REVIEW ARTICLE

Influence of trace elements on dental enamel properties: A review

Zeeshan Qamar,[1] Prof. Dr. Zubaidah Binti Haji Abdul Rahim,[2] Hooi Pin Chew,[3] Tayyaba Fatima[4]

Abstract

Dental enamel, an avascular, irreparable, outermost and protective layer of the human clinical crown has a potential to withstand the physico-chemical effects and forces. These properties are being regulated by a unique association among elements occurring in the crystallites setup of human dental enamel. Calcium and phosphate are the major components (hydroxyapatite) in addition to some trace elements which have a profound effect on enamel. The current review was planned to determine the aptitude of various trace elements to substitute and their influence on human dental enamel in terms of physical and chemical properties.

Keywords: Dental enamel, Trace elements, Chemical properties, Hydroxyapatite.

Introduction

In human body, dental enamel is the hardest tissue. It is highly mineralised containing 92-96% inorganic, 1-2% organic and 3-4% water components in terms of weight/weight (W/W). Inorganic component mostly comprises hydroxyapatite (HAp) which has a chemical formula $Ca_{10}(PO_4)_6(OH)_2$ (calcium hydroxyapatite).[1] Organic part comprises proteins, mainly amelogenin, ameloblastin and tuftelin, with traces of proteoglycans and lipoids which mostly disappear upon enamel maturation. The distinctive composition of dental enamel makes it non-vital, brittle and irreparable. Therefore, underlying support from dentine is obligatory for proper functioning of tooth. On involvement of dentine with caries, the unsupported enamel fractures effortlessly.

The thickness of human dental enamel varies at different surfaces of tooth. The enamel is thickest at the cusps and thinnest at the cervical margin of a human tooth. The layer of enamel covering the cusps is about 2.5mm thick. The enamel retains more hardness and resistance towards wear and these assets make it distinctive from all other tissues of human body. The surface enamel layer is harder, radio-opaque and less porous compared to the subsurface.[1,2]

Enamel wears off with ageing and the teeth also become dark. The reason for the darkening of teeth is reduced enamel thickness which occurs because of wear, trauma and other factors, as a consequence the colour of dentine beneath is reflected on the surface.[3] Boyde defined the structure of dental enamel and expounded that crystals are organised within enamel prisms that run from the dentino-enamel junction (DEJ) to the tooth surface.[4]

Substitution of trace elements in tooth enamel

The inorganic component comprises well-defined and tightly-packed nano apatite crystals of calcium and phosphate with small amounts of trace elements. The variation in the arrangement and the size of the apatite crystals of enamel does affect its hardness and optical properties. This raises a point of key interest to the factors affecting and determining the size of the enamel apatite crystals. One of the possible reasons could be that the protein content in enamel might affect the size of the crystals. But Eimar reported that the protein concentration in enamel has no connotation with the crystallographic structure of the mature teeth.[5] Ghadimi described that the presence of trace elements could have an influence on the size of enamel apatite crystals.[6] Some trace elements have potential to expand the lattice parameters of the synthetic apatite crystal cell along the a-axis such as ferrous (Fe^{2+}), ferric (Fe^{3+}), strontium (Sr^{2+}) and zinc ions (Zn^{2+}) (molar fraction >10%) whereas it shrinks with silicate (SiO_4^{4-}), carbonate (CO_3^{2-}), magnesium (Mg^{2+}), Zn^{2+} (molar fraction < 10%) and titanium ions (Ti^{4+}). SiO_4^{4-}, CO_3^{2-}, Zn^{2+}, Fe^{2+}, Fe^{3+} and strontium ions (Sr^{2+}) can increase the lattice parameters along the c-axis, decreased by Mg^{2+}, nickelous (Ni^{2+}), chromic (Cr^{3+}), cobaltous (Co^{2+}) and Ti^{4+}. So, the crystallinity and crystal size (average length of individual crystals) of the synthetic apatite can be increased by elements like Cr^{3+}, Co^{2+} and Ni^{2+} in contrast to SiO_4^{4-}, Zn^{2+}, CO_3^{2-}, Fe^{2+}, Ti^{4+}, Sr^{2+}, cerium ion (Ce^{3+}) and Mg^{2+}.[6,7]

