Palaeodietary Study of Iron Age Population of Gohar Tepe

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Human and animal skeletal remains can provide information about the food chain of inhabitant's nutrition of archaeological sites. The food chain which at it minerals and organic materials transferred from soil to the plants, from plants to the animals, and from herbivores animals to the carnivores, is the benchmark system in ancient diet. Although Chemical interactions between soils, animal and human remains should not be disregarded, chemical data can provide useful information about the paleodiet. In present paper, the concentration of Chemical elements was analyzed in eight premolar teeth of mature individuals as well as a teenager without discrimination of their sex, from discovered graves of Iron Age of Gohar Tepe. The site had been populated until the Iron Age. In addition, we have investigated the ratios of Sr, Ba, Ca, P, with the purpose of studying the food chain from animal to human by analyzing discovered animal teeth in the site. To control diagenetic processes, the calcium/phosphorus (Ca/P) index was used. Furthermore the amount of chemical element in teeth and peripheral soil was compared. Concentration level of Chemical elements was measured by atomic absorption spectroscopy (AAS). The Strontium content and the Ba/Sr ratios show that the human population in Gohar Tepe used products of animal origin somewhat more than plants and terrestrial food more than seafood in their diet.

Keywords: *Paleodiet; Tooth; AAS; Gohar Tepe; Chemical Elements*

Introduction

Recent attention to "palaeodiet" making use of the concentration of trace elements started in the middle of the 20th century (Lambert *et al.* 1979; Zapata *et al*. 2006; Sponheimer *et al.* 2005). Elements most frequently used to reconstruct past diets include Sr, Zn, Ba, and Ca which provide dietary information (Sillen and Kavanagh 1982; Burton et al. 1999).

Barium and Strontium do not have an essential role in the metabolism processes of human body and their accumulation in bone and teeth depend on their consumption therefore these two elements have been applied continual in the reconstruction past human diet (Szostek *et al.* 2009; Ezzo *et al.* 1995). On the other hand, Barium and Strontium have not known physiological function and are therefore classified as non-essential trace elements. This makes them particularly useful for the kind of analysis presented here. Strontium is the most important chemical element in dietary reconstruction (Klepinger 1984; Sandford 1992).

The concentration of Sr and the Sr/Ca ratio decrease when we go forward in the food chain from soil to the plants, from plants to animals, and from herbivore animals to carnivores. In omnivore animals there are approximately between herbivores and carnivores (Burton *et al*. 1999; Sandford 1993; Szostek *et al.* 2003). Because of an important role of trace elements in reconstruct past diets, study of the concentration of chemical elements in skeletal remains in archaeological sites has been carried out not only by anthropologists but also by chemical archaeologists (Sandford 1992; Szostek *et al.* 2003). The chemical composition of skeleton remains, especially teeth which are its most stable part, permits us to reconstruct the diet using concentration of chemical elements in teeth. No much information is available about the economic, social and biological situation of the remained skeletons without written resources and documents. Investigation of chemical composition of teeth for palaeodiet has been performed by several scholars (Arnay-Dela-Rosa *et al.* 2009).

Diagenesis, the postmortem alterations in the chemical constitution of bone, should be noteworthy. Alteration of buried bone through natural diagenesis can vitiate any analysis of ancient

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diet based on concentrations of inorganic elements (Lambert *et al.* 1985). Although chemical reactions between soil and skeletons should not be ignored, the chemical data can be beneficial information about the palaeodie.

Gohar Tepe is a an important site located in eastern parts of Mazandaran province between the cities of Neka and Behshahr (36º 40'42"N 53º24' 07"E) in north of Iran and close to Caspian Sea (fig. 1). This site, which was excavated by Ali

