

Optical Spectroscopic Analysis Techniques to Detect Elemental Profile of Human Teeth Dentine

Saifullah Jamali¹, Muhammad Aslam Khoso¹, Irfan Ali Sanjrani¹, Hussain Saleem^{3*}, Tariq Ali Siyal¹, Muhammad Ashraf^{1,2}, Mansoor Ahmed Memon¹, Ghulam Murtaza¹, Zahid Hussain Arain¹, Zaheer Ahmed Ujjan¹, Muhammad Niaz Laghari¹, Samina Saleem⁴, Nek M. Shaikh¹, Waseem A. Bhutto¹, Abdul Majid Soomro¹, Altaf H. Nizamani¹

*Corresponding Author: hussainsaleem@uok.edu.pk

¹Institute of Physics, University of Sindh, Jamshoro, Pakistan.

²Department of Basic Science, Dawood University of Engineering and Technology, Karachi, Pakistan.

³Department of Computer Science, UBIT, University of Karachi, Karachi, Pakistan.

⁴Karachi University Business School (KUBS), University of Karachi, Karachi, Pakistan.

Abstract

Numerous articles under the study and the examination of heavy metals in human teeth have been published in recent years. The heavy metal poisoning is a widespread issue emerged in toxicology area these days. It has been discovered that long-term exposure to heavy metals typically present in traces, in our everyday meals, drinking water, and in the environment as pollution causes heavy metal poisoning in human beings. Industrial effluents, Coal and Oil, as well as a variety of consumer items, such as cosmetics, can all cause this type of exposure. Teeth, which are often thought of as exoskeleton parts, store heavy metals with a high affinity and represent long-term exposure information. In this study, we have chosen and examined the sections of dentine instead, then examined the entire tooth. We have combined the work done on the examination of heavy metals in human teeth using several instrumental approaches e.g. "Optical Spectroscopic Techniques" to detect elemental profile of human teeth in the current study.

Keywords:

Heavy metals; Human teeth; Optical Spectroscopic techniques; Pollution; Toxic metals;

1. Introduction

The purpose of this study is to look at the current state of elemental analysis from the hard tissue dentine of teeth. Humans have two sets of teeth at any one moment during their lives. The "Deciduous" or "Temporary" or "Milk teeth" are the name given to one set of teeth. There are a total of 20 teeth in each jaw i.e. 04 incisors, 02 canines, and 04 molars, for a total of 10 teeth in each jaw that erupt at the age of six months and shed at the age of six years [1]. The permanent teeth, which are the second set of teeth, take their place next. There are 32 teeth in each jaw, including 04 incisors, 02 canines, 04 premolars, and 06 molars. The calcification of the permanent teeth's roots is complete until the age of 22~25 years. The crown and root sections of the tooth are the two primary elements [1] [2].

The crown of the tooth is visible in the mouth, whereas the root of the tooth is in the jaw and drops beyond the gum line. The anatomy and structure of human tooth is shown via Fig.1 [67]. Enamel, dentine, pulp, cementum, and the

periodontal ligament are the most important components of teeth. Enamel, which is made up of rock-hard minerals and calcium phosphate, is the outermost and toughest component of the tooth [3] [4] [5] [6] [7] [8] [9] [10] [11]. The dentine is a hard tissue that lies underneath the enamel and is filled with tiny channels [12] [13]. When cold or heat penetrates the tooth through this channel, enamel is damaged, causing sensitivity or discomfort [14] [15]. Pulp is the live, softer middle component of the tooth. Pulp contains nerves, connective tissue, and blood arteries. The connective tissue layer that links the roots to the jawbone and gums is known as cementum [16]. The periodontal ligament is a soft tissue that cushions the tooth against the jaw. Bony and mucosal structures surround the tooth [17] [18] [19].

Humans have been exposed to increased quantities of heavy metals through both the terrestrial and aquatic environments since the dawn of time [20]. Metals having large atomic numbers, atomic weights, and densities are known as heavy metal. Some of these metals, such as calcium, magnesium, zinc, and iron, are needed for human health, whereas others, such as cadmium, arsenic, lead, nickel, chromium, copper, and mercury, have been shown to be very hazardous to humans. Ingestion, inhalation, and dermal contact are all ways for heavy metals to enter the human body [21] [22]. Heavy metals accumulate in numerous organs, including tissues, bones, and teeth, and their chronic toxicity has an impact on human health [23] [24] [25]. Toxic metals are slowly diffuse into the human body. Once it is diffused there, they are difficult to remove and build up in the body permanently [26]. Heavy metals aid in the management of human body systems such as homeostasis, transport, and binding to cell components due to their chemical coordination and oxidation-reduction capabilities [27].

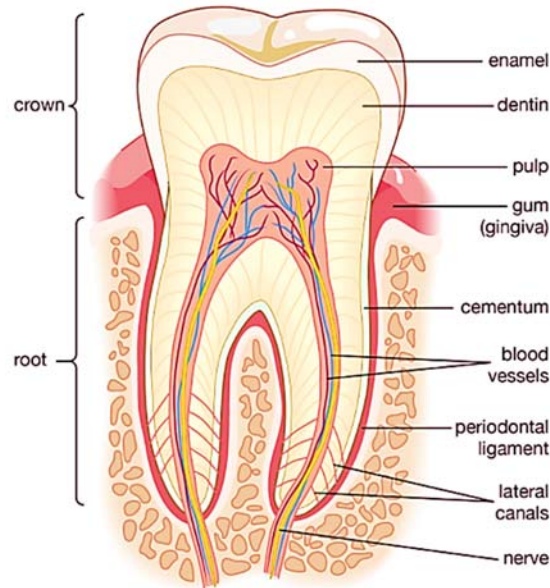


Fig. 1. Anatomy and Structure of Human Tooth [67].

By removing native metals from their normal binding sites, they connect with protein sites, producing cell dysfunction and, ultimately, severe toxicity. Heavy metal accumulation varies by organ [28]. The amount of heavy metals absorbed into the body is determined by their binding to carrier molecules. The metallothionein (small proteins) is important for heavy metal storage since it is responsible for the metal binding characteristics [29] [30] [31]. They can induce hepatotoxicity by forming a cysteine-metallothionein complex in the liver when they bind to cysteine-rich proteins like metallothionein [32] [33]. Heavy metals are absorbed into calcified tissues throughout the development stage when they are exposed to heavy metals in their mineral phase. Information regarding recent exposures can be found in urine and blood. Hair and fingernails can represent information on exposure durations ranging from a few months to years, but they can also be polluted by dyes, shampoo, nail paints, airborne dust, and other contaminants [30]. Heavy metals accumulate with strong affinity in calcified tissues (bones and teeth), and this knowledge reflects long-term exposure. Bones are generally available for sample, but teeth provide a lasting record of recent and/or historical heavy metal exposure.