Nevertheless, trace elements are lower in concentration, but they have a significant role to maintain body health. Source of trace elements in human body is via ingestion of food or on exposure to the environment, incorporated into the apatite structure of the enamel. Curzon investigated the role

[1]Department of Oral Biology, Ziauddin University, Karachi, Pakistan, [1,2,4]Department of Oral Biology and Biomedical Sciences, [3]Department of Restorative Dentistry, Faculty of Dentistry, University Malaya, Kuala Lumpur, Malaysia.
Correspondence: Prof. Dr. Zubaidah Binti Haji Abdul Rahim.
Email: zubaidarster@gmail.com, zubaidar@um.edu.my

of trace elements in preventing dental caries suggesting fluoride ion (F^-), aluminium ion (Al^{3+}), Fe^{2+}, Fe^{3+}, selenium ion (Se^{4+}) and Sr^{2+} are associated in decreasing the prevalence (low risk) of dental caries, whereas presence of manganese (Mn^{2+}), cupric (Cu^{2+}) and cadmium ion (Cd^{2+}) is associated with increasing the risk.[8]

Despite the significance of trace elements on enamel haemostasis, their role on the crystallographic and physical properties remains mysterious.[9,10] The connotation between physical and chemical properties of tooth enamel with trace elements incorporated will be discussed in detail.

Fluoride (F^-)

F-can replace hydroxyl ion in the apatite crystal structure. This substitution causes a change in behaviour and solubility of apatite.[1]

Selenium (Se^{4+})

A non-metallic element, spread widely in nature absorbed by the body via food intake or inhalation.[10] Monteil reported Se^{4+} incorporation in synthetic HApvia anionic exchange of phosphate with selenite (1:1 substitution ratio).[11] Ghadim reported Se^{4+} has an association with enamel lattice parameters along the a-axis and c-axis.[5] The ionic radius of Se^{4+} (0.50 Å) is larger than that of P^{5+} (Phosphate) (0.35 Å), For that reason, on substitution in synthetic HAp the lattice parameter increases.[12]

Chromium (Cr^{3+})

A heavy metal which is present in small amount in human body has an aptitude to control fat and sugar metabolism. In addition, it has the potential to increase growth of muscles and tissues. The ionic radius of Cr^{3+} (0.69 Å) is smaller than Ca^{2+} (0.99 Å), though Cr^{3+} has a potential to substitute Ca^{2+} in synthetic HAp. Thus, Cr^{3+} will decrease the lattice parameters along a-axis and c-axis.[7]

Nickel (Ni^{2+})

Ni^{2+} (0.72 Å) is a toxic metal with the ionic radius larger than of Ca^{2+} (0.99 Å). Therefore, on substituting Ca^{2+} it will decrease the lattice parameters along c-axis of the cell in synthetic Hap.[8] It substitutes Ca^{2+} (I) to bond with oxygen and phosphates to form nickelous phosphate $Ni_3(PO_4)_2$.[13] The observations were analogous to that of Ghadimi who reported inverse association between Ni^{2+} concentration and the crystal size in tooth enamel.[5] Furthermore, the study reported a strong relation of Ni^{2+} with the occurrence of carbonate type B though the phenomenon is unknown and yet to be studied.[5]

Cobalt (Co^{2+})

Co^{2+} is a toxic environmental metal that enters the body via food, water and air. Elkabouss reported Co^{2+} substitutes Ca^{2+} in synthetic HAp and is written as $Ca_{10-x}Co_x(PO_4)_6(OH)_2$.[15] The maximum exchange of cobalt with calcium is 1.35wt% of Co^{2+}.[14] Ghadimi reported the Co^{2+} incorporation in enamel has strong negative association in substitution of carbonate type B (carbonate ion substituting phosphate group).[5,15] However, further studies are required to explore the actual phenomena.