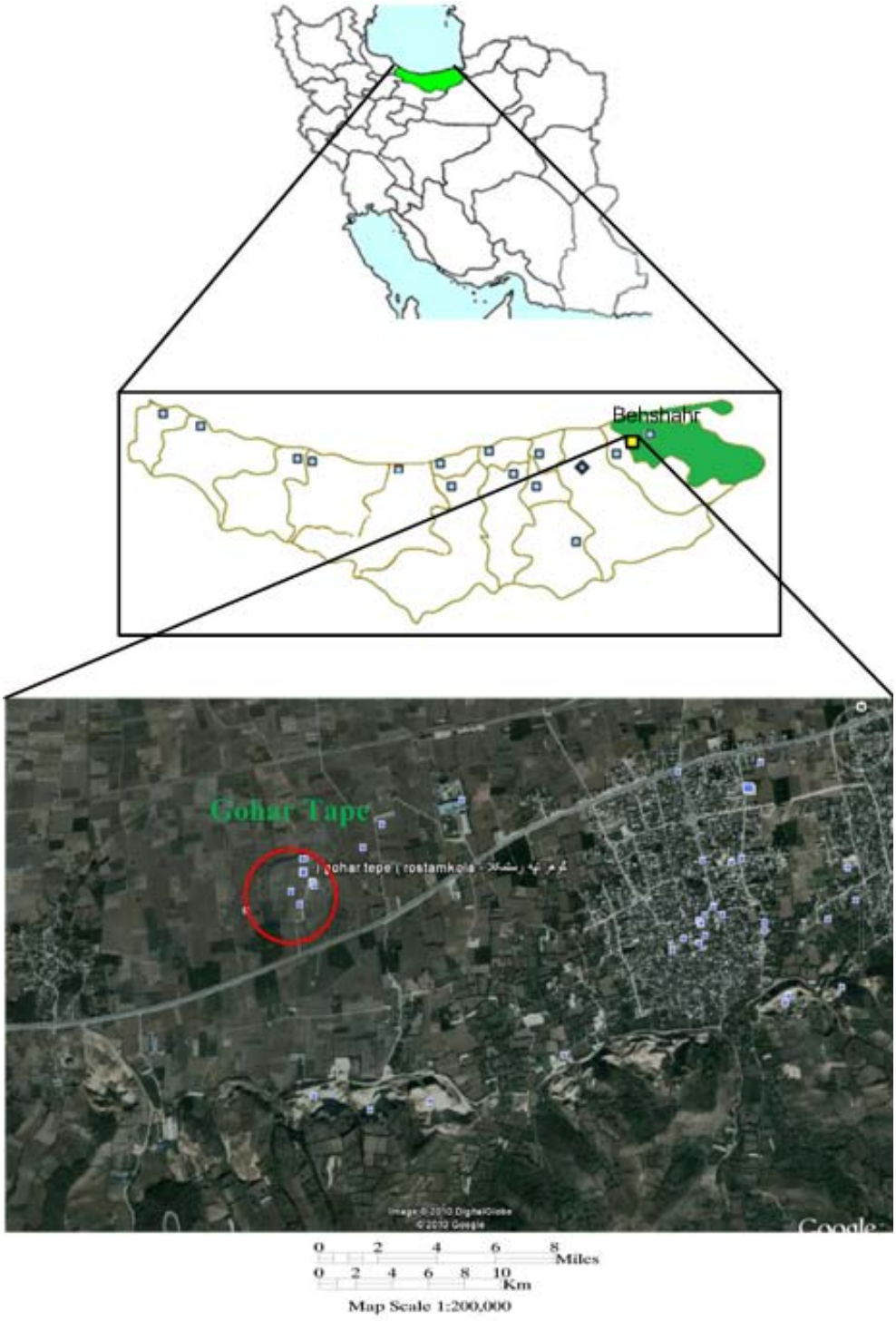

Fig.1:*Gohar Tepe, the site from which samples were obtained.*

Mahforoozi, is a key site in this part of the country. During the Middle Bronze Age (2500 – 2000 BC), the settlement covered more than 10 ha, but was later abandoned in the Late Bronze Age (2000 _ 1500 BC) and was used again between Iron Age II – III periods (1200 – 600 BC) as a cemetery (Soltysiak and Mahfroozi, 2007). 29 human skeletons were discovered from this site. A few skeletal samples has been studies Soltysiak and Mahforouzi (2007). Due to spread of dental caries, at least one tooth of each individual in this population had carious lesion (among 65% of adults). The inhabitants of this settlement had access to rich resources including marine resources as well as fertile agricultural and forest resources. Also, the location has possibly been greatly prosperous during the early and middle Bronze Age since cultural materials have been found in the site.

