Review Article

A review on heavy metal profiling from teeth for forensic purposes

Aiman Rito Chophi[1*], Rajinder Singh Chandel[2]

[1]*Research Fellow,*

[2]*Associate Professor,*

Department of Forensic Science,
Punjabi University, Patiala, Punjab- 147002, India.

* Corresponding author: ritochophi@gmail.com

ABSTRACT

Heavy metal profile from teeth are utilized in various discipline of sciences such as medical, environment, geology, and anthropology to indicate individuals health status, risk of exposure to harmful environments, and to estimate age and region of certain artifacts or human skeletal remains. The elements profile from teeth are also studied in the field of forensic science, in cases where skeletal remains are found, with the objective to provide information about the origin of species, age, gender, and provenance, and to ultimately narrow down the remainsto the level of individualization, if possible. However, there are number of variables which affect the distribution of heavy metals in teeth- for instance: types of teeth (e.g. incisor and canine), region or part of teeth (e.g. crown and root), and different method of analysis employed. And due to this influence of variables, it becomes difficult to compare and interpret results. Thus this review seeks to find a direction (though there are no particular way), at least in terms of similar or contradictory reported results, and to find which variables (heavy metals) contributes more to the variance in a particular dataset, or which heavy metals are more significant for estimating various parameters that an analyst wishes to observe. The present article also highlights the application of heavy metal profiling in forensic cases as and when the exhibits are received in forensic laboratories.

Key Words: forensic sciences; heavy metals; individualization; discrimination; provenance.

INTRODUCTION:

Heavy metals are naturally present in the environment and they are also added to the environment through human anthropogenic activities [1]. Some heavy metals are beneficial for human health while others have adverse effect, and therefore, the concentration of these elements in the body have been studied to a great extent. Gdula-Argasinskaet al.[2] demonstrated in black vole (C. glareolus)that, change of heavy metals in the environment leads to change in the concentration levels present in the teeth. This can reflect the animal's diet and also the site (polluted or non-polluted) [3]. The level of concentration may indicate individual's health status [4, 5], nutrition [6], and risk of exposure to harmful elements present in a certain region or locality [7-9]. In this regard, body tissues and body fluids are usually utilized to indicate the short term exposure and concentration levels of heavy metals [1]. Hard tissues such as bones and teeth accumulate most heavy metals absorbed in the body and offers valuable information with respect to long term exposure to heavy metals. Of the two hard tissues, heavy metal profiling in teeth is reported to be of preferred choice as teeth are not subjected to turnover,easy sampling [10], and teeth's ability to resist degradation and withstand harsh and extreme conditions of heat; even well preserved for hundreds and thousands of years [11]. The elemental profile from teeth is thus being studied in various discipline such a medicine, environmental studies, public health, anthropology, and in the field of archeological research. Elemental profiling in teeth has also drawn considerable attention of the forensic scientists, who seeks to answer questions pertaining to identity, provenance (geographical origin), age, and sex of an individual when the body or buried body remains are recovered after the tissues have been highly degraded. This could arise from concealed bodies, mass disaster cases such as natural calamities, accidents and war, arson, and in cases where the person was attacked using acids. Sometimes, the criminal may also deliberately mutilate the body after committing a crime to hamper the criminal investigation process [12, 13]. Thus the distribution of heavy metals in teeth, and along with the standard data of background environment (diet, soil, water etc.) and dental records (restorations and fillings) can reveal important information, and serve as associative evidence in criminal investigations [12]. Heavy metal profiling also offers the advantage over molecular genetic techniques such as DNA by providing the age of an individual and which could be helpful in certain cases like missing person cases. A number of studies have reported various elements such as Pb, Fe, Ca, Mn, Cd, Cr, Cu, Zn, K, Na, Mg, Al, Ba, Sr, U, Ni, Co, Sn, Sb, etc. from various parts or region of teeth (enamel, dentin, and roots). These elements have mostly been analyzed using atomic absorption spectroscopy (AAS), Inductive Coupled Plasma-Mass spectrometer (ICP-MS), and X-ray fluorescence. The detail analyses of teeth using these techniques, and the advantages and limitations of each technique have been reviewed by Singh et al. [13]. Recently, isotopic ratios of elements such as Sr, Nd, Ca, and radio-nucleotides such as Ra, and Th have also been analyzed from teeth [14,15]. Heavy metals are deposited on teeth during the formation of mineralization process, and isotopic signature of dental enamel is indicative of