Lead (Pb^{2+})

A heavy metal that is harmful and poisonous for the human body,[9] Pb^{2+} (1.2 Å) can replace Ca^{2+} ions (0.99 Å) at 2nd site of Ca in synthetic Hapand written as $Pb_{(10-x)}Ca_x(PO_4)_6(OH)_2$.[16] Pb^{2+} sorption by HAp in lower concentration decreases crystal size.[16] It was supported by an investigation proving a negative correlation of Pb^{2+} with the size of enamel apatite.[5]

Manganese (Mn^{2+})

Mn^{2+} is another trace element also being incorporated in enamel by ingestion via food, air and water.[9] Mn^{2+} also has a potential to replace Ca^{2+} in HAp.[17] Various studies have conveyed the capability of Mn^{2+} to incorporate in synthetic HAp without disturbing the crystal domain size.[17]

Iron (Fe^{2+}/Fe^{3+})

It is an element of indispensable importance, enters the body via ingestion of food, predominantly green vegetables. Low reported Fe^{2+} lower concentration in enamel and its effect on the carbonate type A of synthetic HAp (carbonate ion substituting for both OH^- in the c-axis channel of apatite).[15]

Titanium (Ti^{4+})

The Ti^{4+} (0.68 Å) ionic radius is smaller than Ca^{2+} (0.99 Å). Hence, substitution of Ca^{2+} ions in synthetic HAp by Ti^{4+} decreases the lattice parameters and crystal domain size,[6] thus making an agreement with other studies related to the Ti^{4+} concentration in enamel.[5]

Effect on Solubility of Enamel

The incorporation of F^- in the apatite declines its solubility product (Ksp), making the apatite structure more acid resistant. Eanes also proposed that the disarranging of OH^- by F^- has a stabilising outcome on the crystal lattice.[18] This action of F^- is significant in caries prevention. Frazier found that the addition of F^- in apatite structure during tooth development augments the crystallinity of the apatite.[19]

Muhler reported insignificant effect of Se^{4+} increasing concentration on enamel resistance to dental erosion/ caries, whereas Bowen studies suggested a direct relation of Se^{4+} with enamel dissolution.[20,21]

Cr^{3+} has a disadvantage due to its potential of altering the enamel appearance. Its effect on the dissolution of enamel and caries resistance is yet to be investigated.

Rodrigues reported that Co^{2+} incorporation alters the colour of enamel to cream or white compared to the control group, but has no effect on the micro-hardness of the tooth.[22]

Studies by Gerlach described that teeth exposed to high Pb^{2+} concentrations did not show macroscopic variations in the mature and developing enamel.[23] Brudevold found that the manifestation of dental caries is more prevalent in individuals with higher Pb^{2+} in tooth enamel, though they also had higher content of F^- as well in enamel compared to the normal individual.[24]

Currently Ti^{4+} is commonly used in biomaterials and bio-applications.[25] Being inert, it is biocompatible with the human body. Having high strength, lower modulus of elasticity and density, it is compatible enough to integrate with bone and other tissues, making its wide use for implants.

Buzalaf suggested that increasing iron concentration has inhibitory effect on the dissolution, therefore iron supplements in the form of food and beverages can play a dynamic role in reducing dental caries and erosion.[26] The mechanism of protection is not well established, but Torell proposed that the possible reason could be the formation of an acid-resistant and thin hydrous iron oxide coating on the surface.[27]

Al^{3+} concentration increases with age in the human body and its high content can lead to brain and skeletal disorders. Higher content of Al^{3+} can cause enamel discolouration. A negative correlation was reported between the Al^{3+} concentration and length of enamel cracks, proposing that enamel with lower content of Al^{3+} tends to have longer cracks and vice versa.[5] The phenomenon of this correlation is yet to be scrutinised. Putt investigated the effect of Al^{3+} on human enamel and stated that all compounds except those forming stable Al-complexes have a potential to decrease enamel dissolution.[28]