People in this site kept and used domesticated animals for their nutrition. The animal remains found in the site include boar, sheep, cattle and goat; besides these animals, perhaps wild animals had been hunting. It shows that domesticated animals had been used and possibly in some months of the year people carried out agricultural activities.

The aim of present study is to show the concentration changes in the levels of chemical elements accumulation in teeth in order to have a better understanding of dietary of the inhabitants. The chemical elements whose concentration may provide information about the diet are Sr, Zn, Ca, Ba, and P.

Sampling and Experimentation

A number of human skeletons, which are dated back to Late Bronze Age and Iron Age II – III, were sampled randomly. Human teeth were also sampled from different age and sex. Investigation of trace elements concentration in tooth was carried out on 9 samples of the second premolar of human teeth and 9 samples of the premolar and molar of animal teeth. Some of the animal remains were directly related to the burials as grave goods and some of them have been found from the stratified layer. The teeth show no sign of fossilization. From the graves under study four soil samples were collected and transferred to laboratory after being put in the polythene bags (fig.2).

Fig.2: *Human tooth sample and respective skeleton from Gohar Tepe (Iran).*

Each tooth without root (only enamel and dentine), was brushed and washed several times under tap water and finally washed in distilled water. Samples were further cleaned in an ultrasonic bath as described by Szostek and cooperators (Szostek *et al.* 2003). They were then dried at the temperature of 80°C in an oven and weighed with accuracy of 0.00001 g. Afterwards, the samples were immersed in a 4:1 mixture of 65% nitric acid (Merck) and perchloric acid (70% Suprapur, Merck). In order to digest them, solutions were transferred to volumetric flasks and diluted to 10 mL with ultrapure water. A part of this solution chosen for determination of calcium in tooth was further diluted to 1:1000. Zinc solution was also diluted to 1:100. Samples were finally submitted to Department of medicine for detection of following elements: Ca, Ba, Sr and Zn.

In order to analyze all the elements except for phosphorous, the obtained solution was submitted to Atomic Absorption Spectrophotometer (AAS) laboratory of Medical Faculty of Tarbiat Modares University, and analyses were performed using model AAS.670 made by Shimadzo in Japan. They were subsequently submitted to ICP laboratory in faculty of basic sciences of Tarbiat Modares for phosphorous analysis. The soil of the grave was also brought to AAS lab when being prepared by digestion and finally statistical analysis was carried out using SPSS software version 16.

Results and Discussion

Chemical composition was derived from Iron Age skeletons in Gohar Tepe reported from several graves uncovered in Gohar Tepe. For this purpose, As stated above, a number of human teeth, herbivore and omnivore animal teeth disregarding their age and sex, were sampled randomly. The concentration of trace elements in human teeth and the graves identification numbers (ID) illustrated in Table. 1

The values of Ca/P ratio were 2.17 in human teeth samples (enamel and dentine), 2.21 in herbivore animal teeth and 2.25 in omnivore animal teeth (fig. 3). In fresh bones of modern populations, the Ca/P ratio ranges from 1.8 to 2.19. This result documented by several researchers (Szostek *et al.* 2009; Sillen 1989). Our results indicate that the samples under study approximately have not been diagenetically altered which may demonstrate that the condition of the preserved material was good; but this is not sufficient index and other factor such as PH of the soil and concentration of elements in the soil of grave should be investigated.