the environment and the diet consumed during enamel formation. Dentin, which is a composite material containing inorganic collagen matrix, is reported to be richer in organic content and biologically more active than enamel [1].

Forensic significance of heavy metal profiling

Osseous versus non-osseous versus dental in origin: Often, a forensic anthropologists maybe required to establish whether the exhibit is of osseous or dental in origin, particularly when burned or charred fragments of materials are submitted for examination. In such a scenario, heavy metal profiling could be of immense assistance as demonstrated by Christensen et al. [16]. The authors employed X-ray fluorescence (XRF) technique for the analysis of various materials: bone, teeth and other materials such as wood, glass, plastic, metal, and minerals and subjected these materials to chemicals, burning and weathering conditions. Based on the concentration ratio of Ca and P, the materials were classified as osseous or dental in origin. Unaltered teeth had dental Ca/P mean concentration around 4.02 (s.d=0.83), while osseous tissue had mean concentration around 4.92 (s.d=1.19). Likewise other subjected materials were classified based on Ca/P ratios. Minerals and wood also showed similar Ca level but P was reported to be absent. Minerals such as synthetic hydroxyapatite (implants for human body) and rock apatite were found to have similar composition to that of teeth [17]. However, the authors reported that minerals can be identified through their crystalline structure. Principal component analysis (PCA) analysis showed distinct clustering among the osseous and non-osseous materials and osseous and dental tissues were differentiated with 94% accuracy using quadratic discriminant analysis(QDA) from non-osseous and other chemical materials. Both the tests demonstrated the power of portable XRF over Scanning electron microscope-Energy dispersive X-ray (SEM-EDX)[18] in terms of providing on-site screening tool capability with reasonable cost and non-destructive method of sample analysis.

Origin of species

Establishing the origin of species is an important aspect in forensic science, as the material identified as non-human may be excluded from subsequent analyses; if it is determined not to be of forensic significance [17]. However, non-human teeth are also of valuable evidence in cases of illegal wild-life poaching and trading cases [19]. Nganvongpanit et al. [19] studied 15 species of animals, mostly mammals to investigate the distribution of heavy metals in teeth and tusks of animals using XRF technique. In elephant, 16 elements were found in deciduous teeth and 18 elements were found in permanent teeth. The elements Si and Mo were reported to have been found only in tusk. Elements such as (P, Ca, Fe, Rh, and Pb) were found common in all the species. Concentration of other elements varied among different animals and different teeth types. The highest classification accuracy (73%) of animal species was provided by a combination of different element ratios (Ca/P + Ca/Zn + Ca/Fe + Ca/Sr + Zn/Fe). From a forensic standpoint, low classification accuracy was achieved. However, for some animal species such as dog, pig, goat, tapir, monkey, and elephant,

100% classification accuracy was reported. The same authors working on another study [20] reported 83.2% classification accuracy between human versus non-human using step wise discriminant analysis. A good number of samples were collected for the said study: human (n=111), and animals (n=173) representing 20 animal species. The same elemental ratio (Ca/P + Ca/Zn + Ca/Fe + Ca/Sr + Zn/Fe) was utilized. Similarly,Buddhachat et al. [21] utilized XRF technique for differentiating eight different species from different body parts such as antlers, teeth, bone etc. Ten elements-Si, P, Ca, Ti,Mn, Fe, Ag, Cd, Sn and Sb were found in all species while Al, was reported to be present only in human, monkey and elephant species. A classification accuracy of 78.4% was reported in this study using canonical discriminant plots derived from molar teeth across six species. The elemental composition of dugong (*Dugongdugong*)tusks [22] also showed varied compositions which could be differentiated from the elephant and tigers teeth with correct prediction accuracy of 98.2%. The reason for high prediction accuracy was attributed to habitats as dugong lives in sea along the coastal habitat. Elemental profile from these studies suggests that fairly high classification and prediction accuracy can be generated, and thus the information obtained using this non-destructive technique XRF can be used as screening tool and aid in forensic investigations. The limitation is that, the instrument cannot detect light elements (H to He) and isotopes. Besides the analysis of heavy metals in teeth, organic contents in teeth have also been analyzed using Raman spectroscopy [23-25] to investigate the different origin of species.

