test was run, the difference in LEH amounts between High Mound IVB and Lower Mound (pvalue-0.225662) (Table 2) and between High Mound IVB, Lower Mound and Dinkha Tepe (p-value- 0.445793) (Table 3) was not statistically significant. Thus, no significant difference existed in the LEH distribution between the groups from the cemeteries and the one from the citadel which means that no sample inclusion bias was detected.

When the chi-square test was done onto the age categories from High Mound IVB (Table 6, Figure 10) and Lower Mound (Table 4, Figure 9), the High Mound IVB difference was not statistically significant (pvalue- 0.666211). The group from the Lower Mound however had a pvalue of 0.045392 which is less than 0.05. Therefore the null hypothesis is rejected, and there may be an association between LEH and the age categories in the Lower Mound. Looking at the distribution of LEH about the age categories, it appears that the LEH tend to be the most prevalent from children to young adults, but is less prevalent in mature and older adults. This fits with the observation by Goodman and Armelagos in 1988 that more LEH was associated with younger age of death.

When the chi-square test was used on the difference of LEH between sexes in High Mound IVB (Table 7, Figure 11), the difference was found to be not statistically significant (p-value 0.29953). However, when the chi-square test was used on the LEH difference between sexes in Lower Mound (Table 5, Figure 8), the difference was significant (p-value 0.000608). From the data, there seems to be a greater tendency for the males in the cemetery to show more linear enamel hypoplasia lines.

The average linear enamel hypoplasia events per individual were calculated between age categories in Lower and High Mound (Figure 9, Figure 12). The younger the individual the

greater number of enamel hypoplasia was found and is aligned with previous research (Smay-Toebbe 2005, Goodman & Rose 1991).

DISCUSSION

Hasanlu is an extraordinary collection for physical anthropologists to work with. The two cohorts of skeletons retrieved from High Mound and Lower Mound represent two very different life trajectories in contemporaneous times. The High Mound sample is representative of a catastrophe model. Most of the skeletons retrieved from that location, and that date back to Period IVB (1100-800BCE) fell during the invasion of the city. These people are believed to have been healthy before their deaths, and so the sample should capture what the typical stress markers of a "normal" population would look like.

The Lower Mound of Hasanlu consists mostly of a cemetery that has many burials, the majority of which date back to Period IV and V. Because the population in Hasanlu during Period IV and Period V seems continuous from the archaeological record, the Lower Mound sample seems to fit a typical attrition model where individuals died from being sickly. Thus, the comparison of stress markers between the two locations can reveal the biological effects of stress on the healthy and the sick.

Dinkha Tepe is another contemporaneous collection that is only 15 miles away from the Hasanlu site (Muscarella 1974). This is another cemetery collection dating to Hasanlu Period IV and Period V, but the burials were arranged slightly differently. Comparisons between the stress markers on the Dinkha Tepe sample with the Hasanlu samples could show the relationship between the locations and signal if any environmental or developmental differences exist.

In this survey, it was found that the linear enamel hypoplasia in all three sites were greater than 50% which indicates that there was significant overall stress in the environment. The LEH prevalence between the three sites was not statistically significant. This means that no significant difference in LEH existed between the supposed catastrophe model (High Mound) and the supposed attrition model (Lower Mound). This is in concordance with results from the investigations by Smay-Toebbe (2005) and Selinsky (2009). No significance between the High and Lower Mound LEH's means no great inclusion bias exists in the samples from all the locations. Because the numbers are not skewed toward more LEH in the cemetery or less LEH in the cemetery, the data neither supports the osteological paradox theory nor the representationalist theory. It may be that the sample size from Hasanlu is too small to capture the selection bias, or it may be that there is selection bias, but the bias varies between age categories and evens out overall.

The lack of statistical difference between the amounts of linear enamel hypoplasias in the two cemeteries, Dinkha Tepe and Lower Mound show that the two populations buried there are similar. Despite the differences in burial style, the individual from Hasanlu and Dinkha Tepe cemeteries seemed to live with similar environments, stressors and perhaps social rank since the data does not show any significant differences in health status.

In High Mound, no statistical significance was attributed to the age category or sex.

Linear enamel hypoplasia was just as likely to show between the young and the old, and between males and females.

In Lower Mound, there was a statistical significance in the different distributions of LEH between age categories, and between males and females. It seems that younger skeletons

(children, subadults, and young adults 35 years or less (Selinksy 2009)) were more likely to be affected by LEH in the cemetery than the older ones. This finding implies that the people who died young were likely to suffer stressors early in their lives, and this idea is in accordance with the representationalist hypothesis of LEH appearance. This finding is in contrast with the argument produced by Selinsky (2009) who found more LEH in High Mound and finds support for Wood's (1992) Osteological Paradox Hypothesis.