The Zn^{2+} concentration ranges from 430 to 2100 ppm varying on different surfaces of the tooth. A large proportion of Zn^{2+} in enamel is deposited before tooth eruption, whereas its post-eruptive deposition is irregular. Studies with synthetic HAp showed that Zn^{2+} is readily acquired by the apatite as it competes with Ca^{2+} and can attain position on the apatite crystal, making the HAp resistant to the acid dissolution.[29]

Trace elements have their respective cariogenic effects (Table-1).[30]

Table-1: Trace elements with their anti-cariogenic effect.[30]

Anti-Cariogenic Effect	Elements
Strong	F^-, P^{+4}
Mild	Cu^{2+}, Sr^{2+}, B, Li^+, Au^+
Weak/Doubtful	Be^{2+}, Co^{2+}, Mn^{2+}, Sn^{2+}, Zn^{2+}, Br, I^-
Inert	Ba^{2+}, Al^{3+}, Ni^{2+}, Fe^{2+}, Fe^{3+}, Pd^{2+}, Ti^{4+}
Caries Promoters	Se^{4+}, Mg^{2+}, Cd^{2+}, Pt^{2+}, Pb^{2+}, Si

Fluoride ion (F^-), Phosphorous ion (P^{+4}), Cupric ion (Cu^{2+}), Strontium ion (Sr^{2+}), Boron ion (B), Litium ion (Li^+), Gold ion (Au^{3+}), Beryllium (Be^{2+}), Cobaltous ion (Co^{2+}), Manganous ion (Mn^{2+}), Stannous ion (Sn^{2+}), Zinc ion (Zn^{2+}), Bromide ion (Br^-), Iodide ion (I^-), Barium ion(Ba^{2+}), Aluminium ion (Al^{3+}), Nickelous ion (Ni^{2+}), Ferrous ion (Fe^{2+}), Ferric ion (Fe^{3+}), Palladium ion (Pd^{2+}), Titanium ion (Ti^{4+}), Selenium ion (Se^{4+}), Magnesium ion (Mg^{2+}), Cadmium ion (Cd^{2+}), Platinum ion (Pt^{2+}), Plumbous ion (Pb^{2+}), Silicone ion (Si).

Distribution of trace element

Trace elements have variable distribution in enamel. Cardenas reported that higher content of I^- and Yb^{2+} is detectable near DEJ than the outer layer of enamel i.e., approximately 0.04%.[31] Whereas 0.1% of Al^{3+} is present in the outer layer of enamel and lowest near DEJ, absence in the intermediate surface.[31] For bromine ion (Br^-) nearly 0.01% is present in the inner layer of the enamel; 0.06% of silicon ion (Si) is reported in the entire enamel thickness, particularly in the inter-cuspid zone of the tooth; and potassium ion (K^+) and Tn^{4+} have an equal distribution throughout the enamel thickness.[31]

Cardenas observed 0.09% of Sr^{2+} near DEJ and on the cuspal tips.[31] He further reported the presence of 0.08% barium ion (Ba^{2+}) in the middle of both buccal and palatal cusps.

Optical phenomena due to trace elements

Increasing content of trace elements in enamel leads to variations in the optical properties. Incorporation of higher Fe^{2+}/Fe^{3+} or I-content leads to black pigmentation of teeth. Increase in concentration of silver nitrate ($AgNO_3$) and Mn^{2+} leads to brown or black pigmentation of teeth. Ni^{2+} and Cu^{2+} produce green stains whereas Cd2+leads to yellow staining.[32]

Studies have reported the association between crystal domain size and the optical properties of Ti^{4+}. The phenomenon states that the larger the crystal domain size, the higher is the transparency. The reason behind this fact is that more light can be scattered from the enamel comprising small crystals.[4]

Hardness

Ti^{4+} is absorbed via ingestion of food like candies, sweets and chewing gums.[33] Ghadimi reported the presence of Ti^{4+} in tooth enamel and its association with the increasing hardness of tooth.[4,5] Since there is an inverse association reported to crystal domain and nanocrystal size in enamel, therefore it increases the enamel hardness. The effect on hardness by other trace elements has not been reported yet; the possible reason could be the negative effects on the dental enamel and compromised aesthetics.