Dental tissues are extremely hard, consisting of a substance comparable to that of bones. They are often regarded as an exoskeleton component [34] [35]. Because human teeth are biological tissue that is easily accessible. The heavy metal analysis technique has been used to classify individuals from human teeth in terms of heavy metal absorption and exposure. It is not necessary to investigate the entire tooth. Just chosen portions of dentine are required [36]. Human dentine tissues are part of the exoskeleton and do not undergo mineral phase turnover [37]. It has a number of benefits over other bio indicators, making it a credible

indicator of harmful heavy metal exposure in the environment. Because the dentine is surrounded by enamel and cementum, it is unaffected by the oral environment. Odontoblasts are located in the pulp and produce dentine constantly until the tooth is lost. Hard tissues of the teeth (dentine and enamel) are more useful than soft tissues such as the kidney and liver because they are not vulnerable to heavy metal changeover once ingested [34] [38]. Because metals may pass across the placental barrier, teeth also contain heavy metals ingested by the mother during pregnancy, as their production begins in the prenatal period [24] [39].

Shed deciduous teeth can be gathered from dental clinics, schools, or approaching households. Permanent teeth are gathered from dental clinics where they are commonly pulled for orthodontic or other purposes. Ancient teeth are collected from graveyards [40]. Teeth may be preserved for a long period; the only precaution is to keep them out of an acidic or leaching environment. As a result, calcified tissues on teeth preserve different heavy elements to which they are exposed and give a reliable historical record [41]. Teeth may be used to estimate age, gender, ethnicity, social standing, employment, and habits, therefore their examination is also utilized for identification. It takes a great deal of effort and care to get accurate results or data through analysis [42] [43] [44].

Laser Induced Breakdown Spectroscopy (LIBS), Atomic Absorption Spectrometry (AAS), Particle Induced X-Ray Emission (PIXE), X-Ray Fluorescence (XRF), Inductively Coupled Plasma – Mass Spectroscopy (ICP-MS), Laser Ablation Inductively Coupled Plasma-Mass Spectroscopy (LA-ICP-MS) are some of the instrumental methods used to analyze.

2. Techniques used for Analysis

Using various spectroscopic analysis techniques and several investigations have been undertaken to identify heavy metals in human teeth. The analytical procedures shown in Fig.2, discussed such as (1) Laser Induced Breakdown Spectroscopy (LIBS), (2) Atomic Absorption Spectrometry (AAS), (3) Particle Induced X-Ray Emission (PIXE), (4) X-Ray Fluorescence (XRF), (5) Inductively Coupled Plasma – Mass Spectroscopy (ICP-MS), (6) Laser Ablation Inductively Coupled Plasma-Mass Spectroscopy (LA-ICP-MS) were used for investigation of heavy metals.

2.1 Laser Induced Breakdown Spectroscopy (LIBS)

Laser Induced Breakdown Spectroscopy is a flexible technique for identifying the elements present in the material under investigation. It aids in the identification of metals and non-metals in less than a second with little sample preparation and no waste formation, as well as the detection of exogenous and endogenous substance elements in people and animals.

2.2 Atomic Absorption Spectroscopy (AAS)

It is the most widely used technique for detecting trace elements in various materials, particularly in dental samples.

2.3 Particle Induced X-Ray Emission (PIXE)

Particle Induced X-Ray Emission (PIXE) is a fast and non-destructive approach for simultaneous multi-element analysis with sensitivity in the parts-per-million (ppm) range across the board.

2.4 X-Ray Fluorescence (XRF)

Without inflicting harm to the specimens, XRF gives essential elemental information.

2.5 Inductively Coupled Plasma – Mass Spectroscopy (ICP-MS)

It is one of the most used techniques for elemental analysis since it can deliver high levels of sensitivity, precision, and accuracy. Various scholars have used this approach in a number of investigations.

2.6 Laser Ablation Inductively Coupled Plasma-Mass Spectroscopy (LA-ICP-MS)

This specific approach of laser ablation-inductively coupled plasma-mass spectrometry is beneficial for determining the change of the elemental composition in the sample. This method has been used by several researchers to assess the level of heavy metals in human tooth samples.

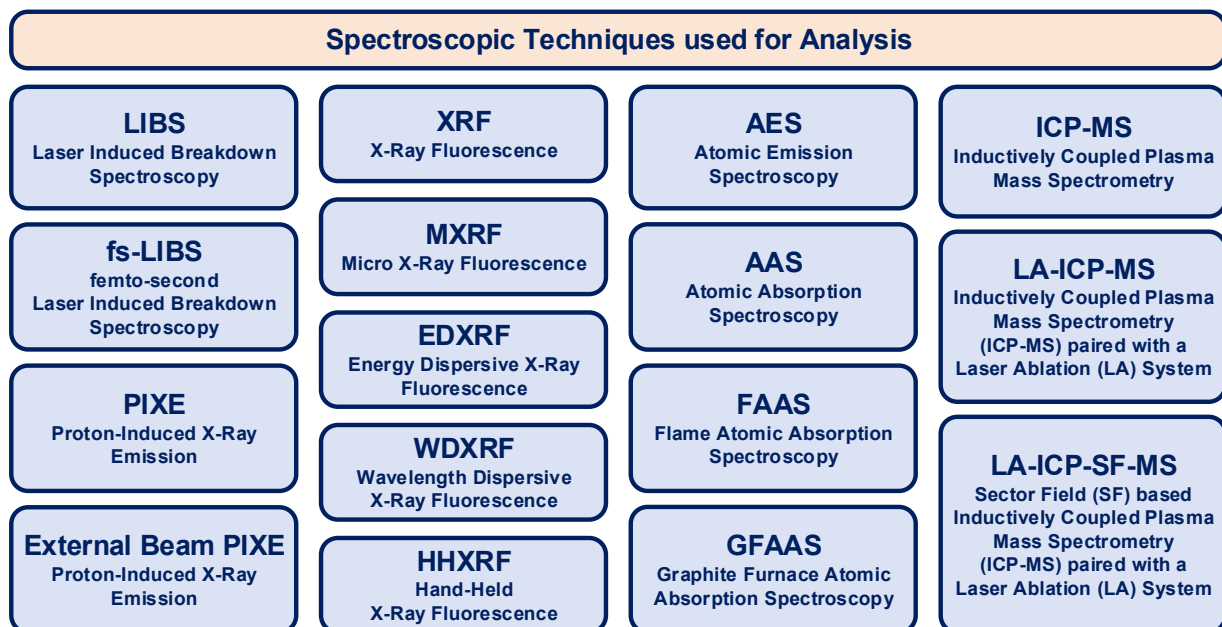


Fig.2. Various Spectroscopic Techniques used for Analysis.