The statistical significance between the amount of LEH in males and females in Lower Mound might support an association between males who suffer early childhood stress and dying. Perhaps only the males who had significant irreparable stress events would die while the others without childhood stress would live. This difference in LEH and sex was similar to the finding of Palubeckaite (2001) who saw in his Lithuanian sample that males tended to have more LEH than females. However, unlike the one here, Palubekcaite's difference was not significant. A statistical significance indicates an association but not a specific meaning. Therefore the explanation for why in Lower Mound, all the men had linear enamel hypoplasia present is up for speculation.

The information retrieved from this study has various limits. First, with only one researcher, the data is collected using the judgments of only one person. Thus, some hypoplasia may be over or undercounted, and the mistake unchecked by another person. As mentioned before, the skeletal collections of Hasanlu and Dinkha Tepe are not full, and thus may skew the results in a direction that is unknown. Some teeth are missing, and it is harder to interpret the effects of stress on only a few teeth.

In terms of measurements, it is difficult to find samples with full crown heights. Hence, the overall measurements and timing of LEH may be off, which could drastically change how many LEH are matched, between teeth and fit the LEH criteria. Also, the crown height shows slight variations between populations and between genders (Reid & Dean 2006). Because there is no data table for Iranian enamel formation times, the corresponding age that relates to the distance from the cement enamel junction to the defect was calculated using a Northern European sample. Finally, while statistical correlations or non-correlations are interesting to note, the sample size is quite small and could skew the data.

Further studies could involve multiple researchers taking the same measurements on the teeth of these skeletons once more, a full analysis of the demographic traits of the Dinkha Tepe collection, or perhaps an attempt at a microscopic view so that the arguments regarding how much LEH is prevalent in teeth in the collection can finally be resolved.

CONCLUSIONS

The aim of this study was to survey the teeth in the Hasanlu and Dinkha Tepe collection for the presence of linear enamel hypoplasia and to compare the results for any statistical associations that may arise within and between the groups. The study found that the overall sum of linear enamel hypoplasias was not significantly different between populations. The hypoplasia for High and Lower Mound Hasanlu could thus be combined. The Dinkha Tepe hypoplasia numbers were also combined with Hasanlu since the populations were located close together and contemporaneous with one another. The overall LEH prevalence calculated was 64% (Figure 4) and can be a representation of the LEH in the multiple tepes of the Early Iron Age in the Solduz River Valley region.

The 64% prevalence of LEH in the Hasanlu/Dinkha Tepe region can be compared to the prevalence of LEH in other contemporary societies in the region. In a study on Early Iron Age skeletons (1100-900 BCE) from Pella, Jorden, Griffin and Donlon (2007) found a linear enamel hypoplasia frequency of 47.5% on incisors and 88.7% on canines. These statistics flank the 64% found in Hasanlu/Dinkha Tepe. Comparison of the two numbers shows the LEH in both populations were similar to one another signaling that high prevalence of LEH may have been typical for that time in the Middle East. This may be a reflection of the environmental conditions in the site. Though at the time of the invasion, there was a significant harvest, perhaps there was a higher disease load or inadequate nutrition in the area.

While 64% of a population affected by LEH is rather high, it is by no means the highest. Some studies have found LEH's in 99% of the population studied (Méndez Colli et al. 2009). Others studies have lower LEH rates. Guatelli-Steinberg (2004) found LEH's affecting only 38.2% of Neanderthal skeletons and 38.1% of modern Inuit foragers. In developed modern human populations, the frequency of hypoplasis is usually less than 10 percent of the population (Goodman & Rose 1991). Thus, Early Iron Age Hasanlu seems more stressed than modern populations today. However, the LEH frequency in a population is highly dependent on the environment. Thus, context should always be considered when developing explanations.

The information found in this study provides clues to the health of the Hasanlu and Dinkha Tepe populations as well as similarities in their health status. These results can support further investigations into the populations studied.

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Figure 1.

Period	Dates
I	13m and 14w cent. CE
II	c. 300-c. 275 BCE
IIIA	c. 600 - c. 300 BCE
IIIB	c.750-c. 600 BCE
IVA	c. 800-750 BCE
IVB	c.1100- c. 800 BCE
IVC	c.1250-c. 1100 BCE
V	c. 1450 - c. 1250 BCE
VI	c. 1600 - c. 1450 BCE
VII	Late 4m cent c. 1600 BCE

Figure 2. From Muscarella 1974 Bga, burial 17



Figure 3.

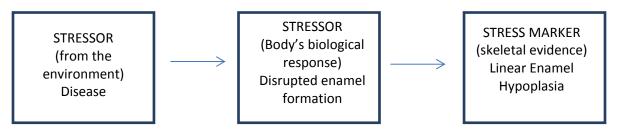


Figure 4.