Age estimation

In certain scenario where human skeletal remains are uncovered in forensic investigations, and there are no clue as to the identity of an individual, the estimation of age and stature contributes to the profiling of the individual. Age estimation can assist in eliminating certain individuals out of a large pool and narrow down the search for the missing person [26]. In the field of forensics, bones have largely been studied for age estimation purpose. It has been reported that age estimation in child children is more accurate than in adults. In adults, only few age-dependent features are suitable for estimation, and other features physiologically degenerate or are affected by endogenous or exogenous factors and can result in errors [27]. Morphological methods typically provide age estimation errors of about of ± 10 years in adults [26]. The estimation of age carried out by racemization method (conversion of L-aspartic acid to its D-form in teeth) also gives estimation errors of about ±5 years. Therefore, search for new techniques which can provide and predict the age to high accuracy are continually being explored. Since, the heavy metals deposited on teeth during the formation and mineralization process are retained and are not subjected to turnover; a considerable amount of work has also been carried out from teeth [28]. A recent technique, accelerator mass spectroscopy was utilized by Alkasset al. [26] for analyzing 95 teeth (mostly premolars and molars) of 84 individuals for age estimation and origin of an individual. With respect to age estimation, birth estimation error was derived from two sets of teeth (n=12, and n=66) by analyzing isotope 14C content and the authors reported age correlation accuracy of 1.9±1.4 years and 1.3±1 year respectively [26].

Nganvongpanit et.al [20] analyzed heavy metals on 444 teeth of 111 skulls and reported that there was a correlation between concentration of heavy metals and age. Elements such as Fe ($R^2 = 0.121$), Zn ($R^2 = 0.148$), Cr ($R^2 = 0.249$), Ni ($R^2 = 0.121$), Sn ($R^2 = 0.246$), Sb ($R^2 = 0.239$), Cd ($R^2 = 0.239$) and Ag ($R^2 = 0.097$), and Ca/P ratio ($R^2 = 0.088$) showed negative correlation with age. Kosa et al. [29] found a significant decrease in the Ca/P ratio in older individuals using electron probe microanalysis. In the mentioned study [20] the type of teeth (e.g. canine) showed different and uneven distribution of heavy metals among the teeth, and parts of teeth (crown and root) showed that there exists statistical difference in elements distribution with roots having high Si, S, Fe and Ca/P ratio. It was mentioned that since the teeth was analyzed using Hand Held XRF tool, enamel could not be distinguished from dentin. For age estimation, it was reported that four representative teeth was taken from each skull but no specified type or parts of teeth has been mentioned. Sehrawat1 & Monika Singh [30] employedwavelength-dispersive X-ray fluorescence spectrophotometer (WD-XRF) analyzer on molar teeth for age estimation. Amongst the various regression model generated for age estimation, the model with variable O and K showed the highest correlation coefficient ($R^2=0.055$, $p<0.05$). Utilizing the following equation regression equation was generated (age = 57.13. 0.316 × O + 105.113 × K) using 100 collected modern teeth, the ancient teeth (excavated human body) was predicted to be of males in the age (19 to 48 years). However, due to low correlation coefficient generated, the results could not be affirmed, and the authors suggested more works are

required with larger number of samples. AL-Qattan et al. [31] analyzed 398 permanent teeth 1stpremolars and third molars of 192 males and 206 males. Heavy metals in dentin varied from (0.1-0.5g) and correlation coefficient of 0.654 was observed with standard error of 5.95 years ($R^2=52.3$). The difference in true age and calculated age was found to vary by 1.3±4.8 years.

