Palaeopathological Analyses of Human Skeletons: Statistical Analysis of Health and Disease among the Chalcolithic and Bronze Age Populations at Tepe Hissar

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Abstract: During the approximately 2500-year history of the site (ca. late-5th to early-2nd millennium BCE), Tepe Hissar experienced a number of cultural, economic, and social transformations, which were accompanied by conflict, population movements, an increase in industrial activities, as well as episodes of site abandonment and reoccupation. This study investigates the impact of these socio-cultural-economic changes on the health of the people buried at Tepe Hissar. This study examines the pathological indicators of nutritional stress and skeletal metabolic disease using a sample of 313 adult individuals, stratified by sex and age, as well as by chronology. The chi-square test was employed to compare the significant differences in the prevalence of metabolic disease within and between the Tepe Hissar populations grouped by sex and age category. For those samples too small to be subjected to chi-squared significance testing, Fisher's exact test was used. Both calculations were conducted in Excel and SPSS 20. The results of these tests showed that males and females of all age groups from each of the three periods at Tepe Hissar experienced similar episodes of health and stress during their lives. Specifically, approximately half of the individuals from each period experienced some form of metabolic skeletal disease. Statistical analyses did not show any significant increase or decrease in the prevalence of nutritional stress or other metabolic diseases among this population between periods. The results of this research indicate that the socio-cultural-economic changes and events that occurred at this site, particularly during Hissar II and III, did not have a significant impact on the general health and disease of the population. This research suggests that the society at Tepe Hissar had access to similar levels of food resources and experienced similar living conditions over time from the late-5th to the early-2nd millennium BCE. The high rate of metabolic bone diseases in each period, however, could have been due to exposure to toxic elements such as lead and arsenic which were widely utilized in metallurgical production at Tepe Hissar throughout its history.

Keywords: Metabolic Diseases, Human Skeletal Remains, Chalcolithic and Bronze Age, Lead and Arsenic Poisoning, Tepe Hissar.

Introduction

The concept of stress has been defined as the repetitive physiological reaction of individuals and populations to a disruptive variable or combination of variables and insults, and has much in common with the concept of adaptation (Buikstra and Cook 1980; Bush and Zvelebil 1991; Goodman *et al*. 1988). Stress is used in a variety of contexts in anthropological research, e.g. nutritional, environmental, mechanical or functional, psychological, and physiological (Bush 1991; Goodman 1991; Larsen 1987). Factors such as a change in subsistence patterns, food shortage leading to famine, malnutrition, diseases, a change in socio-cultural structures and lifestyle and economy, political instability, environmental degradation, industrial activities, migration, increase in population size, and climate change have significant impacts on a population exposed to these kinds of stressors (Curtis *et al*. 2005; Goodman 1991; House *et al*. 1994; Pearlin *et al*. 2005; Temple 2007; Turner and Avison 2003). However, these factors might often interact with each other and in so doing can worsen other factors. Stress as a physiological disruption cannot be directly measured; however, different skeletal and dental changes may be used to infer stress (Goodman *et al*. 1988). If physiological stress becomes chronic then it may interrupt growth and leave its signature on bones and teeth, but it depends on the type of stressor involved (Brickley and Ives 2008; Buikstra and Cook 1980; Duray 1996; Goodman *et al*. 1988). For example, health

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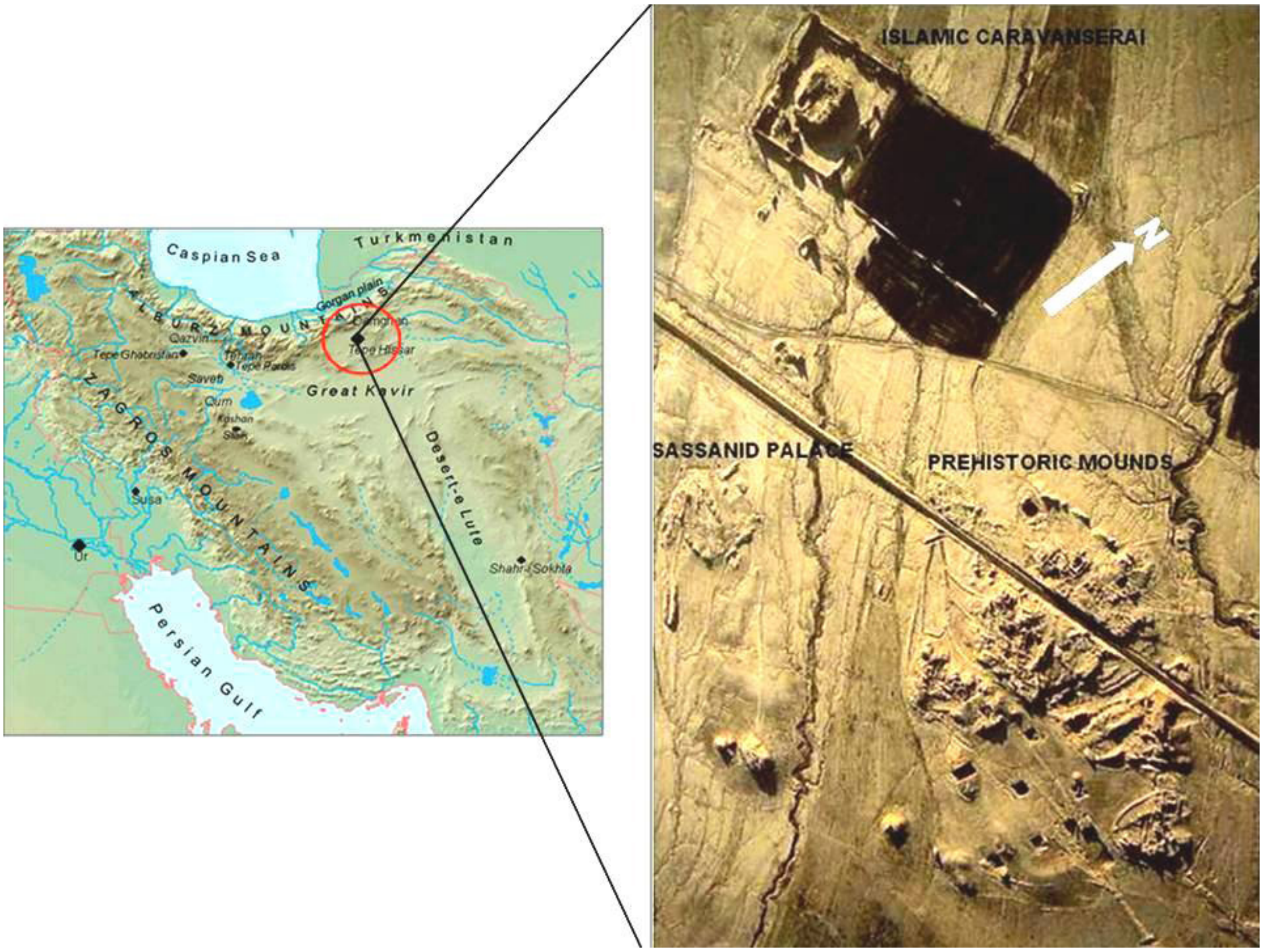