Figure 5.

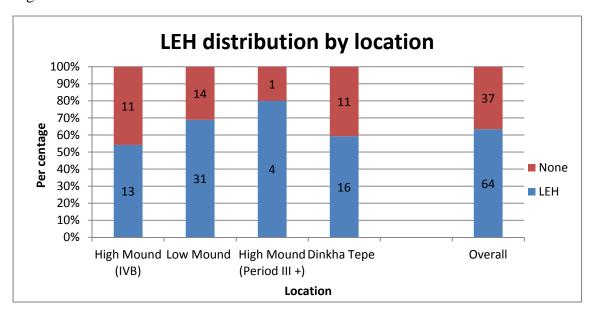


Figure 6

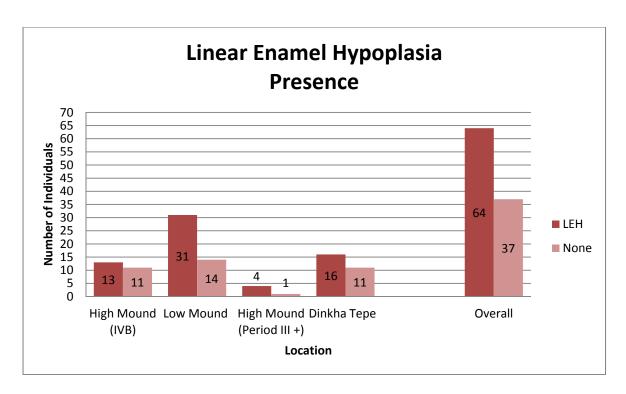


Figure 7

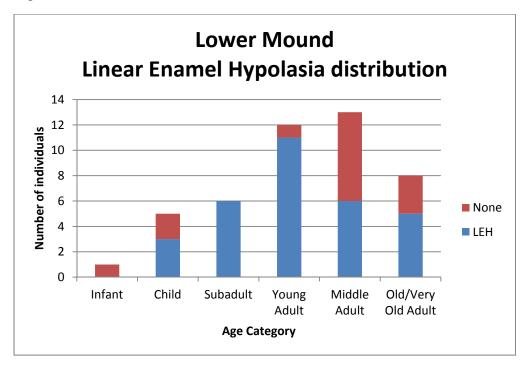


Figure 8

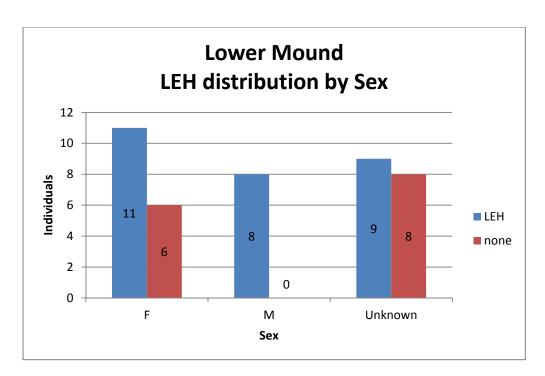


Figure 9

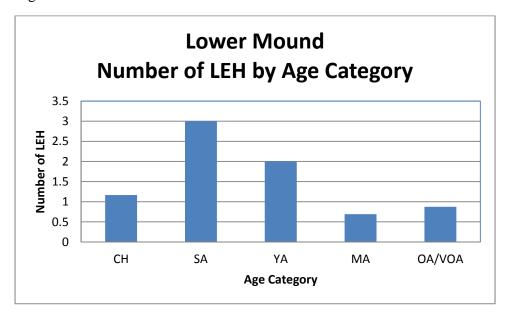


Figure 10

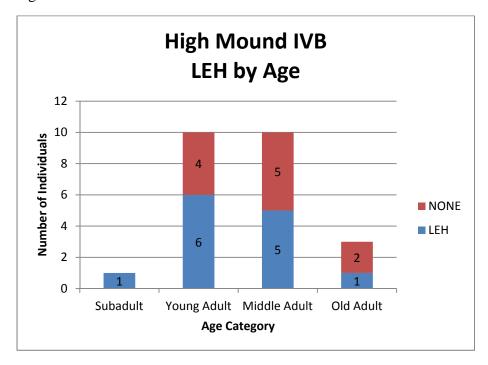


Figure 11

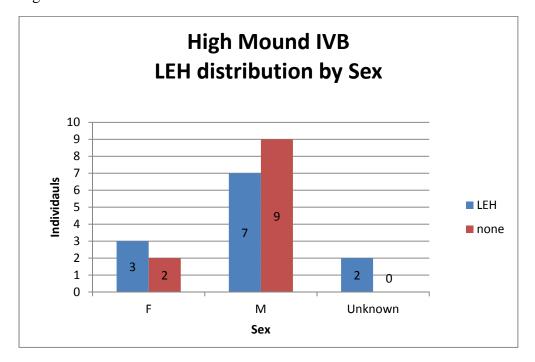


Figure 12

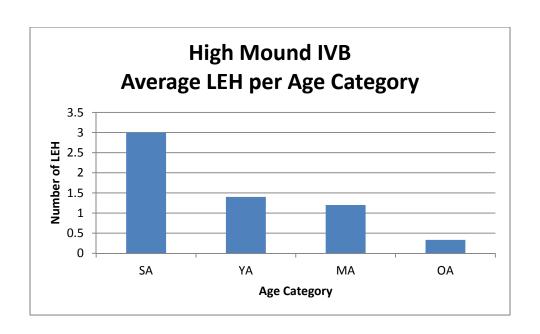


Table 1.

Observed	LEH	None	Total Number
High Mound (Period	13 (54.2%)	11 (45.8%)	24
IVB)			
Lower Mound	31 (68.9%)	14 (31.1%)	45
High Mound (Period III	4 (80%)	1 (20%)	5
and earlier)			
Dinkha Tepe (66's,	16 (59.3%)	11 (40.7%)	27
69's), 59-4-102 and 59-			
4-110			
Total	64	37	101

Table 2.

<u>Chi-square test</u> Between High Mound and Lower Mound hypoplasias

Observed	LEH	None	Total Number	Expected	LEH	None	(O-E)^2/E	
High Mound (IVB)	13	11	24	High Mound (IVB)	15.304 35	8.6956 5	0.346961	0.61065
Low Mound	31	14	45	Low Mound	28.695 7	16.304 3	0.18505	0.32568
Total	44	25	69				df= 1	X2= 1.468
							p-value= 0.225662	

Table 3.

<u>Chi-square test</u> Between High Mound, Low Mound and Dinkha Tepe

Observed	LEH	None	Total Number	Expected	LEH	None	(O-E)^2/E	
High Mound (IVB)	13	11	24	High Mound (IVB)	15	9	0.266667	0.444444
Low Mound	31	14	45	Low Mound	28.125	16.875	0.293889	0.489815

Dinkha Tepe	16	11	27	Dinkha Tepe	16.875	10.125	0.04537	0.075617
Total	60	36	96				X2=1.6158	df=2
							p-value=	
							0.445793	
							Not	