Determination of gender

Heavy metals in teeth are also being exploited to see if gender can be determined based on its distribution. The above cited authors [20] also analyzed teeth of 46 males and 62 females and observed that out of elements such as (Al, Cu, Si, Zn,P,Zr, S, Mo, Ca, Ag,Ti, Cd, V, Sn, Cr, Sb,Mn, W, Fe,Pb, Ni, and light elements)six elements: Si, Cr, Mn, Zn, Zr and W showed considerable difference in human gender. Mann-Whitney U-test was performed on the data, and Mn washigher in males ($p < 0.05$), but the proportions of the others were significantly higher in females($p < 0.05$). A stepwise discriminant analysis was performed which provided only 65.5% and 65% accuracy and precision respectively. The authors opined that even though some elements are significantlydifferent between males and females, elemental profile might have littlepotential as an effective tool for sex identification. Sehrawat and Singh[30] reported that Mg, S, and Sr were significantly different between males and females. However, univariate discriminant function analysis resulted in only 57% correct individual gender. Thus, this study supports the statement that heavy metals are not effective for sex determination. Similarly, the referred articles [9, 32-33] found no significant difference for Pb and Cd between the two genders though the

levels were relatively higher in males. AL-Qattan et al.[31] reported that the mean dentin lead concentration was significantly higher in males than in females (6.8± 4.7 mg/g and 5.6 ± 4.6 mg/g respectively; p= 0.015), but no discriminant function was performed in this study. However, more studies should be carried out to find which variable can give high accuracy and precision for sex determination.

Forensic provenance

Heavy metals and particularly isotope ratios of hydrogen (H), carbon (C), nitrogen (N), oxygen(O), sulfur (S), strontium (Sr) and lead (Pb) in teeth act as %natures recorders+. Particularly, Oxygen from water play an important role in the formation of bioapatite (a group of minerals present in bone or tooth enamel), and strong correlation(R^2 = 0.76) between bioapatite and 18O values of water have been found to exist [11].These elements enter the tissue through food and water consumed or may even enter the body through inhalation or dermal contact [1]. The profiling of these elements thus can reflect persons life history including diet, provenance (geographical origin), and travel-movements [34]. The profiling of elements to assign the possible geographical location was utilized in a case in 2002, where an unidentified body was recovered. The combined analysis of stable isotope ratios of light (H, C, and N) and heavy elements (Pb, Sr) on the body tissues indicated that an individual belonged to Romania [35]. Kooter et al.[14] utilized the isotope ratio of 87Sr/86Sr to investigate the spatial distribution of human dental enamel and tap water. While tap water correlated to Sr ratio, no correlation was observed between tap water and human enamel. Tap water is a mixture of groundwater and river bank infiltration, and it contributed to wide range of observed tap water Sr ratios. Thusunless elements present in water are correlated to human teeth,proxy for mobility for modern globalized(Dutch) context was reported to be invalid,and indication of geographical origin becomes difficult. The authors observed that Sr and water hardness (Mg and Ca) were significantly correlated-consequently there were variation due to water hardness. It was reported that in modern times, due to globalization of food supply and globalization of diet intake, forensic provenance from Sr ratios has become difficult, as compared to ancient population where food were of local origin. In this study[14]98% of Dutch inhabitants were found to have 87Sr/86Sr ratios between 0.7088 and 0.7099. The exposure to environment (industrial) can also help in locating geographical origin of an individual. This was demonstrated through the work of [9] to assess the fluoride environmental pollution in Tunisia. Gufsa, which is a place exposed to phosphate quarries had significantly higher fluoride concentration in teeth (mean=6793.1) as opposed to region Tunis which had mean concentration of 1008.8.Kamenovet al.[34] carried out similar work to compare teeth data from Bulgaria and surrounding areas. The authors found that isotope of C,O and Pb contributed to significant difference between differentiating local versus foreigners and commented that these elements appear to be controlled by locally imbibed water. Particularly, Pb isotope showed high precision in locating Bulgarian region, however it was stated that due to changes in the present natural environment, Pb isotopic data available for ancient humans cannot be directly used for describing modern day human teeth data.