When diagenesis occurs, concentration of elements in the soil and bone would tend to become increasingly similar to each other. Results of the research by Pate and Hutton, (Pate and Hutton 1988) reveal ion exchange between some parts of skeleton

Graves Number	Elements						
	Ca (%)	P (%)	Ba (ppm)	Sr (ppm)	Zn (ppm)	Ca/P	Sr/Ca
AI2XX (F32)	46.39	17.65	8844	295	276	2.63	6.36
AL2XXI (F6)	37.44	12.99	6623	105	220	2.88	2.8
AH2XXII (F8)	42.25	13.84	7564	137	245	3.05	3.24
Ah2xxI F37	30.86	14.22	5320	117	155.8	2.17	3.79
Ah2xxI F6	26.58	15.06	4587	133	206	1.76	5
Al2xx (F32)	25.16	13.92	5253	108	242	1.8	4.29
AJ2xx (C35)	21.55	13.01	5470	208	226	1.66	9.65
Ah2xx (F33)	26.33	13.73	6116	144	227	1.92	5.47
AJ2xx (C 53)	27.88	16.31	5586	84.4	248	1.71	3.03
Average	31.60444	14.52556	6151.444	147.9333	227.3111	2.175556	4.847778
SD	8.48	1.55	1332.23	65.21	33.44	0.539	2.15

Table.1: *Concentration of trace elements in human teeth from the Gohar Tepe.*

with soil. The movement of ions between bones and soil from areas with higher concentrations to those with lower concentrations shows that diagenesis, i.e. postmortem alterations, has occurred (Szostek *et al.* 2009). .In some cases dentine is not completely resistant to diagenesis process (Kohn et al 1999; Budd et al 2000). In present study, mean concentrations of chemical and trace elements in teeth found in graves and animal teeth and also soil have been provided in figs. 4 - 6. Bone zinc (Zn), have been considered as indicator of high-protein diets (Velasko *et al.* 1997). In fact, concentration of Zn is provided by milk and dairy products. The content of Zn between the soil of graves and teeth was similar (fig. 4). Zn concentration was approximately 227 ppm in human teeth; it was about 182 ppm in herbivore animal and about 117 in omnivore animal. The concentration of Zn in the soil was measured to about 260 ppm. Because of high density of Zn in the soil, which was sampled from the graves, and also low PH, which will be described in the following discussion, perhaps the Zn concentration of dentine was altered by postmortem changes.

Barium (Ba) is useful element to separate marine from terrestrial mammal bone. Its level is high for terrestrial mammal and low for marine species (Klepinger 1984). Furthermore, the ratio of Ba/ Sr can provide information about food web .Ba/ Sr ratios are considerably lower in sea water and marine organisms than in fresh water and terrestrial organisms, (Szostek *et al.* 2009; Burton and Price 1990).

The value of Ba content is high either in animal teeth and soil as depicted in fig. 5, but this value in human teeth is much higher than others samples. So, there is a risk that the animal samples under study have been diagenetically altered, but the human teeth samples have not been affected by diagenesis process. It is notable to state that the Ba ratio is much higher than Sr ratio in human teeth.

The PH of soil was ca. 5.5, because of low PH of soil graves, it is suggested that the high barium content in human teeth may be due to its intensive accumulation throughout an individual's life; it has been suggested that the concentration of barium (Ba) is higher in acidic water than alkaline water, (Bergfield 2007).