As for teeth with higher Pb^{2+}, on longitudinal micro-hardness of mature enamel, no transformation was observed.[23]

Sources of trace elements

The concentration of Al^{3+}, iodide ion (I^-), Se^{4+}, Sr^{2+}, Ni^{2+}, Cu^{2+}, Mn^{2+}, plumbous ion (Pb^{2+}), and Co^{2+} are higher in dental enamel than dentine which has abundant Fe^{2+}/Fe^{3+} and F^-. The amount of trace elements varies in different enamel layers. Elements such as Pb^{2+}, Zn^{2+}, Mn^{2+}, Fe^{2+}/Fe^{3+} and F^- are more in the outer layer than in the sub-and inner layers. This relates to the reports that the elements such as Mn^{2+} and Fe are incorporated in enamel from external environment or deposited during the process of calcification. The sources of trace elements entering human body are air, food and water. Below we will discuss dental appliances and fluids as possible sources of trace elements in dental enamel.

Saliva

Saliva, an oral fluid, is rich in several trace elements. It continuously bathes the teeth and is known to alter the composition of dental enamel. The composition and concentration of saliva varies between individuals. The range of various detectable trace elements in saliva are known (Table-2).[34]

Table-2: Concentration of trace elements in saliva.[34]

Element	Range of Elements
Cu^{2+}	(20-321 μgL^{-1})
Zn^{2+}	(28-358 μgL^{-1})
Mg^{2+}	(2.53-34.13 mgL^{-1})
Ca^{2+}	(21.5-170 mgL^{-1})
Al^{3+}	(25-102 μgL^{-1})
Sr^{2+}	(9-26 μgL^{-1})
Mn^{2+}	(2.3-5 μgL^{-1})
Fe^{2+}/Fe^{3+}	(25-160 μgL^{-1})
Si	(2-3 μgL^{-1})
Na^+	(257.49-276.8 mg/L)
K^+	(958.73-994.704 mg/L)
Cr^{3+}	(0.8-3.6 μgL^{-1})

Cupric ion (Cu^{2+}), Zinc ion (Zn^{2+}), Magnesium ion (Mg^{2+}), Calcium ion (Ca^{2+}), Aluminium ion (Al^{3+}), Strontium ion (Sr^{2+}), Manganous ion (Mn^{2+}), Ferrous ion (Fe^{2+}), Ferric ion (Fe^{3+}), Silicone ion (Si), Sodium ion (Na^+), Potassium ion (K^+), Chromic ion (Cr^{3+}).

Sodium ion (Na^+), Mg^{2+}, Zn^{2+} and K^+ are trace elements that are abundantly found in saliva and also in human dental enamel.[34]

Orally Administered Formulations

Zn^{2+}, F^- and Sr^{2+} containing mouthwashes and toothpastes are commercially available which are the possible sources leading to ionic substitutions in dental enamel.

Oral Appliances

Dental prosthesis such as cast partial denture could be a possible source of trace elements in patients with missing teeth. As in the fabrication of prosthesis, the alloys that are commonly used include Co^{2+}, Cr^{3+}, Fe^{2+}/Fe^{3+}, Ni^{2+} and Ti^{4+}.[35] Therefore the saliva of patient with cast partial dentures has higher concentration of Cr^{3+}, Ni^{2+}, Co^{2+}, and Fe^{2+}/Fe^{3+} metals than that of a normal individual having natural dentition without any artificial denture.

Porcelain which is used for making dental prosthesis comprises glass matrix and leucite crystalline phase, the later consisting of various elements particularly Si (57-66%), Ba^{2+} (15-25%), Al^{3+} (7-15%), Na^+(7-12%), K^+(7-15%) and lithium ion (Li^+)(0.5-3%). Thus, this could be a possible reason for the increasing content of trace elements in enamel and saliva.