3. Literature Review and Discussion

Number of researchers put their efforts to investigate and identify heavy metals in human teeth using various chemical analysis techniques and procedures in this field especially Laser Induced Breakdown Spectroscopy i.e. LIBS. The detailed description of work studied, and techniques applied with the outcome and analysis is shown in Table-I.

Alhasmi et al. [45] measured the quantities of hazardous metals including Lead, Cadmium, and Arsenic in the roots of removed teeth from smokers and non-smokers using LIBS technique. The relevant elemental concentrations for nonsmokers found as Lead (*Pb*), Cadmium (*Cd*), and Arsenic (*As*) were 23~29, 0.26~0.31, and 0.64~11 ppm; while for smokers, 35~55, 0.33~0.51, and 0.91~1.5 ppm; and for the control group, 0.17~0.31, 0.01~0.05, and 0.05~0.09 ppm.

Khalid et al., [46] used LIBS to examine human deciduous tooth samples. *Ca*, *Fe*, *Sr*, *Zn*, and *Pb* elemental concentrations were found to be greatest in enamel, followed by dentine, and finally Cementum.

LIBS was used to classify human teeth by Suyanto et al., [47] where *Ca*, *F*, *Si*, *Zn*, *Na*, *Sn*, *Ar*, *Li*, *K*, *Ce*, *Fe*, *Mn*, *Ti*, *Al*, *Cr*, and *P* have all been discovered in Indonesian human teeth.

Alvira et al., [48] established a novel technique for determining *Sr/Ca* alterations in human lower third molar enamel. The relative quantity of Strontium (*Sr*) was measured using femto-second Laser Induced Breakdown Spectroscopy (fs-LIBS).

Using AAS, Al-Jubouri et al., [49] found Cadmium (*Cd*) levels in Iraqi employees' blood, hair, saliva, and tooth samples. The exposed subjects' Cadmium levels were found to be higher.

Fischer et al., [50] used AAS to investigate variations in the concentration of certain elements (*Mn*, *Fe*, *Mg*, *Cu*, *K*, *Cr*, *Pb*, *Cd*, and *Ca*) in deciduous teeth. Teeth of older children had a statistically significant lower concentration than those of younger children.

Fischer et al., [51] also used AAS to investigate age-related alterations in human teeth. The yearly rise in *Pb* content in tooth tissues is around 0.1 g/g.

Using Flame Atomic Absorption Spectroscopy (FAAS), Olovic et al., [52] assess the amount of 12 metals in 23 tooth samples from two cities in Bosnia and Herzegovina. *Ca*, *Na*, *Mg*, *K*, *Cu*, *Zn*, and *Fe* were detected in abundance in the samples examined. Gender, geographic location, and smoking differences were more pronounced between intra groups than between inter groups.

Using GFAAS, Olympio et al., [53] investigated the risk variables linked with elevated lead levels in tooth enamel in 160 adolescents.

Using GFAAS, Barton et al., [1] examined 300 samples of 6-year-old children for lead and cadmium in deciduous teeth, scalp hair, and capillary blood. The levels of *Pb* in teeth and blood were shown to have a positive link.

Using external beam PIXE, Rao et al., [54] calculated the trace elements in various regions of human teeth. *P*, *Ca*, *V*, *Mn*, *Fe*, *Ni*, *Cu*, *Zn*, *Ba*, *As*, *Sr*, and *Pb* were measured on various sections of human teeth in this study.

Rautray et al., [55] used external PIXE to investigate the elemental profiles of enamel, cementum, and cavities in human teeth. The concentrations of ten elements, namely *P*, *Ca*, *Fe*, *Zn*, and *Pb*, in enamel are higher than those in cementum.

Opera et al., [56] studied teeth using PIXE and found *Ca*, *Cr*, *Cu*, *Fe* and *Zn*.

Dias et al., [57] used Micro X-Ray fluorescence to investigate changes in lead distribution in various bone and tooth tissues. *Pb* buildup was also measured antemortem and postmortem. The pulp and root of the tooth structure had the highest level of lead.

Guerra et al., [33] used Energy Dispersive X-Ray Fluorescence (EDXRF) to assess the distribution of hazardous components in amalgam-treated teeth.

Teruel et al., [58] used Wavelength Dispersive X-Ray Fluorescence (WDXRF) to compare the chemical makeup of human teeth to that of other animal species. Human and bovine enamel and dentine species were determined to be the most comparable among the other species studied.

Nganvongpanit et al., [59] suggested a novel approach for evaluating using Hand-Held X-Ray Fluorescence (HHXRF) if a suspected tooth is genuine human teeth or not. It was discovered that human teeth can be distinguished from those of other animals, but that sex cannot be ascertained from tooth samples.

Amr et al., [60] used ICP-MS to compare the content of trace elements in the children's primary and permanent teeth. Permanent teeth had larger concentrations of *Na*, *Mg*, *Al*, *Fe*, *Ni*, *Cu*, *Sr*, *Cd*, *Ba*, *Pb*, and *U* than children's teeth, but much less *Mn*, *Co*, *As*, *Se*, *Mo*, and *Bi*.

Asaduzzaman et al., [34] used ICP-MS to examine human teeth dentine in order to determine metal exposure owing to pollution. Chinese teeth have greater metal levels than Indian and Malay teeth, according to the study.

ICP-MS and Atomic Emission Spectroscopy (AES) were used by Escudero et al., [61] to analyze the content of 25 trace elements in 150 human coronal dentine. Both hazardous and important elements were discovered in coronal dentine, with increases in toxic (*Pb*, *Li*, *Sn*) and essential (*B*, *Ba*, *K*, *Sr*, *S*, and *Mg*) concentrations linked to tooth age but independent of sex.