Fig. 1. Map of Iran- geographic location of Tepe Hissar and aerial view of the site of Tepe Hissar (Mousavi and Sumner 2012: Pl.19)

related stressors such as dietary deficiency (malnutrition or nutritional deprivation) can cause metabolic bone disease with different pathological lesions on both bones and teeth (Aufderheide *et al*. 1998: 405; Ortner *et al*. 2001; Stuart-Macadam 1989). The study of multiple indicators of skeletal and dental stress, in combination with other types of data, such as is found in archaeological, historical and epidemiological sources, has an important position in bioarchaeological research for assessing subsistence and dietary changes and general levels of health in prehistoric and historic communities (Goodman 1993; Keenleyside, 1998).

This research investigated the bioarchaeological pattern of nutritional stress and metabolic bone diseases among Tepe Hissar populations to test the hypothesis that the socio-cultural-economic changes that occurred at Tepe Hissar over time had significant impact on health and nutritional status of people. The population of Tepe Hissar experienced widespread cultural and economic changes, as well as interpersonal conflict/violence, and population movements during their lives, over an interval of ca. 2500 years (Afshar 2015; Afshar *et al*. in press; Dyson and Howard 1989; Schmidt 1933, 1937). There is currently no published information regarding nutritional stress and metabolic disease from this site or from other archaeological sites in Iran for comparative study, and therefore this study will be a landmark for future bioarchaeological and palaeopathological research in Iran.

The site of Tepe Hissar

Tepe Hissar represents one of the largest known settlements in the north-eastern Central Iranian Plateau (Fig. 1). It is located along the southern slopes of the Alburz Mountains on the Damghan Plain, and sits astride a major trade route of the "Silk Road" which connects Central Asia in the East to Mesopotamia and the Persian Gulf in the West (Pigott *et al*. 1982). The archaeological materials and C14 dating show the site was inhabited during the late 5th to the early 1st millennium BC through the historic phases (300-600 AD) to the Islamic period (middle period- Schmidt 1933, 1937; Dyson and Howard 1989; Roustaei 2010).

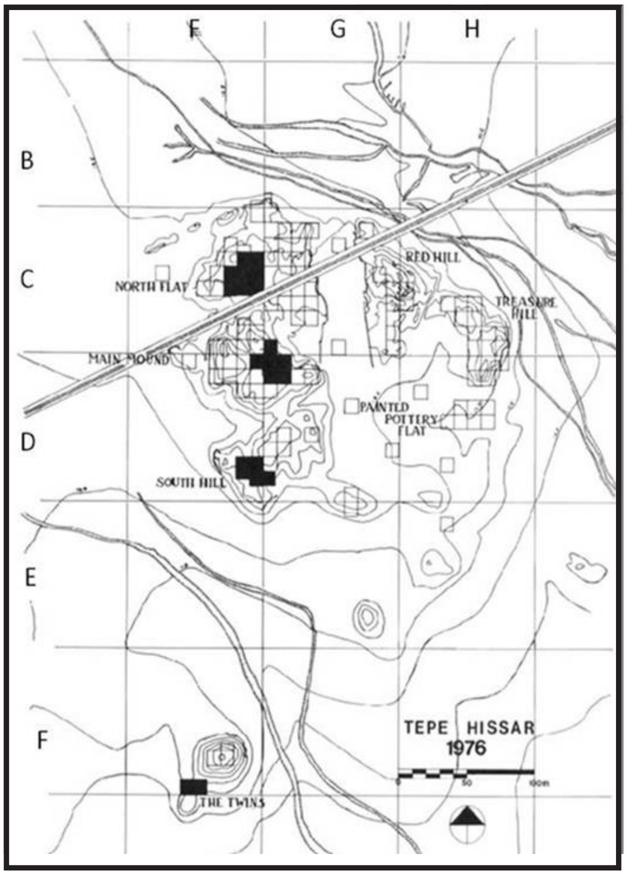

Fig. 2. Plan of Tepe Hissar excavations: black squares (restudy team in 1976), and white squares (Schmidt 1933- Dyson and Tosi 1989)

In 1931-32 and 1932-33, before the Second World War, the prehistoric mound of Tepe Hissar was first systematically excavated by the University of Pennsylvania Museum under the direction of Erich Schmidt (Schmidt 1933, 1937). Based on seriation of pottery types found in the 1637 graves excavated, Schmidt (1937) identified three periods that were subdivided into 8 sub-periods: IA, IB, IC, IIA, IIB, IIIA, IIIB, and IIIC (Fig. 2). In 1971, Sumner, and in 1974 Sumner, Howard, Tosi and Bagherzadeh attempted to find more information about the stratigraphy, architecture, technology and ecology of Tepe Hissar (Dyson 1985). In 1979, a re-investigation project was undertaken by the University of Pennsylvania Museum, Turin University, and the Iran Centre for Archaeological Research (Dyson and Howard 1989). In more recent times in 1995, 2006 and 2010, research was carried out solely by the Iranian team, directed by Yaghmaei and Roustaei (Roustaei 2010). Archaeological evidence shows that, during the Chalcolithic and Bronze Ages (late 5th to the 2nd millennium BC), the site experienced widespread cultural and economic changes represented by transformations in its material culture, mortuary practice, technology and crafts produced, long distance exchange networks, as well as interpersonal violence and conflict. Site abandonment and reoccupation occurred periodically at the site, and has traditionally been explained by the "arrival" of new populations in this locality (Afshar 2015; Afshar *et a*l. in press; Dyson and Howard 1989; Schmidt 1933, 1937). During its life the population of the site, however, experienced stressful situations.