Clinical Implications

Trace elements can enter the structure of tooth enamel and affect its physico-chemical properties. Ghadimi reported that there are several sources for trace elements that get incorporated into enamel structure post-eruptively.[5] These include saliva, dental prosthesis or dental porcelain. Future studies will have to be performed to define the effect of saliva and dental prosthesis on tooth enamel structure.

Metallic parts of the oral prosthesis usually comprise Co^{2+}, Ni^{2+} and Cr^{3+}. The use of these metals can lead to sensitivity and allergy, therefore titanium dentures were introduced in dentistry. It has been reported that titanium is beneficial for the tooth enamel by making them whiter and harder.[5] Thus, it can be suggested that the use of titanium containing materials or appliances should be promoted to be used in dentistry.

Conclusion

The existence of trace elements in the enamel influences the properties of human dental enamel. The Ti^{4+} concentration in enamel is related to the lightness, hardness and the crystal domain size along the c-axis of the enamel. Iron has a negative relation with the type Acarbonate, whereas Co^{2+} and Ni^{2+} have a direct relation in forming type B carbonate. Al^{3+} concentration is inversely related to the enamel cracks. Se^{4+} has positive connection with the a-axis and c-axis of the lattice parameters whereas Ni^{2+} and Cr^{3+} have negative association with the c-axis. Pb^{2+} and Mn^{2+} have positive relation with the c-axis of the crystal domain size in dental enamel. The F-increases the resistance against acid attack and caries more potently compared to other trace elements. The use of various elements other than Zn^{2+}, Cr^{3+}, Co^{2+}, Ti^{4+}, Cu^{2+} and F^- is not promoted due to their mostly negative effect on either the crystal lattice or other physico-chemical properties.

Disclaimer: None.

Conflict of Interest: None.

Source of Funding: None.