Table-I. The study of various chemical analysis techniques to identify heavy metals in human teeth.

S. No	Author (Researcher)	Technique used	Description	Outcome	Ref.
1	Alhasmi et al.	LIBS Laser Induced Breakdown Spectroscopy	Measured the quantities of hazardous metals including Lead (<i>Pb</i>), Cadmium (<i>Cd</i>), and Arsenic (<i>As</i>) in the roots of removed teeth from smokers and non-smokers.	The relevant elemental concentrations for nonsmokers found as <i>Pb</i> , <i>Cd</i> , and <i>As</i> were 23~29, 0.26~0.31, and 0.64~11 ppm; while for smokers, 35~55, 0.33~0.51, and 0.91~1.5 ppm; and for the control group, 0.17~0.31, 0.01~0.05, and 0.05~0.09 ppm.	[45]
2	Khalid et al.	LIBS Laser Induced Breakdown Spectroscopy	Examined human deciduous tooth samples.	<i>Ca</i> , <i>Fe</i> , <i>Sr</i> , <i>Zn</i> , and <i>Pb</i> elemental concentrations were found to be greatest in Enamel, followed by Dentine, and finally Cementum.	[46]
3	Suyanto et al.	LIBS Laser Induced Breakdown Spectroscopy	Classified human teeth.	<i>Ca</i> , <i>F</i> , <i>Si</i> , <i>Zn</i> , <i>Na</i> , <i>Sn</i> , <i>Ar</i> , <i>Li</i> , <i>K</i> , <i>Ce</i> , <i>Fe</i> , <i>Mn</i> , <i>Ti</i> , <i>Al</i> , <i>Cr</i> , and <i>P</i> have all been discovered in Indonesian human teeth.	[47]
4	Alvira et al.	fs-LIBS femto-second Laser Induced Breakdown Spectroscopy	Established a novel technique for determining <i>Sr/Ca</i> alterations in human lower third molar enamel.	The relative quantity of Strontium (<i>Sr</i>) was measured.	[48]
5	Al-Jubouri et al.	AAS Atomic Absorption Spectroscopy	Examined human blood, hair, saliva, and tooth samples in Iraqi employees.	Found Cadmium (<i>Cd</i>) levels in Iraqi employees' blood, hair, saliva, and tooth samples. The exposed subjects' Cadmium levels were found to be higher.	[49]
6	Fischer et al.	AAS Atomic Absorption Spectroscopy	Investigated variations in the concentration of certain elements (<i>Mn</i> , <i>Fe</i> , <i>Mg</i> , <i>Cu</i> , <i>K</i> , <i>Cr</i> , <i>Pb</i> , <i>Cd</i> , and <i>Ca</i>) in deciduous teeth.	Teeth of older children had a statistically significant lower concentration than those of younger children.	[50]
7	Fischer et al.	AAS Atomic Absorption Spectroscopy	Investigated age-related alterations in human teeth.	The yearly rise in <i>Pb</i> content in tooth tissues is around 0.1 g/g.	[51]
8	Olovic et al.	FAAS Flame Atomic Absorption Spectroscopy	Assessed the amount of 12 metals in 23 tooth samples from two cities in Bosnia and Herzegovina.	<i>Ca</i> , <i>Na</i> , <i>Mg</i> , <i>K</i> , <i>Cu</i> , <i>Zn</i> , and <i>Fe</i> were detected in abundance in the samples examined. Gender, geographic location, and smoking differences were more pronounced between intra groups than between inter groups.	[52]
9	Olympio et al.	GFAAS Graphite Furnace Atomic Absorption Spectroscopy	Investigated the risk variables linked with elevated lead levels in tooth enamel in 160 adolescents.	Found rise in lead (<i>Pb</i>) levels in tooth enamel in adults.	[53]
10	Barton et al.	GFAAS Graphite Furnace Atomic Absorption Spectroscopy	Examined 300 samples of 6-year-old children for lead and cadmium in deciduous teeth, scalp hair, and capillary blood.	The levels of <i>Pb</i> in teeth and blood were shown to have a positive link.	[1]
11	Rao et al.	External Beam PIXE Proton-Induced X-Ray Emission	Calculated the trace elements in various regions of human teeth.	<i>P</i> , <i>Ca</i> , <i>V</i> , <i>Mn</i> , <i>Fe</i> , <i>Ni</i> , <i>Cu</i> , <i>Zn</i> , <i>Ba</i> , <i>As</i> , <i>Sr</i> , and <i>Pb</i> were measured on various sections of human teeth in this study.	[54]
12	Rautray et al.	External Beam PIXE Proton-Induced X-Ray Emission	Investigated the elemental profiles of enamel, cementum, and cavities in human teeth.	The concentrations of ten elements, namely <i>P</i> , <i>Ca</i> , <i>Fe</i> , <i>Zn</i> , and <i>Pb</i> , in enamel found higher than those in cementum.	[55]
13	Opera et al.	PIXE Proton-Induced X-Ray Emission	Studied teeth structures.	Found <i>Ca</i> , <i>Cr</i> , <i>Cu</i> , <i>Fe</i> and <i>Zn</i> .	[56]
14	Dias et al.	MXRF Micro X-Ray Fluorescence	Investigated changes in lead distribution in various bone and tooth tissues. <i>Pb</i> build-up i.e. ante-mortem and post-mortem was also measured.	The pulp and root of the tooth structure had the highest level of lead.	[57]
15	Guerra et al.	EDXRF Energy Dispersive X-Ray Fluorescence	Assessed the distribution of hazardous components in amalgam-treated teeth.	Identified hazardous components in teeth.	[33]

Table-1 (Continued...). The study of various chemical analysis techniques to identify heavy metals in human teeth.