The article stressed that compilation of new multi-isotope dataset based on human teeth with known origin is essential. Plomp et al.[36]remarked that multi-isotope profiling is becoming more significant in forensic and archaeological provenance. The authors utilized Neodymium isotope from human dental enamel to examine whether Nd isotope composition in human enamel reflects the geological area in which it was formed. It was observed thatfor 83.3% of the individuals, the variation in Nd isotope composition of human enamel is indistinguishable from the geology of the location. Though, Nd isotope was found to be majorly controlled by geology, other factors such as human anthropogenic activities, and globalization of food market were suspected to be influencing the outcome of the result.

Individualization

Heavy metal profiling cannot identify a person outright. It only serves as an additional tool in tandem with other genetic techniques. However, in certain cases, restorative resin materials which have distinct elemental composition may enhance the identity of the individual. In this aspect, Bush et al.[37] demonstrated that 100% success rate in locating the tooth resin (placed) and 100% success rate in identification of resin brand. Some resin samples could not be analyzed by XRF - in cases where accessibility was an issue (mal-aligned teeth and curvature of arch)technique as this tool must be placed no further than 2mm from the object.In samples subjected to cremation processed cremains, though there was reduced weight and percentage in size, and hindered in recognizing the resin fragments, XRF was successfully used to discriminate each

individual in the study group. The authors opined that when dental record exists, identification of individual can be achieved by recognition of restorative brands. Castro et al. [38] attempted to discriminate 7 individuals from different layers of teeth (enamel and dentin plus cementum layer), and 14 individuals using the whole tooth. PCA plot of elements (Mg, Mn, Fe, Cu, Zn, Rb, Sr, Ba and Pb) of different individuals showed clustering of samples of the same individual in both the cases, but classification accuracy and precision were not reported. The samples analyzed were also quite low in number. Thus the authors believe that chemometric methods such as PCA in tandem with LDA can be utilized on more number of samples for classification accuracy and also for generation of predicting models. Interpretation of results should be carried out in a most cautious manner as there are several factors affecting the elements distribution in teeth. Amr et al.[39] reported that there are significant difference in concentration of metal between primary and permanent teeth. Elements like Na, Mg, Al, Zn, Sr, Pb, Cd, and Ba were reported to be higher in permanent teeth, while elements like Mn, Mo, Ag, and Bi was reported to be lower in permanent teeth. The authors also observed that there was significant difference between Al, Mg, Sr, Cd, Ba and Pb levels in pulp of healthy and carious teeth. Teeth with roots were found to have more concentration of Pb, and Zn than teeth without roots, and concentration of Pb were also found higher in canines than in molars, which were found to have high concentration of Zn and Hg [40]. In addition, smoking and beverages are known to affect the elements distribution profile in teeth. The cited articles [33, 41] reported that

heavy metals such as Zn, Cu, Pb, Ni, Co, and Cd were significantly different between smokers and non-smokers while the effect of beverage was dependent on the ph.

