Biocompatibility In Orthodontics

Amoli Singh[1], Nidhin Philip Jose[2], Siddarth Shetty[3], Shravan Shetty[4], Ashith M.V[5]

Dr. Amoli Singh, B.D.S[1], Dr. Nidhin Philip Jose, M.D.S[2], Dr. Siddharth Shetty M.D.S[3], Dr. Shravan Shetty[4], Dr. Ashith MV[5]

[1]Post graduate student, Department of orthodontics and Dentofacial Orthopaedics, Manipal College of Dental Science, Mangalore Affiliated to Manipal Academy of Higher Education Pin code- 575001
[2]Associate Professor, Department of Orthodontics and Dentofacial Orthopaedics, Manipal College of Dental Science, Mangalore Affiliated to Manipal Academy of Higher Education Pin code- 575001
[3]Professor, Department of Orthodontics and Dentofacial Orthopaedics, Manipal College of Dental Science, Mangalore Affiliated to Manipal Academy of Higher Education Pin code- 575001
[4]Assistant Professor, Department of Orthodontics and Dentofacial Orthopaedics Manipal College of Dental Science, Mangalore Affiliated to Manipal Academy of Higher Education Pin code- 575001
[5]Associate Professor, Department of Orthodontics and Dentofacial Orthopaedics, Manipal College of Dental Science, Mangalore, Affiliated to Manipal Academy of Higher Education Pin code- 575001

Email: [1]amolisngh@gmail.com, [2]nidhin.philip@manipal.edu, [3]shravan.shetty@manipal.edu, [5]ashith.mv@manipal.edu

Contribution Details:

	Contributor 1	Contributor 2	Contributor 3	Contributor 4	
Concepts	✓		✓		
Design	✓	✓			
Definition of intellectual content	✓				
Literature search	✓	✓		✓	
Clinical studies					
Experimental studies					
Data acquisition	✓	✓			
Data analysis				✓	

Statistical analysis				✓	
Manuscript preparation	✓				
Manuscript editing	✓		✓		
Manuscript review		✓	✓		
Guarantor					

Abstract: This review aims to summarise the properties, interactions, biocompatibility and associated adverse reactions of the commonly used orthodontic materials in clinical practice.

The recent years have seen the introduction of a variety of new orthodontic products necessitating the need for an all-inclusive training in the dental materials field. It is essential for orthodontists to have the basic knowledge about composition of these materials, their structure and properties. Clinically the interactions of orthodontic materials with the hard tissues in the oral cavity and with other dental materials can lead to the issue cytotoxicity and allergic reactions for both patients and doctors. The background about these interactions will allow the orthodontist to make correct material selections which will provide for adequate treatment mechanics and simultaneously equip him/her with the knowledge to deal with the complex effects of the orthodontic materials in the oral environment

Several electronic databases were searched for relevant research and material regarding biocompatibility and interactions of the commonly used orthodontic materials in clinical practice.The Directory of Open Access Journals (DOAJ), meta Register of Controlled Trials, WHO, Digital Dissertations and Google Scholar were used to look for the relevant data. The references and citations of already existing reviews and articles on the issue of biocompatibilty were also searched.

This review is mainly done to attain better knowledge about the biocompatibility of different materials that are used in the orthodontic practice and their associated effects.

Key-words: Biocompatibility, cytotoxicity, allergic reactions

Key Messages: This review summarises the interactions, biocompatibility and associated adverse reactions of the commonly used orthodontic materials. It will allow the orthodontist to make correct material selections to provide adequate treatment mechanics and simultaneously equip him with the knowledge to deal with the complex effects of these materials.

1. INTRODUCTION:

There is no bio-material which is truly inert. Clinically the interactions of orthodontic materials with the hard tissues in the oral cavity and with other dental materials can lead to the issue cytotoxicity and allergic reactions for both patients and doctors. The background of interactions will allow the orthodontist to make correct material selections which will provide for adequate treatment mechanics and simultaneously equip him with the knowledge to deal with the complex effects of these materials in the oral environment.[1-2]

This review aims at summarising the properties, interactions, biocompatibility and associated adverse reactions of the clinically used orthodontic materials.

Methodology:

Several electronic databases were searched for relevant research and material regarding biocompatibility and interactions of the commonly used orthodontic materials in clinical practice.The Directory of Open Access Journals (DOAJ), meta Register of Controlled Trials, WHO, Digital Dissertations and Google Scholar were used to look for the relevant data. The references and citations of already existing reviews and articles on the issue of biocompatibilty were also searched.