S. No	Author (Researcher)	Technique used	Description	Outcome	Ref.
16	Teruel et al.	WDXRF Wavelength Dispersive X-Ray Fluorescence	Compared the chemical makeup of human teeth to that of other animal species.	Human and bovine enamel and dentine species were determined to be the most comparable among the other species studied.	[58]
17	Nganvongpanit et al.	HHXRF Hand-Held X-Ray Fluorescence	Suggested a novel approach for evaluating genuineness of a suspected human tooth.	It was discovered that human teeth can be distinguished from those of other animals, but that sex cannot be ascertained from tooth samples.	[59]
18	Amr et al.	ICP-MS Inductively Coupled Plasma Mass Spectrometry	Compared the content of trace elements in the children's primary and permanent teeth.	Permanent teeth had larger concentrations of <i>Na</i> , <i>Mg</i> , <i>Al</i> , <i>Fe</i> , <i>Ni</i> , <i>Cu</i> , <i>Sr</i> , <i>Cd</i> , <i>Ba</i> , <i>Pb</i> , and <i>U</i> than children's teeth, but much less <i>Mn</i> , <i>Co</i> , <i>As</i> , <i>Se</i> , <i>Mo</i> , and <i>Bi</i> .	[60]
19	Asaduzzaman et al.	ICP-MS Inductively Coupled Plasma Mass Spectrometry	Examined human teeth dentine to determine metal exposure owing to pollution.	Chinese teeth have greater metal levels than Indian and Malay teeth, according to the study.	[34]
20	Escudero et al.	ICP-MS and AES Inductively Coupled Plasma Mass Spectrometry and Atomic Emission Spectroscopy	Analysed the content of 25 trace elements in 150 human coronal dentine.	Both hazardous and important elements were discovered in coronal dentine, with an increase in toxic (<i>Pb</i> , <i>Li</i> , <i>Sn</i>) and essential (<i>B</i> , <i>Ba</i> , <i>K</i> , <i>Sr</i> , <i>S</i> , and <i>Mg</i>) concentrations linked to tooth age but independent of sex.	[61]
21	Castro et al.	LA-ICP-SF-MS Sector Field (SF) based Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Teeth samples from 14 distinct people were examined and performed quantitative analysis on bone and tooth samples.	It was discovered that the elemental makeup of enamel, dentine, and cementum layers enhance individual separation.	[43]
22	Abdullah et al.	LA-ICP-MS Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Examined the relative quantities of Lead (<i>Pb</i>), Mercury (<i>Hg</i>), and Manganese (<i>Mn</i>) in the enamel areas of deciduous teeth from children with ASDs and HDB.	There were no significant variations in neurotoxicant levels between children with Autism Spectrum Disorders ASD and those with High Levels of Disruptive Behavior HDB.	[62]
23	Hare et al.	LA-ICP-MS Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Done bio-imaging of trace elements in teeth.	<i>Pb</i> , <i>Zn</i> , and <i>Cd</i> concentrations were found to be greater in dentine and the areas close to the pulp.	[63]
24	Hanc et al.	LA-ICP-MS Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Studied the elements migration in human teeth.	Found Aluminium (<i>Al</i>), Barium (<i>Ba</i>), and Lanthanum (<i>La</i>).	[64]
25	Sr. Guede et al.,	LA-ICP-MS Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Analysed 23 teeth enamel and dentine samples from a Muslim community in Tauste (North Spain) in order to discover trace components and examine Medieval Muslim food trends.	The findings revealed that some people had high <i>Pb</i> levels as a result of occupational exposure to anthropogenic lead. The chemical makeup differed by sex and age, and it was shown to be linked to food consumption.	[65]
26	Horton et al.	LA-ICP-MS Inductively Coupled Plasma Mass Spectrometry (ICP-MS) paired with a Laser Ablation (LA) system	Investigated the links between dentine biomarkers of <i>Mn</i> , <i>Zn</i> , and <i>Pb</i> and later childhood behaviour.	Prenatal dentine <i>Mn</i> has been shown to be protective, while high postnatal <i>Mn</i> has been shown to increase the chance of negative behaviors. Additionally, increased <i>Mn</i> , <i>Zn</i> , and <i>Pb</i> concentrations have a negative influence on behavior.	[66]

Castro et al., [43] used a sector field based inductively coupled plasma mass spectrometry (LA-ICP-SF-MS) paired with a laser ablation system to perform quantitative analysis on bone and tooth samples. Teeth samples from 14 distinct people were examined, and it was discovered that using the elemental makeup of enamel and dentine, as well as the cementum layers, enhanced individual separation.

Using LA-ICP-MS, Abdullah et al., [62] looked the relative quantities of Lead (*Pb*), Mercury (*Hg*), and Manganese (*Mn*) in the enamel areas of deciduous teeth from children with Autism Spectrum Disorders (ASDs) and High Levels of Disruptive Behavior (HDB). There were no significant variations in neurotoxicant levels between children with ASD and those with HDB.

Hare et al., [63] used LA-ICP-MS to do bio-imaging of trace elements in teeth. *Pb*, *Zn*, and *Cd* concentrations were found to be greater in dentine and the areas close to the pulp.

Hanc et al., [64] studies the elements migration in human teeth using LA-ICP-MS and found Aluminium (*Al*), Barium (*Ba*), and Lanthanum (*La*).

Sr. Guede et al., [65] used LA-ICP-MS to analyze 23 teeth enamel and dentine samples from a Muslim community in Tauste (North Spain) in order to discover trace components and examine Medieval Muslim food trends. The findings revealed that some people had high *Pb* levels as a result of occupational exposure to anthropogenic lead. The chemical makeup differed by sex and age, and it was shown to be linked to food consumption.

Using LA-ICP-MS Horton et al., [66] investigated the links between dentine biomarkers of *Mn*, *Zn*, and *Pb* and later childhood behavior. Prenatal dentine *Mn* has been shown to be protective, while high postnatal *Mn* has been shown to increase the chance of negative behaviors. Additionally, increased *Mn*, *Zn*, and *Pb* concentrations have a negative influence on behavior.

4. Conclusion

For the identification of traces of heavy metals present in human teeth, analytical techniques such as AAS, GFAAS, ICP-MS, LA-ICP-MS, LIBS, XRF, PIXE, and electrochemical approaches have been used.

These methods can efficiently aid in the identification of various heavy metals that accumulate in human teeth as a consequence of a variety of causes such as environmental pollution, anthropogenic trace metal exposure, and an individual's nutritional state, all of which result in harmful results.

XRF, out of all of these methods, has the potential to non-destructively differentiate human teeth from those of other mammalian species based on their chemical composition.

However, atomic absorption spectrometry (AAS) is frequently utilized for the examination of human teeth, but electrochemical approaches for the detection of heavy metal residues are not generally employed.