Conclusion

Heavy metals in teeth are continually being studied to aid in identification of a person from skeletal remains or forensic geo-locating of origin. So far, a wide range of elements have been reported, but there are huge variation and factors affecting the distribution of heavy metals, and no specific biomarker to indicate age, gender, or for provenance approximation seems to exist- though isotopes on non-metals such as carbon, oxygen, nitrogen, and that of heavy metals such as Sr and lead are reported to provide good estimation. The best estimation can only made from a number of heavy metals combined provided that the standard data, both individual and environment exists for comparison purposes. Few studies that have utilized multivariate statistics have reported poor correlation model for age estimation, and some studies have reported that the determination of gender using heavy meal profile might have little potential as an effective tool. More studies are required to recognize which elements offer effective estimation. Similarly for forensic provenance, the distributions of metals in teeth are becoming homogenous due to similar food diet intake- a result of globalization in food supply. This issue needs to be addressed. Some authors have suggested that analysis of local dietary plant samples can also be examined in the near future. Thus at the moment, the use of heavy metals in forensic cases at best can only be utilized for screening purposes, and be utilized as associative evidence.

REFERENCES

1. K. Asaduzzaman, M.U. Khandaker, N.A.B. Baharudin, Y.B.M. Amin, M.S. Farook, D.A. Bradley, O. Mahmoud, Heavy metals in human teeth dentine: a bio-indicator of metals exposure and environmental pollution, Chemosphere (2017), doi: 10.1016/j.chemosphere.2017.02.114.

2. J. Gdula-Argasinska, J. Appleton, K. Sawicka-Kapusta, B. Spence, Further investigation of the heavy metal content of the teethof the bank vole as an exposure indicator ofenvironmental pollution in Poland Environmental Pollution 131 (2004) 71-79.

3. J. Appleton, K.M. Lee, K. Sawicka-Kapusta, M. Damek, M. Cooke, The heavy metal content of the teeth of the bank vole(Clethrionomysglareolus) as an exposure marker of environmentalpollution in Poland Environmental Pollution 110 (2000) 441-449.

4. M. Saiki, L.K. Adachi, E.M. Adachi, Elemental comparison in sound and carious human teethby instrumental neutron activation analysis, J.RadioanalNuclChem, 282 (2009)29. 32DOI 10.1007/s10967-009-0262-9.

5. R.V. Rao, P.M. Anupama, D.G. Mahesh, A.W. Iqbal, Y. Ramakrishna, P. Venkateswarulu, Estimation of trace elements in various parts of human teeth using external beam pixe. International, Journal of Physics and Applications, 2 (2010) 123-134.

6. C.J. Brown,S.R.N. Chenery, B. Smith, C. Mason, A. Tomkins, G.J. Roberts, L.Sserunjogi, J.V. Tiberindwa, Environmental

influences on the trace element content of teeth-implications for disease and nutritional status, Archives of Oral Biology 49 (2004) 705· 717.

7.]B. Kamberi, F. Koçani, E.Dragusha, Teeth as indicators of environmental pollution with Lead, Journal of Environmental &Analytical Toxicology, 2 (2012) 2-5, DOI: 10.4172/2161-0525.1000118.

8. K. Grünke, H.J. Stärk, R. Wennrich, U.Franck, Determination of traces of heavy metals (Mn, Cu, Zn, Cd and Pb)in microsamples of teeth material by ETV-ICP-MS. Fresenius J Anal Chem, 354 (1996) 633. 635.

9. A.B. Saida, C. Telmoudi, K. Louati, F. Telmoudi, D. Amira, M. Hsairi, A. Hedhili, Evaluation of the reliability of human teethmatrix used as a biomarker for fluorideenvironmental pollution, AnnalesPharmaceutiquesFrançaises, 78 (2020), 21-33.

10. M.A. Amr, A.F.I. Helal, Analysis of trace elements in teeth by ICP-MS:Implications for caries, Journal of Physical Science, 21 (2010) 1-12.

11. L.A. Chesson, W.Meier Augenstein, G.E. Berg, C.P. Bataille, E.J. Bartelink, M.P. Richards, Basic principles of stable isotope analysis in humanitarian forensic science, In: R.C. Parra, S.C. Zapico, D.H. Ubelaker (Ed), Forensic Science and Humanitarian Action: Interacting with the dead and the living, First Edition, John Wiley & Sons Ltd. 2020.