Archaeological evidence for the three Tepe Hissar periods

Hissar I Period (ca. 4300-3700 BC) – Painted Pottery: The archaeological sequence at Tepe Hissar indicates a sudden appearance and development of the settlement (Hissar I period) in the late 5th millennium BC (Schmidt 1937; Dyson and Howard 1989). The archaeological evidence indicates that the first residents of this site were sedentary agricultural people, who lived in houses made up of small and medium sized rooms and poorly built with mudbrick (chineh) (Schmidt 1933: 341-2). The main pottery of this period was "painted pottery", with advanced art, elaborate decoration and standardized forms; it was handmade in the earlier phase (IA) but turned to wheel made later on (Schmidt 1933: 344; 1937: 39). The archaeological evidence from the earliest settlement of Tepe Hissar showed an elaborate cultural assemblage indicating considerable wealth and craft specialization (Pigott *et al*. 1982). Schmidt (1937: 300) indicates that the appearance of spindle whorls could be associated with spinning of wool and the production of woollen clothing in this period. Copper objects uncovered at Hissar I show that the early settlers of this site, in addition to pottery making and stone working, were also familiar with copper implements and copper making technology in the late 5th millennium BC (Schmidt 1937: 301 PL.XVI- Fig. 2.5).

Hissar II Period (ca. 3700-2900 BC) – The Introduction of Grey-Ware: During the early 4th millennium BC, Tepe Hissar entered a new period. The late Hissar I period gave way to Hissar II, distinguished by the appearance of "unpainted dark grey pottery" (Schmidt 1937). This was followed by a remarkable increase in industrial activities, workshops, new materials (e.g. clay counters, accounting artefacts, weapons), a large scale production of metal objects (e.g. copper) and semi- precious beads/ lapis-lazuli (Schmidt 1933, 1937; Tosi 1989), and the growth organization and control of production through administration (Dyson, 1987; Dyson and Remsen 1989: 79; Tosi and Bulgarelli 1989: 40-41). It is believed that changes from painted to grey pottery were not without external influence (McCown 1942: 11), and were perhaps due to large scale infiltration or invasion of people (Schmidt 1937: 302).

Objects including "mullers", "hand grinder", and "mortars" discovered from this period (Schmidt 1933), suggest that people practiced agriculture as in previous periods and this corresponds to the archaeobotanical analyses (Costantini and Dyson 1990) and recent bioarchaeological studies of human remains from this site (Afshar 2015). The results of investigation of industrial debris on the surface of Tepe Hissar in 1976 testified that this site was an important manufacturing centre with intensive development of handicrafts and other activities during the mid 4th millennium BC (Hissar II). Various industrial areas showed metallurgy, semi-precious stone making (lapis lazuli, beads, soapstone, etc.), and pottery making, all of which were active simultaneously in different combination over many centuries at Tepe Hissar (Tosi 1989: 14). Workshops were located in "open areas" on abandoned structures of earlier periods and far from the main settlement (Tosi and Bulgarelli 1989: 44-50). Nevertheless, the evidence of fire, ashes, "burnt human remains", "violence- related skull trauma", new forms of copper weapons, and destruction evident in buildings dated to Hissar II suggests that these cultural changes may have been accompanied by "traumatic" events, particularly at the end of this period (IIB ~3400 BC- Late Chalcolithic) (Afshar 2015; Afshar *et al*. in press; Schmidt 1937). It is suggested that these changes impacted the health, well being and the diet of Hissar II people. Nevertheless, the site during Hissar II finally "collapsed" in the early 3rd millennium BC and gave way to the distinctive Hissar III period, suggesting that a "dynamic force" or "foreign influence" changed life for the people there (Schmidt 1937). The site was abandoned and used as a burial ground for a short period, but was reoccupied again with new people (Tosi and Bulgarelli 1989:44).

Hissar III Period (ca. 2900-1800 BC): Hissar III is the era of burnished/incised grey pottery, and the majority of the grey ceramics were handmade as opposed to the wheel-made pottery from Hissar II (Schmidt 1933: 393, 1937: 179-180). Archaeological finds from Hissar III demonstrate intensive craft specialization, an advanced culture, and social differentiation during the mid-3rd millennium BC (Dyson and Remsen 1989: 95-96; Howard 1989: 68; Schmidt 1937). At this time Tepe Hissar was occupied by specialized craftsmen smelting copper, and working interesting objects in gold, silver, lead, lapis lazuli, carnelian, turquoise, alabaster, and many other materials during the 3rd millennium BC (Schmidt 1933, 1937; Tosi 1989). At the end of the third millennium BC (late Hissar III), there is evidence of new forms of pottery, a new burial tradition (e.g. warriors, rich burials), as well as new objects, for example alabaster vessels and copper "wands" (sticks) in this phase (Schmidt 1937: 182). The occurrence of specific archaeological material in late Hissar III, such as Central Asian cultural evidence (BMAC, e.g. alabaster, calcite objects, miniature columns), and "warriors" suggests movements of people from Central Asia, but this also indicates more of an intrusion and organized warfare rather than "trade" (Hiebert 1998: 151-155; Hiebert and Lamberg-Karlovsky 1992; Lamberg-Karlovsky 2002; Parpola 1998: 124).

Nevertheless, the evidence of burning and destruction in buildings from the beginning of the Hissar III period, "charred" human skeletal remains, "violence- related skull trauma", the occurrence of several "mass burials" or "communal burials", with some containing only several adult skulls only, while others included a number of disarticulated/"interlocked" skeletons of adult individuals, the appearance of some single burials with missing skulls, or burials with only a skull, and the appearance of advanced forms of copper weapons and the "Warrior, all suggest that during Hissar III, particularly from the middle to late Hissar III period, the site may have experienced intra- or inter group conflict/violence and cultural changes during Hissar III may have not been peaceful (Afshar 2015; Afshar *et al*. in press; Dyson and Remsen 1989: 97; Schmidt 1933, 1937; Schmidt's unpublished archive).

At the end of the Bronze Age (early 2nd millennium BC), most sites on the Iranian Plateau, southern Turkmenistan, and the Indus Valley were abandoned for the first time in centuries (Hiebert 1998: 151). The Bronze Age settlement of Tepe Hissar was abandoned (Schmidt 1937: 308) at the same time as the fall of Turang Tepe and Shahr-i Sokhta in north-and south-east Iran, and Altyn-Tepe and Namazga-Tepe in Turkmenistan, as well as the collapse of both capitals of the early Indus-valley civilization, Harappa and Mohenjo-Daro (Masson 1988: 135).

The questions to be explored in this study are: did the cultural transformations and events that occurred during different time periods at Tepe Hissar affect the health and well-being of people particularly those from the Hissar II and III periods? What health problems did they suffer from? Were there differences between males and females or between different age groups? The hypotheses derived from these questions are: 1) the socio-cultural and economic transitions that occurred at Tepe Hissar, and particularly in Hissar II and III, did have an impact on the health and well-being of the population of the site; 2) the health outcomes at Tepe Hissar during these two periods differed between males and females, as well as between different age groups.