References

- [1] Barton, H.J., 2011, Advantages of the use of deciduous teeth, hair, and lood analysis for lead and cadmium bio monitoring in children. A study of 6-year-old children from Krakow (Poland), *Bio. Trac. Elem. Res.*, 143, 637 – 658. <https://doi.org/10.1007/s12011-010-8896-6>
- [2] Goran Koch, S.P., 2017, *Pediatric dentistry: A clinical approach*, John Wiley Sons, 408.
- [3] Maeda, H., 2020, Aging and senescence of dental pulp and hard tissues of the tooth, *Front Cell Dev. Bio.*, 8, 1417. <https://doi.org/10.3389/fcell.2020.605996>
- [4] Li, Z., Liu, L., Wang, L., and Song, D., 2021, The effects and potential applications of concentrated growth factor in dentin – pulp complex regeneration, *Stem, Cell Ther.*, 12, 1 – 10. <https://doi.org/10.1186/s13287-021-02446-y>
- [5] Foster, B.L., 2012, Methods for studying tooth root cementum by light microscopy, *Int. J. Oral Sci.*, 43 (4), 199 – 128. <https://doi.org/10.1038/ijos.2012.57>
- [6] Trenouth, M.J., 2015, The origin of the terms enamel, dentine and cementum, *R. Coll. Surg. Engl.*, 5, 26 – 31. <https://doi.org/10.1308/204268514X13859766312638>
- [7] Chatzistavrou, X., Papagerakis, S., Ma, P.X., and Papagerakis, P., 2012, Innovative approaches to regenerate enamel and dentin, *Int. J. Dent.*, 856470. <https://doi.org/10.1155/2012/856470>
- [8] Andermatt, L., and Ozcan, M., 2021, Micro shear bond strength of resin composite cement to coronal enamel/dentine, cervical enamel, cemento enamel junction and root cementum with different adhesive systems, *J. Adhes. Sci. Tech.*, 35, 2079 – 2093. <https://doi.org/10.1080/01694243.2021.1872195>
- [9] Ovy, E.G., Romanyk, D.L., Nir, C.F., and Westover, L., 2021., Modelling and evaluating periodontal ligament mechanical behaviour and properties: A scoping review of current approaches and limitations, *Orth. Craniofac. Res.* <https://doi.org/10.1111/ocr.12527>
- [10] Nakano, Y., Denbesten, P., and Goldberg, M., 2021, Structure of collagen derived mineralized tissues (dentin, cementum and bone) and non-collagenous extra cellular matrix of enamel, *Extr. Mat. Bio. Dent. Tiss. Struct.*, 10, 3 – 34.

- [11] Zhai, Q., Dong, Z., Wang, W., Li, B., and Jin, Y., 2018, Dental stem cell and dental tissue regeneration, *Front. Med.*, 132 (13), 152 – 159. <https://doi.org/10.1007/s11684-018-0628-x>
- [12] Duangto, P., Janhom, A., Prasitwattanaseree, S., Mahakkanukrauh, P., and Iamaroon, A., 2016, New prediction models for dental age estimation in Thai children and adolescents, *Forensic Sci. Int.*, 266, 583.e1 – 583.e5. <https://doi.org/10.1016/j.forsciint.2016.05.005>
- [13] Nganvongpanit, K., Buddhachat, K., Piboon, P., Euppayo, T., and Mahakkanukrauh, P., 2017, Variation in elemental composition of human teeth and its application for feasible species identification., *Foren. Sci. Int.*, 271, 33 – 42. <https://doi.org/10.1038/srep46167>
- [14] Lin, M., Xu, F., Lu, T.J., and Bai, B.F., 2010, A review of heat transfer in human tooth – experimental characterization and mathematical modeling, *Dent. Mater.*, 26, 501 – 513. <https://doi.org/10.1016/j.dental.2010.02.009>
- [15] Dostalova, T., Jelinkova, H., Remes, M., Šulc, J., and Němec, M., 2016, The use of the Er: YAG laser for bracket debonding and its effect on enamel damage, *Photom. Las. Surg.*, 34, 394 – 9. <https://doi.org/10.1089/pho.2016.4115>
- [16] Morgenthal, A., Zaslansky, P., and Fleck, C., 2021, Cementum thickening leads to lower whole tooth mobility and reduced root stresses: An in silico study on aging effects during mastication, *J. Struct. Biol.*, 213, 107726. <https://doi.org/10.1016/j.jsb.2021.107726>
- [17] Agrawal, A., and Tiwari, A.K., 2021, Dentoalveolar Fractures, *Max. Tra.*, 159 – 176.
- [18] Mohammadi, A., Hassani, A., and Fazlisahehi, O., 2021, Use of buccal fat pad in facial cosmetic surgery, *Integr. Proced. Facial Cosmet. Surg.*, 887 – 901.
- [19] Galler, K.M., and D'Souza, R.N., 2011, Tissue engineering approaches for regenerative dentistry, *Regen. Med.*, 6, 111 – 124. <https://doi.org/10.2217/rme.10.86>
- [20] Ali, H., Khan, E., and Ilahi, I., 2019, Environmental chemistry and ecotoxicology of hazardous heavy metals: Environmental persistence, toxicity, and bioaccumulation, *J. Chem.*, 6730305. <https://doi.org/10.1155/2019/6730305>
- [21] Engwa, G.A., Ferdinand, P.U., Nwalo, F.N., and Unachukwu, M.N., 2019, Mechanism and health effects of heavy metal toxicity in humans, *Pois. Mod. Worl.*
- [22] Duruibe, J.O., and Egwurugwu, M.O.C.O, 2007, Heavy metal pollution and human biotoxic effects, *Int. J. Phys. Sci.*, 2, 112 – 118.
- [23] Jamali, S., Bhutto, W.A., Nizamani, A.H., Saleem, H., Khaskheli, M.A., Soomro, A.M., Sahito, A.G., Shaikh, N.M., Saleem, S., 2019, Spectroscopic Analysis of Lithium Fluoride (LiF) using Laser Ablation, *IJCSNS*, 19 (8), 127-134. http://paper.ijcsns.org/07_book/201908/20190819.pdf
- [24] Fortes, J., Maria, D., Carceles, P., Luna, A., Laserna, J., 2006, *International journal Legal Media*, <https://doi.org/10.1007/s00414-041-1131-9>
- [25] Jan, A.T., Azam, M., Siddiqui, K., Ali, A., Choi, I., and Haq, Q.M.R., 2015, Heavy metals and human health: mechanistic insight into toxicity and counter defense system of antioxidants, *Int. J. Mol. Sci.*, 16, 29592 – 29630. <https://doi.org/10.3390/ijms161226183>
- [26] Nawrocka, A., Piwonski, I., Sauro, S., Porcelli, A., Hardan, L., and Szymanska, M.L., 2021, Traditional microscopic techniques employed in dental adhesion research - Applications and protocols of specimen preparation, *Biosens.*, 11, 408. <https://doi.org/10.3390/bios11110408>
- [27] Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B.B., and Beeregowda, K.N., 2014, Toxicity, mechanism and health effects of some heavy metals, *Inter. Toxicol.*, 7, 60. <https://doi.org/10.2478/intox-2014-0009>
- [28] Honda, K., Sahrul, M., Hidaka, H., and Tatsukawa, R., 1983, Organ and tissue distribution of heavy metals, and their growth-related changes in antarctic fish, pagothenia borchgrevinki, *Agric. Biol. Chem.*, 47, 2521 – 2532. <https://doi.org/10.1080/00021369.1983.10865986>
- [29] Calisi, A., Lionetto, M.G., De Lorenzis, E., Leomanni, A., and Schettino, T., 2014, Metallothionein induction in the coelomic fluid of the earthworm *Lumbricus terrestris* following heavy metal exposure: A short report, *Biomed Res. Int.*, 109386. <https://doi.org/10.1155/2014/109386>
- [30] Samuel, M.S., Datta, S., Khandge, R.S., and Selvarajan, E., 2021, A state of the art review on characterization of heavy metal binding metallothionein's proteins and their widespread applications, *Sci. T. Environ.*, 775. <https://doi.org/10.1016/j.scitotenv.2021.145829>
- [31] Li, L.S., Meng, Y.P., Cao, Q.F., Yang, Y.Z., Wang, F., Jia, H.S., Wu, S.B., and Liu, X.G., 2016, Type 1 metallothionein (ZjMT) is responsible for heavy metal tolerance in *Ziziphus jujuba*, *Biochem.*, 81, 565 – 573. <https://doi.org/10.3390/ijms160816750>
- [32] Batool, J., Amin, N., Jamil, Y., Shaikh, N.M., and Islam, S.A., 2021, Rapid elemental analysis of human teeth using laser induced breakdown spectroscopy, *Phys. B. Cond. Mat.*, 602, 412495. <https://doi.org/10.1016/j.physb.2020.412495>