References

1. Qamar Z, Anderson P, Hill R. Poly Gamma Glutamic Acid: Effect On Inhibiting Dissolution Of Hydroxyapatite And Enamel. Lambert Academic Publishing, 2012.
2. Berkovitz Ham, Holland GR. Oral Anatomy histology and embryology2011.
3. Duck FA. Physical properties of tissues: a comprehensive reference book. Academic press, 2013.
4. Eimar H, Ghadimi E, Marelli B, Vali H, Nazhat SN, Amin WM, et al. Regulation of enamel hardness by its crystallographic dimensions. ActaBiomater. 2012; 8:3400-10.
5. Ghadimi E, Eimar H, Marelli B, Nazhat SN, Asgharian M, Vali H, et al. Trace elements can influence the physical properties of tooth enamel. Springerplus. 2013; 2:499.
6. Ergun C. Effect of Ti ion substitution on the structure of hydroxylapatite. J Euro Cer Soc. 2008; 28:2137-49.
7. Mabilleau G, Filmon R, Petrov P, Baslé M, Sabokbar A, Chappard D. Cobalt, chromium and nickel affect hydroxyapatite crystal growth in vitro. Acta biomaterialia. 2010; 6:1555-60.
8. Curzon M, Crocker D. Relationships of trace elements in human tooth enamel to dental caries. Arch Oral Biol. 1978; 23:647-53.
9. Kampa M, Castanas E. Human health effects of air pollution. Environ Pollut. 2008; 151:362-7.
10. Malczewska?Toth B. Phosphorus, Selenium, Tellurium, and Sulfur. Patty's Toxicology. 2012.
11. Monteil-Rivera F, Fedoroff M, Jeanjean J, Minel L, Barthes MG, Dumonceau J. Sorption of selenite (SeO 3 2?) on hydroxyapatite: an exchange process. J Colloid Interface Sci. 2000; 221:291-300.
12. Ma J, Wang Y, Zhou L, Zhang S. Preparation and characterization of selenite substituted hydroxyapatite. Mater SciEng C Mater Biol Appl. 2013; 33:440-5.
13. Zhang J, Wang D, Zhou J, Yao A, Huang W. Precise adsorption behavior and mechanism of Ni (II) ions on nano-hydroxyapatite. Water Envir Res. 2010; 82:2279-84.
14. Elkabouss K, Kacimi M, Ziyad M, Ammar S, Bozon-Verduraz F. Cobalt-exchanged hydroxyapatite catalysts: Magnetic studies, spectroscopic investigations, performance in 2-butanol and ethane oxidative dehydrogenations. J Catal. 2004; 226:16-24.
15. Fleet ME, Liu X. Coupled substitution of type A and B carbonate in sodium-bearing apatite. Biomaterials. 2007; 28:916-26.
16. Mavropoulos E, Rossi AM, Costa AM, Perez CAC, Moreira JC, Saldanha M. Studies on the mechanisms of lead immobilization by hydroxyapatite. Envir sci& tech. 2002; 36:1625-9.
17. Li Y, Widodo J, Lim S, Ooi C. Synthesis and cytocompatibility of manganese (II) and iron (III) substituted hydroxyapatite nanoparticles. J Mat Sci. 2012; 47:754-63.
18. Eanes. Enamel apatite. Chemistry, structure and properties. J Dent Res. 1979;58:829.
19. Frazier PD, Little MF, Casciani FS. X-ray diffraction analysis of human enamel containing different amounts of fluoride. Arch Oral Biol. 1967; 12:35-42.
20. Bowen W. Effect of Selenium and Vanadium on Caries Activity in Monkeys (Macaca irus). J Dental Research 1972;51:1285.
21. Muhler JC, Shafer WG. The Effect of Selenium on the Incidence of Dental Caries in Rats. J Dent Res. 1957; 36:895-6.
22. Rodrigues LK, Cury JA, Nobre dos Santos M. The effect of gamma radiation on enamel hardness and its resistance to demineralization.J Oral Sci. 2004; 46:215-20.
23. Gerlach RF, Cury JA, Krug FJ, Line SRP. Effect of lead on dental enamel formation. Toxicology. 2002; 175:27-34.
24. Brudevold F, Aasenden R, Srinivasian BN, Bakhos Y. Lead in enamel and saliva, dental caries and the use of enamel biopsies for measuring past exposure to lead. J Dent Res. 1977; 56:1165-71.
25. Brunette DM, Tengvall P, Textor M, Thomsen P. Titanium in medicine: material science, surface science, engineering, biological responses and medical applications. Springer Science & Business Media, 2012.
26. Buzalaf MA, de MoraesItaliani F, Kato MT, Martinhon CC, Magalhães AC. Effect of iron on inhibition of acid demineralisation of bovine dental enamel in vitro. Arch Oral Biol. 2006; 51:844-8.
27. Torell P. Iron and dental caries. Swed Dent J. 1988; 12:113-24.
28. Putt MS, Kleber CJ. Dissolution Studies of Human Enamel Treated with Aluminum Solutions. J Dent Res. 1985; 64:437-40.
29. Fatima T, Rahim ZB, Lin CW, Qamar Z. Zinc: A precious trace element for oral health care?J Pak Med Assoc. 2016;66:1019-23.
30. Navia J. Prevention of dental caries: agents which increase tooth resistance to dental caries. Inter Dent J. 1972; 22:427.
31. Cardenas JM, Cantu FJG, Rodriguez RO, Murga HM, Gutierrez FO, Preciado RM, et al. Concentration and distribution of trace elements in dental enamel using the energy-dispersive X-ray spectroscopy technique. EurSci J. 2014; 10.
32. Neville BW, Damm DD, Chi AC, Allen CM. In: Neville BW.Oral and maxillofacial pathology, Elsevier: Saunders, 2015.
33. Weir A, Westerhoff P, Fabricius L, Hristovski K, von Goetz N. Titanium dioxide nanoparticles in food and personal care products. Environ Sci Technol. 2012; 46:2242-50.
34. Borella P, Fantuzzi G, Aggazzotti G. Trace elements in saliva and dental caries in young adults. Science of the total environment. 1994;153:219-24.
35. Anusavice KJ, Shen C, Rawls HR. In: Anusavice KJ, Shen C. Phillips Science of dental materials.ElsevierSaunders,2012; pp 592.