Materials and Methods

The skeletal collection from Tepe Hissar is curated at the University of Pennsylvania Museum of Archaeology and Anthropology, Philadelphia, USA (UPM). This collection after Shahr-i Sokhta (curated at the site in Iran), is one of the largest human skeletal collections available from Iran. From the 1637 skeletons uncovered during Schmidt's excavations (1933-1937), 397 skeletons have been housed at UPM since that time, but the rest of the skeletal remains are in an unknown location in Iran. The first systematic bioarchaeological research on human skeletal remains

Table. 1. Hissar I: Metabolic disease profile by sex

Disease	Male	Female	Indeterminate	Total	Comparison	
					X2	P
Cribra Orbitalia	1/5 (20%)	1/1 (100%)	-	2/6 (33%)	2.4	0.333
Porotic Hyperostosis	4/5 (80%)	0/1 (0%)	-	4/6 (67%)	2.4	0.333
Vitamin C Deficiency	0/10 (0%)	0/6 (0%)	0/3 (0%)	0/19 (0%)	-	-
Total Vitamin D Deficiency	3/7 (43%)	3/5 (60%)	1/3 (33%)	7/15 (47%)	0.612	0.736
Residual Rickets/Osteomalacia	2/6 (33%)	3/5 (60%)	1/3 (33%)	6/14 (43%)	0.933	0.627
Osteopenia/Osteoporosis	1/10 (10%)	3/6 (50%)	1/3 (33%)	5/19 (26%)	3.185	0.203
Total Metabolic Diseases	4/10 (40%)	4/6 (67%)	1/3 (33%)	9/19 (47%)	1.351	0.509

P≤0.05

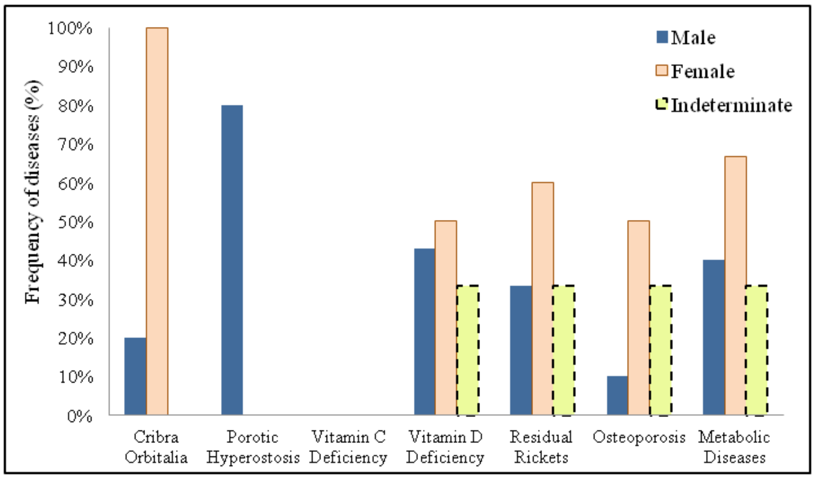

Fig. 3. Hissar I: Metabolic disease profile by sex

from Tepe Hissar was conducted by the author as a doctoral thesis and this article is a small part of her PhD research (Afshar 2015). The focus of this research was the human skeletal remains dating from the early Chalcolithic to the Bronze Age (late 5th- 2nd millennium BC). While there was a total of 368 adult individuals available for study from these periods, indicators of nutritional stress and skeletal metabolic diseases was examined in 313 adult individuals preserved and available for study from the three periods (Hissar I=19, Hissar II=43 and Hissar III=251), by the author. The age categories utilized in this study were based on Buikstra and Ubelaker's (1994: 36) recommendations, but to obtain more nuanced information, the young adult class was divided into two: young adult 1 (YA1, 18-25 years), young adult 2 (YA2, 26-35 years), middle adult (MA, 36-50 years), old adult (O, 50+), and adult (AA, 18+).

Malnutrition or nutritional deprivation is recognized as a major factor for metabolic bone disease occurrence (Dunnigan and Henderson 1997; Holick 2006; Ortner *et al*. 1999). The presence or absence of skeletal indicators of stress and/or dietary deficiency included, porotic hyperostosis and cribra orbitalia (Aufderheide *et al*. 1998; Stuart-Macadam 1991), Vitamin C deficiency, Vitamin D deficiency (including residual rickets/osteomalacia), and osteopenia/osteoporosis (Brickley 2000; Brickley and Ives 2008; Brickley *et al*. 2005, 2010; Holick 2005; Miller *et al*. 1996; Ortner 2003; Resnick and Niwayama 1995) were recorded in each skeleton where present. The chi-square test was used for comparing the significant differences in the prevalence of metabolic disease rates within and between the Tepe Hissar population groups by sex and age category. In the case of small sample sizes not meeting the assumptions of the chi-square test, Fisher's exact test was used (Fletcher and Lock 2005). These calculations were conducted both in Excel and SPSS 20. Only p-values less than 0.05 were considered significant.

Results

Hissar I: Table 1 and Fig. 3 demonstrate the metabolic disease profile for Hissar I by sex. The prevalence of metabolic disease and nutritional stress was examined in 19 individuals (male=10, female=6, indeterminate=3), and nine of those showed pathological bone changes consistent with this condition (47%). The prevalence of metabolic disease in total was higher among females than males. But the statistical analysis showed this was insignificant. Comparing the prevalence of vitamin C deficiency, there was no evidence of this deficiency.