- [33] Guerra, M., Ferreira, C., Carvalho, M.L., Santos, J.P., and Pessanha, S., 2016, Distribution of toxic elements in teeth treated with amalgam using μ -energy dispersive X-ray fluorescence, *Spectrochim. Acta Part B At. Spectrosc.*, 122, 114 – 117. <https://doi.org/10.1016/j.sab.2016.06.006>
- [34] Asaduzzaman, K., Khandaker, M.U., Binti Baharudin, N.A., Amin, Y.B.M., Farook, M.S., Bradley, D.A., and Mahmoud, O., 2017, Heavy metals in human teeth dentine: A bio-indicator of metals exposure and environmental pollution, *Chemo.*, 176, 221 – 230. <https://doi.org/10.1016/j.chemosphere.2017.02.114>
- [35] Sharma, G.K.S., 2020, Trend in the analysis of heavy metals in human teeth dentine: a review, *J. Indo-Pacific Acad. Forensic Odontol.*, 93 – 112.
- [36] Sahebi, S., Moazami, F., and Abbott, P., 2010, The effects of short-term calcium hydroxide application on the strength of dentine, *Dent. Traumatol.*, 26, 43 – 46. <https://doi.org/10.1111/j.1600-9657.2009.00834.x>
- [37] Koubi, G., Colon, P., Franquin, J.C., Hartmann, A., Richard, G., Faure, M.O., and Lambert, G., 2013, Clinical evaluation of the performance and safety of a new dentine substitute, biodentine, in the restoration of posterior teeth - a prospective study, *Clin. Oral Investig.*, 17, 243 – 249. <https://doi.org/10.1007/s00784-012-0701-9>
- [38] Asaduzzaman, K., 2017, Natural radioactivity and heavy metal pollutants in staple foodstuff and human teeth collected from selected areas in peninsular Malaysia, *Fac. Sci. Univ. Malaya Kuala Lumpur*.
- [39] Neeti, K., Prakash, T., 2013, Effects of Heavy Metal Poisoning during Pregnancy, *Int. Res. J. Environ. Sci.*, 2, 88 – 92.
- [40] Kamalak, H., Canbay, C.A., Yigit, O., and Altin, S., 2018, Physico-mechanical and thermal characteristics of commercially available and newly developed dental flowable composites, *J. Mol. Struct.*, 1156, 314 – 319. <https://doi.org/10.1016/j.jdent.2014.05.009>
- [41] Budd, P., Montgomery, J., Cox, A., Krause, P., Barreiro, B., and Thomas, R.G., 1998, The distribution of lead within ancient and modern human teeth: Implications for long-term and historical exposure monitoring, *Sci. Total Environ.*, 220, 121 – 136. [https://doi.org/10.1016/s0048-9697\(98\)00244-7](https://doi.org/10.1016/s0048-9697(98)00244-7)
- [42] Báez, A., Belmont, García, R., Carlos, J., and Báez, A., 2004, Cadmium and Lead levels in deciduous teeth of children living in Mexico city, *Rev. Int. Contam. Ambient.*, 20, xx – xx.
- [43] Castro, W., Hoogewerff, J., Latkoczy, C., and Almirall, J.R., 2010, Application of laser ablation (LA-ICP-SF-MS) for the elemental analysis of bone and teeth samples for discrimination purposes, *Forensic Sci. Int.*, 195, 17 – 27. <https://doi.org/10.1016/j.forsciint.2009.10.029>
- [44] Kolmas, J., Kalinowski, E., and Wojtowicz, A., 2010, Mid-infrared reflectance micro spectroscopy of human molars: chemical comparison of the dentine-enamel junction with the adjacent tissues, *J. Mol. Struct.*, 1-3 (966), 113 – 121. <https://doi.org/10.1016/j.molstruc.2009.12.023>
- [45] Alhasmi, A.M., Nasr, M.M., Gondal, M.A., Shafik, S., and Habibullah, Y.B., 2015, Detection of toxic elements using laser-induced breakdown spectroscopy in smokers; and nonsmokers; teeth and investigation of periodontal parameters, *Appl. Opt.*, 54 (24), 7342 – 7349. <https://doi.org/10.1364/AO.54.007342>
- [46] Khalid, A., Bashir, S., Akram, M., and Hayat, A., 2015, Laser-induced breakdown spectroscopy analysis of human deciduous teeth samples, *Lasers Med. Sci.*, 309 (30), 2233 – 2238.
- [47] Zaytsev, S.M., Popov, A.M., Zorov, N.B., Suyanto, H., Trisnawati, N.L.P., Putra, I.K., and Suprihatin, I.E., 2018, Characterization of human teeth by laser-induced breakdown spectroscopy, *J. Phys. Conf. Ser.*, 1120, 012018.
- [48] Alvira, P., Tomás-Pejó, E., Ballesteros, M., and Negro, M.J., 2010, Pretreatment technologies for an efficient bioethanol production process based on enzymatic hydrolysis: A review, *Bioresour. Technol.*, 101, 4851 – 4861. <https://doi.org/10.1016/j.biortech.2009.11.093>
- [49] Al-Jubouri, R.H., 2012, Assessment of cadmium levels in Blood, hair, saliva and teeth in a sample of Iraqi workers and detection of dental findings, *J. Bagh Coll. Dent.*, 24.
- [50] Fischer, A., Wiechuła, D., and Misztela, C.P., 2013, Changes of concentrations of elements in deciduous teeth with age, *Biol. Trace Elem. Res.*, 154, 427 – 432. <https://doi.org/10.1007/s12011-013-9744-2>
- [51] Fischer, A., and Wiechuła, D., 2016, Age-Dependent Changes in Pb Concentration in Human Teeth, *Biol. Trace Elem. Res.*, 47 – 54. <https://doi.org/10.1007/s12011-016-0643-1>
- [52] Olovčić, A., Ramić, E., and Memić, M., 2019, Human enamel and dentin: Effect of gender, geographic location and smoking upon metal concentrations, *Clin. Analys.*, 53, 245 – 261. <https://doi.org/10.1080/00032719.2019.1646753>
- [53] Olympio, K.P.K., Naozuka, J., Oliveira, P.V., Cardoso, M.R.A., Bechara, E.J.H., and Günther, W.M.R., 2010, Association of dental enamel lead levels with risk factors