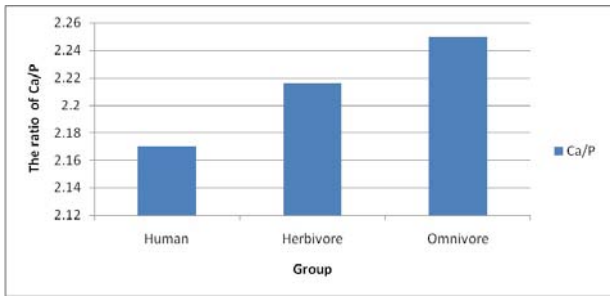

Fig.3:*The ratio of Ca/P in the analyzed groups.*

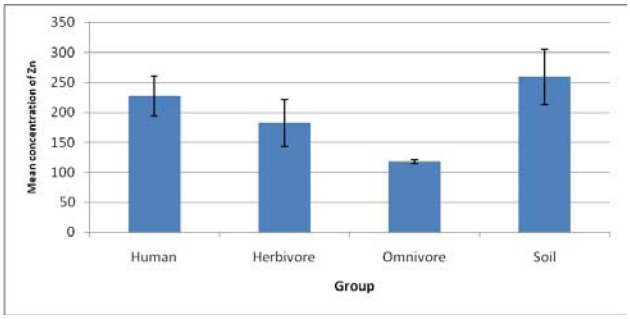

Fig.4:*Mean concentrations of Zn (ppm) in the analyzed groups.*

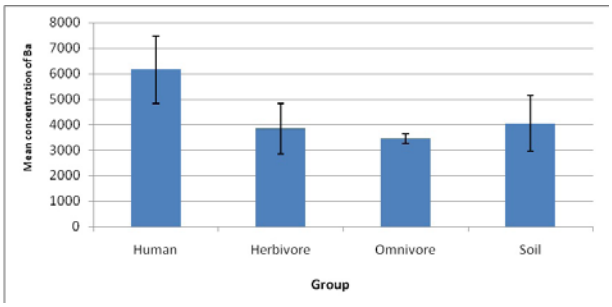

Fig.5:*Mean concentrations of Ba (ppm) in the analyzed groups.*

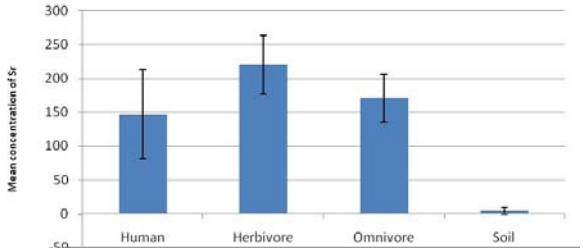

Fig.6:*Mean concentrations of Sr (ppm) in the analyzed groups.*

Sr and Zn are elements emphasized in nutrition because of their least sensitive to diagenesis, (lambert *et al*. 1985; Radosevich 1993). Concentration of Sr is different within human populations, (Sillen and Kavanagh, 1982), it is different among the different ages, as Strontium levels in human bone increase slightly with age, (Price 1989).

Ratio of Sr/Ca in human samples is approximately 60% of what is in herbivorous mammal samples (Schoeninger 1982). The Sr/Ca ratio technique has been shown to effectively distinguish between the diet of faunal species and plants, (Sillen 1981). LSD (Least Significant Difference) test reveals significant differences in Sr and Sr/Ca between human teeth and herbivore animal teeth ($P < 0.05$) but it also shows similarity between concentrations of Sr ($P > 0.05$) between omnivore animals and human (figs.6-7).

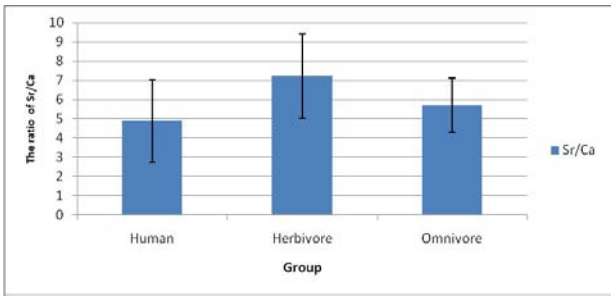

Fig.7:*The ratio of Sr/Ca in the analyzed groups.*

Conclusion

The results show that, there is no significant difference in the levels of Sr in teeth between human and omnivore animal, while there is significant difference in the levels of this element between human and herbivore animal, so we can conclude that the diet of individuals in present case study was based on animal products and also amounts of plants.

The values of Ca/P ratio show that the teeth samples under study have not been diagenetically changed, this may indicate that the condition of the preserved material was good, but because of similarity between the content of Zn in the soil and teeth and also low PH of soil, there is little risk that Zn has been diagenetically changed.

Generally, the high density of trace elements in human teeth samples demonstrates that the peoples of Iron Age population of Gohar Tepe have had access to resources of animal and plant foods.

Despite this factor that the site is close to the Caspian Sea, the values of the Ba and Ba/Sr ratio in human teeth show that the human population in Gohar Tepe used products of terrestrial foods more than marine foods.

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