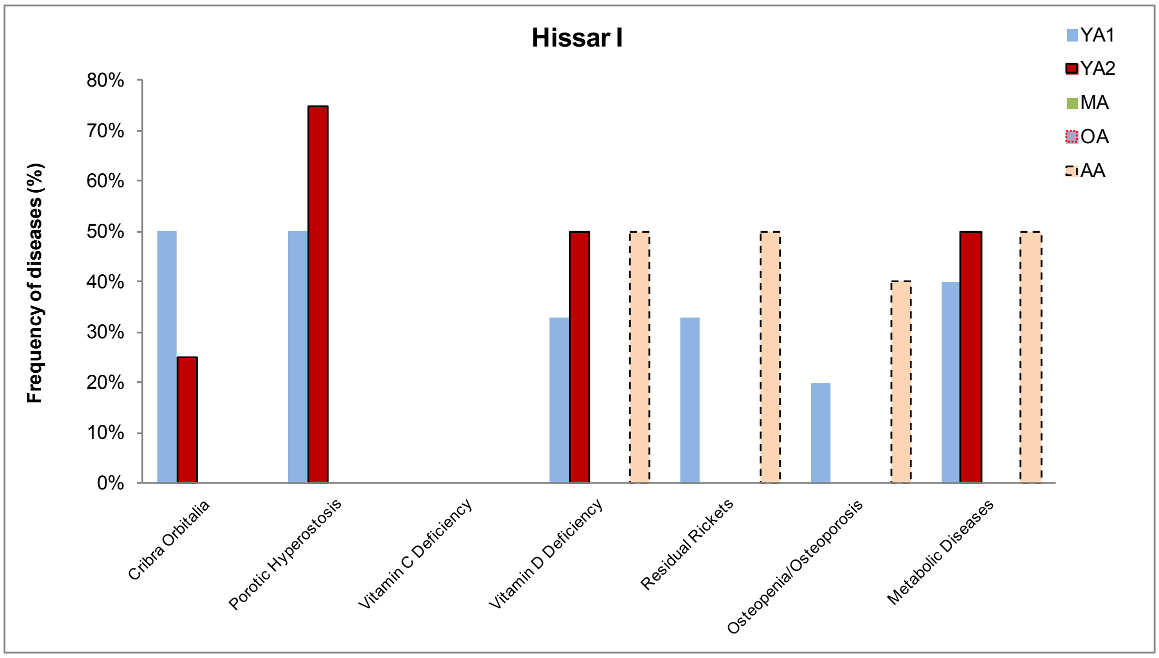

Fig. 4. Hissar I: Metabolic disease profile by age-category

Almost 47% of Hissar I individuals showed bone changes indicative of vitamin D deficiency. The percentage of vitamin D deficiency was 17% higher among females than males (insignificant). The prevalence rate for residual rickets/or osteomalacia in females (60%) was higher than in males (33%) (insignificant). Nineteen individuals were observed for osteopenia/osteoporosis, and almost 26% were affected. However, females were affected 40% more than males (insignificant). Statistical analyses showed all differences between both sexes were minor.

Nine of the 19 individuals preserved for observation were younger than 35 years old (10 individuals belonged to the unaged category (AA)) (Fig. 4). As shown on Fig. 4, the YA2 showed a higher prevalence of pathological lesions related to metabolic disorders compared to YA1, but the statistical analysis indicate these differences were insignificant and small. It is suggested that both YA1 and YA2 from Hissar I were affected equally.

Hissar II: Table 2 and Fig. 5 exhibit the metabolic disease profile for Hissar II by sex. There were 43 individuals preserved well enough to record metabolic diseases in this period (male=15, female=25, indeterminate=3). Thirty individuals showed pathological bone changes consistent with metabolic disorders (70%). The prevalence of metabolic diseases overall was 2% higher among the males compared to females, but it was not statistically significant. This data suggests that both sexes were affected equally.

Among 43 individuals observed for lesions indicative of vitamin C deficiency, none of them showed evidence of scurvy. Almost 61% of individuals showed bone changes indicative of vitamin D deficiency, with the highest prevalence found in males (69%) compared to females (60%) (insignificant). Males demonstrated a higher rate of residual rickets/osteomalacia (54%) compared to the females (50%) (insignificant). Females were 12% less affected with osteopenia/osteoporosis than males (insignificant). These data indicate that differences between both sexes were small and males and females from Hissar II experienced similar rate of nutritional stress and metabolic disorders.

Fig. 6 summarise the prevalence of metabolic diseases in 43 adults by age-category. The prevalence of metabolic disease in this period increased with age, and the OA group showed the highest rate (100%). The prevalence rate was considerably higher among YA1 and YA2, however, statistical tests did not show significant difference. The statistical analysis indicates that all age groups at Hissar II experienced similar rate of stress and metabolic disorders.

Hissar III: Table 3 and Fig. 7 demonstrate the metabolic disease profile from Hissar III by comparing the prevalence of disease between the sexes. Of 251 individuals were preserved well enough to observe the indicators of metabolic disease, 185 showed pathological bone changes consistent with these diseases (74%). The females of this period showed the highest prevalence rate (77%) when compared to the males (71%), however, statistical tests showed these differences were small and not significant. There were 120 individuals with at least one observable orbit, and 66 of those individuals showed CO. The females displayed a higher prevalence of CO (63%) compared to males (49%) (insignificant).

A total of 68% of individuals were affected by Vitamin D deficiency; the rate was identical in males and females (68%) but at 67% for unsexed individuals (insignificant).

Table. 2. Hissar II: Metabolic disease profile by sex

Disease	Male	Female	Indeterminate	Total	Comparison	
					X2	P
Cribra Orbitalia	7/7 (100%)	7/11 (64%)	-	14/18 (78%)	3.273	0.119
Porotic Hyperostosis	6/7 (86%)	9/11 (82%)	-	15/18 (83%)	0.47	1
Vitamin C Deficiency	0/15 (0%)	0/25 (0%)	0/3 (0%)	0/43 (0%)	-	-
Total Vitamin D Deficiency	9/13 (69%)	12/20 (60%)	1/3 (33%)	22/36 (61%)	1.345	0.51
Residual Rickets/Osteomalacia	6/11 (54.5%)	9/18 (50%)	1/3 (33%)	16/32 (50%)	0.424	0.809
Osteopenia/Osteoporosis	6/15 (40%)	7/25 (28%)	1/3 (33%)	14/43 (32%)	0.616	0.735
Total Metabolic Diseases	11/15 (73%)	18/25 (72%)	1/3 (33%)	30/43 (70%)	2.038	0.361