12. B.M. Trapp, S.D.Tallman, The effectsof household corrosive substances on silver amalgam and porcelainfused-to-metal restorations and non-restored teeth, Forensic ScienceInternational https://doi.org/10.1016/j.forsciint.2018.10.004 .

13. M. Singh,J.S.Sehrawat, V. Sharma, Sensitivity and specificity of various techniques for the trace elemental analysis on Teeth: A critical review, Brazilian Journal of Forensic Sciences, Medical Law and Bioethics, 7 (2018) 230-251.

14. L.M. Kootker, E. Plomp, S.T.M. Ammer, V. Hoogland, G.R. Davies,Spatial patterns in 87Sr/86Sr ratios in modern human dental enamel andtap water from the Netherlands: Implications for forensic provenancing, Science of the Total Environment 729 (2020) 3-8.

15. H. Baltas, M. Sirin, F. Senel, F. Devran, Determination of natural radionuclides and somemetal concentrations in human tooth samples intheRize province, Turkey International Journal of Environmental Health Researchhttps, doi.org/10.1080/09603123.2019.1625033.

16. A.M. Christensen, M.A. Smith, R.M. Thomas,Validation of X-Ray Fluorescence Spectrometry for Determining Osseous or Dental Origin of unknown material, J Forensic Sci, 57 (2012), doi: 10.1111/j.1556-4029.2011.01941.x.

17. H.A. Zimmerman, C.J.Meizel-Lambert, J.J. Schultz, M.E. Sigmanb, Chemical differentiation of osseous, dental, and non-skeletal materials in forensic anthropology using elemental analysis, Science and Justice,(2014) http://dx.doi.org/10.1016/j.scijus.2014.11.003.

18. D.H. Ubelaker, D.C.Ward, V.S. Braz, J. Stewart, The use of SEM/EDS analysis to distinguishdental and osseus tissue from other materials, J. Forensic Sci. 47 (2002)1. 4.

19. K. Nganvongpanit, J.L. Brown, K.Buddhachat, C. Somgird, C. Thitaram, Elemental analysis of Asian Elephant (Elephasmaximus) teethusing x-ray fluorescence and a comparison to other species, Biol Trace Elem Res,170 (2016)94. 105,DOI 10.1007/s12011-015-0445-x.

20. K. Nganvongpanit, K. Buddhachat, P. Piboon, T. Euppayo, P. Mahakkanukrauh, Variation in elementalcomposition of human teeth and its application for feasible species identification,Forensic Science International,http://dx.doi.org/10.1016/j.forsciint.2016.12.017.

21. K. Buddhachat, S. Klinhom, P. Siengdee, J.L. Brown, R. Nomsiri, P. Kaewmong, C. Thitaram, P. Mahakkanukrauh, K. Nganvongpanit, Elemental analysis of bone, teeth, horn andantler in different animal species using non-invasive handheld x-ray fluorescence, PLoS ONE 11,doi:10.1371/journal.pone.0155458.

22. K. Nganvongpanit, K. Buddhachat, P. Piboon, T. Euppayo, P. Kaewmong, P. Cherdsukjai, K. Kittiwatanawong, C. Thitaram, Elemental classification of the tusksof dugong (Dugong dugong) by HHXRFanalysis and comparison withother species, Scientific Reports, (2017) DOI: 10.1038/srep46167.

23. G. McLaughlin, I.K. Lednev, Spectroscopic discrimination of bone samples from various species, Am. J. Anal. Chem. 3 (2012) 161. 167.

24. H.G.M. Edwards, N.F. Hassan, N. Arya, Evaluation of Raman spectroscopy and application of chemometric methods for the differentiation of contemporary ivory specimens I: elephant and mammalian species, J. Raman Spectrosc. 37 (2006) 353. 360.

25. M. Shimoyama, H. Maeda, H. Sato, Y. Ninomiya, Y. Ozaki, Nondestructive discrimination of biological materials by near-infrared Fourier transform Raman spectroscopy and chemometrics: discrimination among hard and soft ivories of African elephantsandmammoth tusks and prediction of specific gravity of the ivories, Appl. Spectrosc.51 (8) (1997) 1154. 1158.