							significant	

Table 4.

<u>Chi-square test</u> Between age categories in Lower Mound

Observed	LEH	NONE	Total	Expected	LEH	NONE	(O-E)^2/E	
								1.53580
IN	0	1	1	IN	0.69	0.31	0.69	6
								0.12410
CH	3	2	5	СН	3.44	1.56	0.056279	3
SA	6	0	6	SA	4.13	1.87	0.846707	1.87
								1.99809
YA	11	1	12	YA	8.27	3.73	0.901197	7
								2.16871
MA	6	7	13	MA	8.96	4.04	0.977857	3
								0.10445
OA/VOA	5	3	8	OA/VOA	5.51	2.49	0.047205	8
TOTAL	31	14	45				$X^2=11.32$	df=5
							p-value=	
							0.045392	
							<0.05	
							Significant	

Table 5

<u>Chi-square test</u> Between sex categories in Lower Mound

Obs.	LEH	none	Total	LEH(expected)	None(expected)	
F	11	6	17	19.04	9.52	
М	8	0	8	8.96	4.48	
U	9	8	17	19.04	9.52	
						p-value
Total	28	14	25	X2=14.81	df=2	0.000608

Table 6

<u>Chi-square test</u> Between age categories in High Mound

Obs	LEH	NONE	Total	Ехр	LEH	None	(O-E)^2/E	
						0.45833		0.45833
SA	1	0	1	SA	0.54167	3	0.38782	3
						4.58333		0.07424
YA	6	4	10	YA	5.41667	3	0.06282	2
					5.41666	4.58333		0.03787
MA	5	5	10	MA	7	3	0.03205	9
								0.28409
OA	1	2	3	OA	1.625	1.375	0.240385	1
Total	13	11	24				X2=1.57	df=3
							p-value=	
							0.666211	
							Not	
							significan	
							t	

Table 7

<u>Chi-square test</u> Between sex categories in High Mound

Obs.	LEH	none		Expected	LEH	None	
F	3	2	5	F	2.608696	2.391304	
М	7	9	16	М	8.347826	7.652174	
U	2	0	2	U	1.043478	0.956522	
							p-value
Total	12	11	23		d=2	X2=2.41108	0.29953