P≤0.05

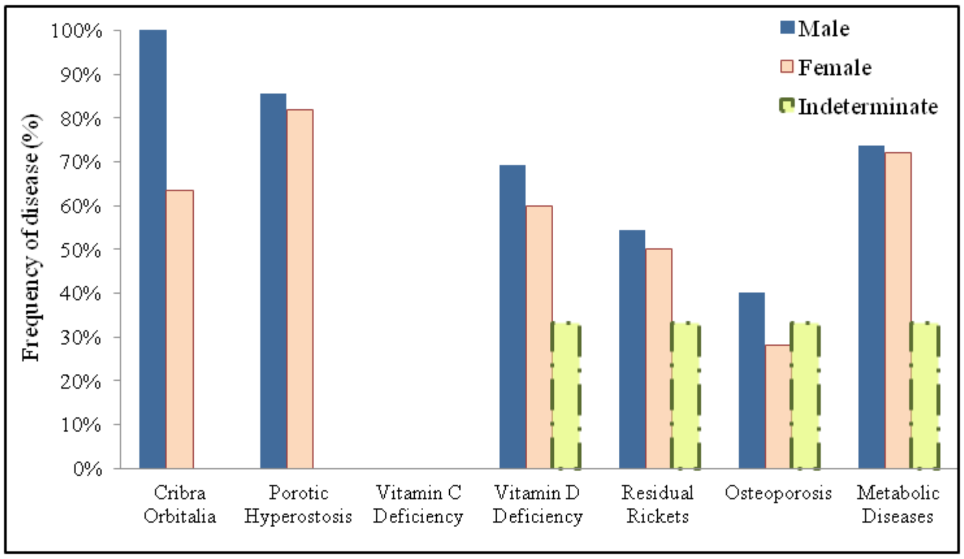

Fig. 5. Hissar II: Metabolic disease profile by sex

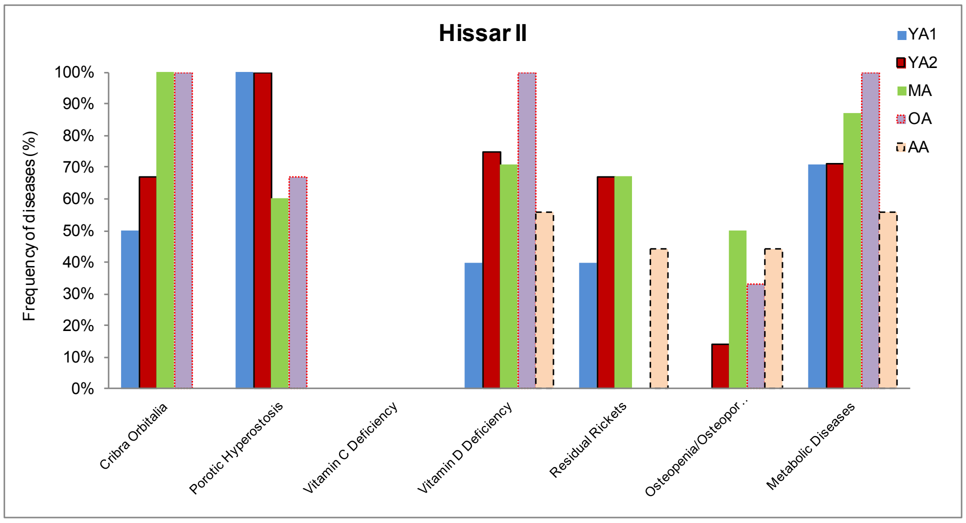

Fig. 6. Hissar II: Metabolic disease profile by age-category

Table. 3. Hissar III: Metabolic disease profile by sex

Disease	Male	Female	Indeterminate	Total	Comparison	
					X^2	P
Cribra Orbitalia	35/71 (49%)	31/49 (63%)	-	66/120 (55%)	2.286	0.141
Porotic Hyperostosis	65/71 (91.5%)	41/49 (84%)	-	106/120 (88%)	1.745	0.249
Vitamin C Deficiency	1/126 (1%)	1/113 (1%)	0/12 (0%)	2/251 (1%)	0.108	0.948
Total Vitamin D Deficiency	72/106 (68%)	67/99 (68%)	8/12 (67%)	147/217 (68%)	0.008	0.996
Residual Rickets/ Osteomalacia	39/86 (45%)	29/88 (33%)	5/12 (42%)	73/186 (39%)	2.834	0.242
Osteopenia/Osteoporosis	49/126 (39%)	54/113 (48%)	4/12 (33%)	107/251 (43%)	2.374	0.305
Total Metabolic Diseases	90/126 (71%)	87/113 (77%)	8/12 (67%)	185/251 (74%)	1.273	0.529

P≤0.05

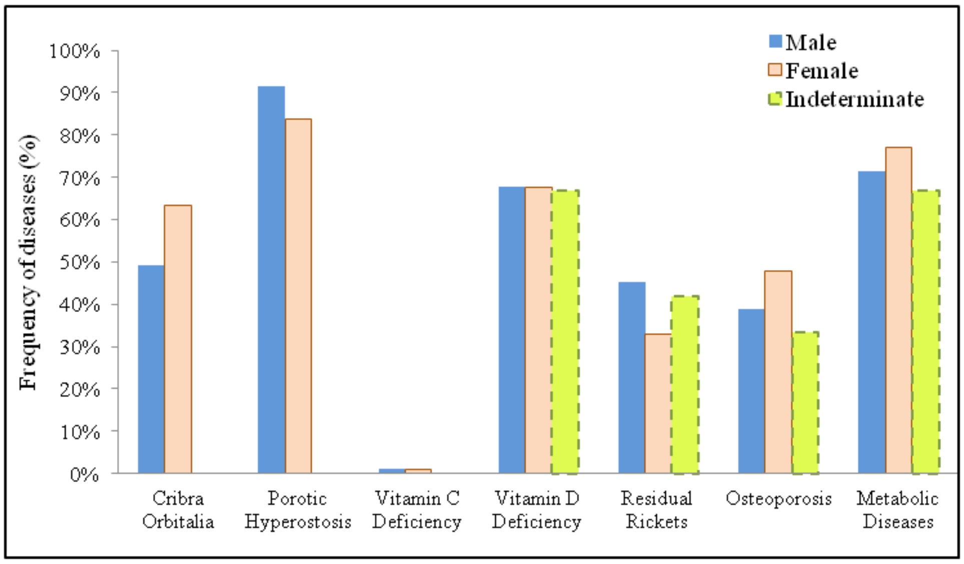

Fig. 7. Hissar III: Metabolic disease profile by sex

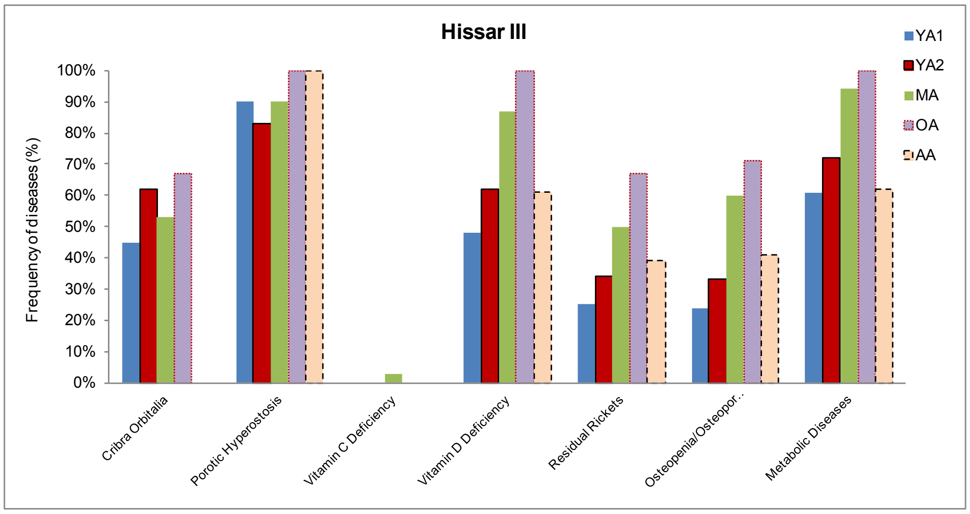

Fig. 8. Hissar III: Metabolic disease profile by age-category

The prevalence of residual rickets/ osteomalacia was 12% higher in males compared to females (insignificant). The females suffered more osteopenia/ osteoporosis (48%) than males (39%) (insignificant). The statistical analysis showed a minor/insignificant difference between males and females from Hissar III, indicating both sexes suffered similar rate of metabolic disorders and nutritional stress.