26. K. Alkass, B.A. Buchholz, H. Druid, K.L. Spalding, Analysis of 14C and 13C in teeth provides precise birth dating andclues to geographical origin, Forensic Sci Int. 209 (2011) 34. 41, doi:10.1016/j.forsciint.2010.12.002.

27. S.C. Zapico, D.H. Ubelaker, Applications of physiological bases of ageing to forensic sciences.Estimation of age-at-death, Ageing Research Reviews 12 (2013) 605. 617.

28. P. Malara, J. Kwapulinski, B.Malara, Do the levels of selected metals differ significantly between the rootsof carious and non-carious teeth?, Science of the Total Environment 369 (2006) 59. 68.

29. kosa et al

30. J.S. Sehrawat, M. Singh, application of trace elemental profile of known teeth for sex and age estimation of ajnala skeletal remains: a

forensic anthropological cross-validation study, Biological Trace Element Research, 193 (2020)295. 310, https://doi.org/10.1007/s12011-019-01712-8.

31. S.I. AL-Qattan, M.A. Elfawal, Significance of teeth lead accumulation in age estimation, Journal of Forensic and Legal Medicine, 17 (2010) 325-328.

32. J. Bayo, S. Moreno-Grau, M.J. Martinez, J. Moreno, J.M. Angosto, J.J. Guille´nPe´rez, L.G. Marcos, J.Moreno-Clavel,Environmentaland physiological factors affecting lead and cadmium levelsin deciduous teeth, Arch. Environ. Contam.Toxicol. 41(2001) 247. 254, DOI: 10.1007/s002440010245.

33. A. Alomary, I.F. Al-Momani, A.M. Massadeh, Lead and cadmium in human teeth from Jordan by atomic absorptionspectrometry: Some factors influencing their concentrations, Science of the Total Environment 369 (2006) 69. 75.

34. G.D. Kamenov, J.H. Curtis, Using carbon, oxygen, strontium, and lead isotopes in modern human teeth for forensic investigations: a critical overview based on data from Bulgaria, J Forensic Sci, (2017) doi: 10.1111/1556-4029.13462.

35. E. Rauch, S. Rummel, C. Lehn, A. Buttner, Origin assignment of unidentified corpses by use of stable isotoperatios of light (bio-) and heavy (geo-) elements· A case report, Forensic Science International 168 (2007) 215. 218.

36. E. Plompa, I.C.C. von Holsteina, J.M. Koornneef, R.J. Smeetsa, J.A. Baartb, T. Forouzanfarb, G.R. Davies, Evaluation of neodymium isotope analysis of human dental enamel as aprovenance indicator using 1013 amplifiers (TIMS), Science & Justice 59 (2019) 322. 331

37. M.A. Bush, R.G. Miller, J.Prutsman-Pfeiffer,P.J. Bush, Identification through x-ray fluorescence analysis of dental restorative resin materials: a comprehensive study of noncremated, cremated, and processed-cremated individuals,_J Forensic Sci, 52 (2007), doi:10.1111/j.1556-4029.2006.00308.x.

38. W. Castro, J. Hoogewerff, C. Latkoczy, J.R. Almirall, Application of laser ablation (LA-ICP-SF-MS) for the elemental analysis of boneand teeth samples for discrimination purposes, Forensic Science International, 195 (2010) 17. 27.

39. M.A. Amr, A.F.I. Helal, Analysis of trace elements in teeth by icp-ms: implications for caries, Journal of Physical Science, 21(2010) 1. 12.

40. H.M. Tvinnereima, R. Eideb, T. Riisec, Heavy metals in human primary teeth: some factors influencing the metal concentrations, The Science of the Total Environment 255 (2000) 21-27.

41. S.S. Najim, M.A. Adnan, Dental erosion by beverages and determination of trace elements in teeth by atomic absorption, American Journal of Analytical Chemistry, 7 (2016) 548-555.