- for environmental exposure, *Rev. Saudi Publica.*, 44, 851 – 858.
- [54] Rao, A.P.R.V., 2010, Estimation of trace elements in various parts of human teeth using external beam PIXE, *IJPA.*, 2, 123 – 134.
- [55] Rautray, T.R., Das, S., and Rautray, A.C., 2010, In situ analysis of human teeth by external PIXE, *Nucl. Inst. Meth. Phys. Res. Sect. Beam Interact. Mater. Atoms.*, 268, 2371 – 2374. <https://doi.org/10.1016/j.nimb.2010.01.004>
- [56] Oprea, C., Szalanski, P.J., Gustova, M.V., Oprea, et al, 2009, XRF detection limits for dental tissues of human teeth, *Vacuum.*, 83, S166 – S168. <https://doi.org/10.1016/j.vacuum.2009.01.054>
- [57] Dias, A.A., Carvalho, M., Carvalho, M.L., and Pessanha, S., 2015, Quantitative evaluation of ante-mortem lead in human remains of the 18th century by triaxial geometry and bench top micro-X-ray fluorescence spectrometry, *J. Anal. At. Spectrom.*, 30, 2488 – 2495. <https://doi.org/10.1039/C5JA00340G>
- [58] Teruel, J.D.D., Alcolea, A., Hernández, A., and Ruiz, A.J.O., 2015, Comparison of chemical composition of enamel and dentine in human, bovine, porcine and ovine teeth, *Arch. Oral Biol.*, 60, 768 – 775. <https://doi.org/10.1016/j.archoralbio.2015.01.014>
- [59] Nganvongpanit, K., Soponteerakul, R., Kaewkumpai, P., Punyapornwithaya, V., Buddhachat, K., et al, 2017, Osteoarthritis in two marine mammals and 22 land mammals: learning from skeletal remains, *J. Anat.*, 231, 140 – 155. <https://doi.org/10.1111/joa.12620>
- [60] Amr, M.A., 2011, Trace elements in Egyptian teeth, *Int. J. Phys. Sci.*, 6, 6241 – 6245. <https://doi.org/10.5897/IJPS09.307>
- [61] Fernández-Escudero, A.C., Legaz, I., Prieto-Bonete, G., López-Nicolás, M., Maurandi-López, et al., 2020, Aging and trace elements in human coronal tooth dentine, *Sci. Rep.*, 10. <https://doi.org/10.1038/s41598-020-66472-1>
- [62] Abdullah, M.M., Ly, A.R., Goldberg, W.A., Clarke-Stewart, K.A., Dudgeon, J.V., Mull, C.G., Chan, T.J., Kent, E.E., Mason, A.Z., and Ericson, J.E., 2011, Heavy metal in children's tooth enamel: Related to autism and disruptive behaviors, *J. Autism Dev. Disord.*, 42, 929 – 936. <https://doi.org/10.1007/s10803-011-1318-6>
- [63] Hare, D., Austin, C., Doble, P., and Arora, M., 2011, Elemental bio-imaging of trace elements in teeth using laser ablation-inductively coupled plasma-mass spectrometry, *J. Dent.*, 39, 397 – 403. <https://doi.org/10.1016/j.jdent.2011.03.004>
- [64] Hanć, A., Olszewska, A., and Baralkiewicz, D., 2013, Quantitative analysis of elements migration in human teeth with and without filling using LA-ICP-MS, *Microchem. J.*, 110, 61 – 69. DOI: 10.1016/j.microc.2013.02.006
- [65] Guede, I., Zuluaga, M.C., Ortega, L.A., Alonso-Olazabal, A., et al., 2017, Analyses of human dentine and tooth enamel by laser ablation-inductively coupled plasma-mass spectrometry (LA-ICP-MS) to study the diet of medieval Muslim individuals from Tauste (Spain), *Microchem. J.*, 130, 287 – 294. <https://doi.org/10.1016/j.microc.2016.10.005>
- [66] Horton, M.K., Hsu, L., Henn, B.C., Margolis, A., Austin, C., et al., 2018, Dentine biomarkers of prenatal and early childhood exposure to manganese, zinc and lead and childhood behavior, *Environ. Int.*, 121, 148 – 158. <https://doi.org/10.1016/j.envint.2018.08.045>
- [67] Britannica, "Britannica Encyclopaedia," Encyclopædia Britannica, Inc., 2013. [Online]. Available: <https://www.britannica.com/science/tooth-anatomy>. [Accessed 2022].