The prevalence of metabolic diseases between the age-categories is summarised in Fig. 8. It illustrates that the frequency of metabolic diseases increased with increasing age, and that YA1 had the lowest rate (61%) seen in the other age-categories. The prevalence increased gradually in YA2 (72%), MA (94%), and OA. The differences in prevalence noted for metabolic diseases between the different age-categories were significant. The highest prevalence rate for vitamin D deficiency was seen among OA, but there was a decrease to 87% and 62% among MA and YA2, respectively. The frequency of the vitamin D deficiency was 14% lower among YA1 than YA2 (significant). The frequency of residual rickets/ osteomalcia increased with increasing age, with the lowest % rate recorded in YA1 (25%) and the highest in the OA (67%) (insignificant). The prevalence rate for osteopenia/ osteoporosis increased with age in this period (significant). The YA1 showed 24% prevalence when compared to 33%, 60%, and 71% in YA2, MA, and OA, respectively.

Comparison of metabolic disease profiles of the Tepe Hissar population- Between periods: Table 4 and Fig. 9 provide a summary of comparative analysis of the prevalence rate of metabolic diseases between the periods at Tepe Hissar pooled by sex and age. Adult health declined over time, and overall metabolic disease increased. The prevalence rate of metabolic diseases was the lowest among individuals from Hissar I, but increased from 47% to 70% in Hissar II and to 74% in Hissar III. However, these differences were statistically insignificant and minor, indicating that people from three periods were almost equally affected.

The overall level of vitamin D deficiency increased from 47% in Hissar I individuals to 61% in those from Hissar II, and this rate increased to 68% among individuals from Hissar III (insignificant).

Table. 4. Metabolic disease profile for Tepe Hissar population, by Period

Disease	Hissar I	Hissar II	Hissar III	Comparison	
				X2	P
Cribra Orbitalia	2/6 (33%)	14/18 (78%)	66/120 (55%)	4.736	0.094
Porotic Hyperostosis	4/6 (67%)	15/18 (83%)	106/120 (88%)	2.559	0.278
Vitamin C Deficiency	0/19 (0%)	0/43 (0%)	2/251 (1%)	0.529	0.912
Total Vitamin D Deficiency	7/15 (47%)	22/36 (61%)	147/217 (68%)	3.306	0.347
Residual Rickets/ Osteomalacia	6/14 (43%)	16/32 (50%)	73/186 (39%)	1.46	0.692
Osteopebia/ Osteporosis	5/19 (26%)	14/43 (32%)	107/251 (43%)	3.335	0.343
Total Metabolic Diseases	9/19 (47%)	30/43 (70%)	185/251 (74%)	6.128	0.106

P≤0.05

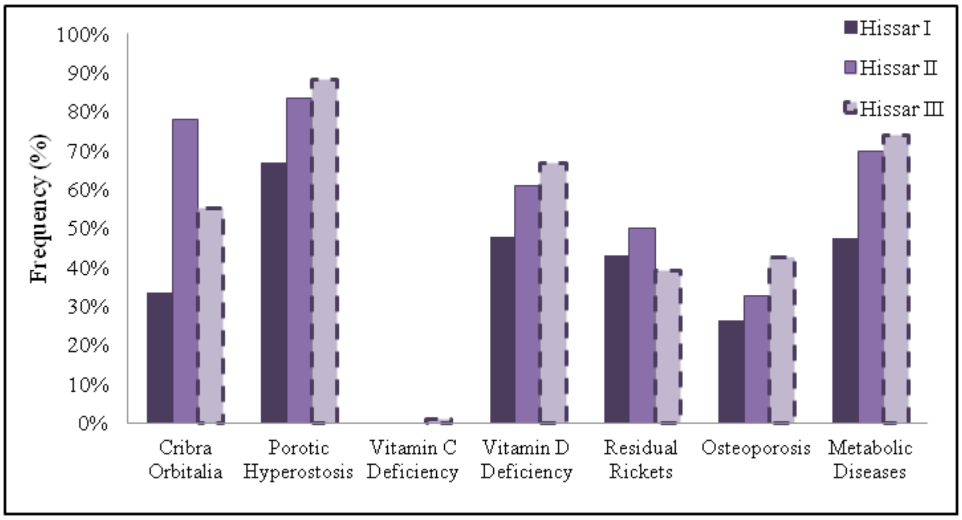

Fig. 9. Metabolic disease profiles for the Tepe Hissar population, by period

Discussion and conclusion

The study of stress indicators and pathological conditions in archaeological populations is an important tool to understand the different aspects of life in ancient human societies (Brickley and Ives 2008: 7). However, these studies are likely to be biased by poor/ambiguous definition of pathological changes, problems with the interpretation of these lesions, limited range of recording techniques, poor preservation and missing bones/teeth, and sampling bias (Wood *et al*. 1992). On the other hand, skeletal remains are inherently biased and tend to record only a limited subset of the stresses to which the body is exposed, for example chronic conditions which persist long enough in the body to lead to changes on the bones or teeth (Cohen 1989: 120; Wood *et al*. 1992). Thus, the lack of pathological changes in a skeleton does not mean that the person has not experienced disease during life, but he/she may have been afflicted by the same disease or deprivation but, because of low resistance, died before any skeletal response could develop (Pinhasi and Bourbou 2008; Wood *et al*. 1992). Therefore, it is impossible to distinguish the difference between healthy individuals with no skeletal lesions, and those who died before disease left any marks on the skeleton. Moreover, genetic variations (e.g. susceptibility and resistance to infectious disease), for example, among humans may affect the frequency of disease from group to group (Pinhasi and Bourbou 2008). Nevertheless, these biases may influence reconstructing population prevalence of pathological conditions from skeletal lesion frequencies (Wood *et al*. 1992), and consequently effect on the reliability of interpretation of health and disease in that population.

The results of this research showed people in each period experienced different episodes of illness and stress. A comparison between the three periods showed a slight decline in health and a slight increase in metabolic bone disease over time. Since these changes were minor, they could not support the hypothesis that socio-cultural economic changes taking place at this site, particularly in Hissar II and III, had a detrimental effect on health and nutritional status of people from these periods.

The presence of cribra orbitalia and porotic hyperostosis in each period at Tepe Hissar suggests the attempts of individuals to "adapt" to adverse environmental conditions and stressors, affecting both sexes equally from different age-groups. This lesion is indicative of iron deficiency anemia as a result of nutritional stress, chronic blood loss, infection or exposure to pathogen, with infection and pathogen exposure (e.g. fungi, viruses, bacteria, and parasites) more often involved than nutrition (Stuart-Macadam 1992).

The Vitamin C deficiency data showed that none of the individuals from Hissar I and Hissar II were affected by scurvy, but that there were two individuals from Hissar III (i.e. only 1% of the study population) who had possible bone changes of this deficiency. This suggests that the population of Hissar always had access to food resources that are rich in Vitamin C (e.g. fresh vegetables, fruits, milk, meat and fish: Ortner 2003: 384; WHO 1999) in terms of their nutritional requirements or appropriate food preparation techniques to preserve Vitamin C in foods. However, limitations such as poor preservation, ambiguous/ limited definition of osseous manifestations of scurvy in adults, and a limited range of diagnostic criteria are factors to consider. Short periods of Vitamin C deficiency also only affect soft tissues; by introducing Vitamin C into the diet most physical signs can be resolved within one or two weeks (Pimentel 2003). These biases, likely limited effective diagnosis in this study underestimating the prevalence rate of scurvy.

An insignificant increase in prevalence rates for total Vitamin D deficiency (residual rickets/osteomalacia, and osteopenia/osteoporosis) was found through time, from 48% in Hissar I, through 61% in Hissar II to 67% in Hissar III. The prevalence rates indicate that almost half of the individuals from each period had less "access" to vitamin D resources (e.g. sunlight) and calcium for a prolonged time during childhood growth or even in adulthood (osteomalacia) (Holick 2006; Kitanaka and Kato 2000), and this rate was similar for both sexes in each period. On the other hand, clothing, living and working conditions also could have put some individuals at risk for developing vitamin D deficiency. For instance, some individuals may have spent most of the day working inside industrial workshops (metallurgy, making pottery) particularly in Hissar II and III, and were not exposed to sunlight.

Nevertheless, the occurrence of Vitamin D deficiency could have been due to exposure to toxic elements such as "lead" and "arsenic" which were widely utilized in metallurgical activities (particularly in Hissar II and III - Tosi 1989). Thornton (2009) found that lead and arsenic were present in metal objects rising from <1Wt% Pb and <3Wt% As in Hissar I to >5Wt% Pb and <6.1Wt% As in Hissar II and peaking in Hissar IIIB-C at 16.1Wt% Pb, 1-5Wt% As. These elements are released during the smelting process or in metal cores or in coal-burning, and produce "stack dust" and "flue gas", contaminating the environment (Roy and Saha 2002; WHO 2010). Lead exposure can also occur through food (e.g. dairy products, meat, fish, grain, and cereal), air, water, and soil contamination, and the concentration of lead found can be between 7.6x105 µg/ m3 in urban areas (e.g. residential areas near workshops) to more than 10µg/ m3 in the vicinity of industrial sources and near smelters (Dart *et al*. 2004: 1423; WHO 2010).

These toxic elements may have increased environmental pollution and affected both workshop and habitation areas at

Tepe Hissar, thus affecting people's health. The distribution of metallurgical artifacts at this site suggests that metal production was wide spread activity among occupied areas of the site (Pigott 1989: 32). Lead interferes with vitamin D synthesis and calcium function and interrupts bone and tooth maturation. It also inhibits haemoglobin production and may cause anemia (Dart *et al*. 2004: 1426-8; Stellman 1998: 23.21; Yu *et al*. 2011). Arsenic affects overall general health and causes bone marrow suppression, bleeding, anemia, leukopenia, protein wasting, and cancer (Abernathy 2001; Fowler *et a*l. 2007: 389; Stellman 1998: 23.05). So lead and arsenic may also have contributed to the occurrence of vitamin D deficiency, cribra orbitalia and porotic hyperostosis at Tepe Hissar. Ostrander (2013) analysed the Coach Lane assemblage (1711-1857 AD.) of North Shields, UK. He found correlations between lead poisoning and rickets, and scurvy; but no direct correlation was found between lead pollution and cribra orbitalia (see Millard *et al*. 2014) and reductions in average stature. Analysis of bone, tooth enamel, and tissue also are used in bioarchaeological research to determine lead pollution/ poisoning in ancient populations (see Budd *et al*. 1998, 2004).

Osteoporosis is a condition correlated with increasing age, however, severe and prolonged inadequate dietary calcium and vitamin D, insufficient sunlight exposure, inefficiency in intestinal mineral absorption, intestinal parasites, parathyroid hormone secretion, and genetics are also accepted as aetiological factors. A diet low in protein, overall malnutrition as well as under-nutrition are also important determinant of peak bone mass and therefore of the risk of osteoporosis (Marcus and Bouxsein 2013; Rauch and Glorieux 2004; Resnick and Niwayama 1995; WHO 2003). Nevertheless, the results of current isotopic analysis (C/N) show sufficiently nutritious diets for each period at this site (Afshar 2015). The occurrence of osteopenia/ osteoporosis among young adults could be an indicator of prolonged nutritional stress and Vitamin D deficiency during childhood or early adulthood, or it could be an indicator of factors which inhibit Vitamin D absorption (see above). However, lead and arsenic pollution may also have played a part. The data suggest that many young adults (except YA2 from Hissar I and YA1 from Hissar II) experienced severe ill-health and malnutrition during childhood or early adulthood, or they may have worked in workshops (e.g. metal smelting) with “less” exposure to daylight and more contact with pollution.

Overall, the archaeological data indicate that parallel to significant cultural shifts and influxes of new people at Tepe Hissar, there was development in technology, an increase in industrial activities, craft specialisation, and accompanying manufacturing workshops, and increasing social complexity (Schmidt 1937; Tosi 1989; Tosi and Bulgarelli 1989), which would have been accompanied by many individuals working in those industries with a division of labor, particularly during Hissar II and Hissar III (4th and 3rd millennium BC). This study has found no significant change in metabolic bone disease (Vitamin C, D deficiency and osteopenia or osteoporosis) in Hissar II and III, compared to Hissar I. The data indicate that the changes that occurred at Tepe Hissar, particularly during the 4th to the early 2nd millennium BC (Hissar II and III), did not significantly impact the health and nutritional status of this population; people from three periods experienced similar frequencies of stress and disease.

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