Latex products:

Latex gloves, elastics and synthetic elastomers are commonly used materials in orthodontic. Latex or natural rubber is a known allergen in dental practice. Due to the mucosal contact during dental treatment the allergic response seen is exaggerated in the already sensitized patients when compared to direct skin contact. This puts the dental patients in a special risk category.[3] The clinical symptoms of latex hypersensitivity includes urticaria, rhino-conjunctivitis, asthmatic reactions, and, in some severe cases, anaphylaxis is also seen. In cases of acute reactions the facial region especially the lips and the mouth are affected. Patients complain of itchy skin and develop a 'nettle rash' appearance. Studies done on dental under graduate students in Canada and Germany revealed that there was an increased incidence of Type 1 hypersensitivity to latex gloves seen in these students over the course of their graduate programme.[4,5]

Nitrile gloves have been advocated as an replacement for latex. They do not contain NRL proteins but still contact urticaria has been reported by nitrile gloves.[6,7] The presence of benzothiazoles has been attributed to the hypersensitive reaction.

Corn-starch is used in the powder of NRL gloves which is known to be involved in causing glove allergy.[8]

Non latex or polyurethane orthodontic elastics can be used as a replacement in patients with latex allergy. Hwang (AJODO 2003) compared mechanical properties of latex and orthodontic elastics made of silicone and assessed their cytotoxic properties. Increased force decay was seen in silicone bands. The silicone bands showed less cytotoxicity than latex. Santos et al. (2010) compared the cytotoxicity between latex and non-latex elastomeric ligatures used in orthodontics. Less cell lysis was seen in non-latex ligatures when compared to polyurethane and latex ligatures.

It is not possible to completely eliminate the use of latex from dental environment. The aim therefore is to reduce the exposure to latex as far as reasonable as possible. Latex free gloves must be worn and powdered gloves should never be used. Synthetic latex free gloves are readily available. These gloves are made from nitrile, polycholoprene, elastyren, and vinyl. In patients with latex allergy, latex free elastics should be used.

Resin based composites:

The primary of usage of resin based composites in orthodontics is seen for bonding of orthodontic brackets. They are complex mixtures containing many substances. These polymers are not inert materials and may release several components when introduced in the oral cavity. Commonly used monomers in adhesive resins are:

- Bis-GMA(Bisphenol A diglycidyl dimethacrylate)
- TEGDMA(Triethylene glycol dimethacrylate).

Release of these methacrylate monomers may continue even one year after polymerisation.[9]

The degree of curing or the polymerisation efficiency is an important property of polymers. It plays a pivotal role in modulating the physical, the mechanical and the biological properties of materials. Biologically reactive elements such as monomers and additives can be released from a weak polymer network. It is predisposed to absorption of water leading to swelling and hydrolytic degradation. This contributes to poor mechanical properties.[10,11] Monomer degradation can also lead to release of metabolic by-products such as TEG (Triethylene glycol), MA(Methacrylic acid, 2,3-EMA (2,3-epoxymethacrylic acid) and formaldehyde all of which can produce toxic effects.[12] Formaldehyde can lead to cytotoxicity and mutagenicity. It also has carcinogenic and pro-allergenic potential.[13]

Composite resins should ideally show minimal polymerization shrinkage and an optimal level of conversion.[14] Bonding failures can occur due to the presence of residual monomers. They can show adverse effects like allergic reactions, cytotoxicity, mutagenicity, and estrogenicity.[15] Studies done on dental composite resins showed that with increased conversion rate of the monomers, there was a reduction in the cellular toxicity. [16]

In light cured monomers the amount of polymerisation is dependent on many variables, such as exposure time, concentration of photo-initiator, the intensity at which the light emitted by the curing unit, and the filler volume.[17] Light cured adhesives have shown to decrease the oxygen inhibition during polymerisation, extending the working time and shorter polymerisation reaction. An added advantage of these adhesives is less formation of formaldehyde when compared to the two-phase systems.

A better peripheral sealing around brackets is obtained with the use of light cured resins as opposed to self-cured systems. Studies done on gingival fibroblasts showed no acute cytotoxicity in either of the groups.[17]

Bisphenol-A (BPA) is a synthetic compound which is used in the process of manufacture of certain resin monomer systems used in orthodontics. The release of this compound in the oral environment has generated interest in orthodontics.[18] Some hormone related effects of BPA have been shown. It has been demonstrated that when a light cured bracket adhesive was used for bonded lingual retainers the release of BPA continued up to one month after the retainer was delivered.[19]

The following recommendations have been made regarding use of these materials in clinics:

• The light-cure tip should be placed as close as possible to the adhesive.

• Before starting the bonding procedure teeth surfaces should be polished with pumice to decrease the potential for release of BPA.

• Direct irradiation (through the bracket) should be avoided and curing should be done around the bracket edges.

• Patients should be advised to rinse their mouths within the first hour after completion of bonding. This is done to prevent hazards from leaching monomers.[20,21]

• Adhesives designed especially for bonded lingual retainers should be used.

Recently, because of these issues many orthodontic adhesives which are free of BPA have come in the market. These adhesives are based on aliphatic dimethacrylates. An experimental BPA-free dental resin based composite adhesive was developed for fixed retainer bonding. Laboratory comparison between this adhesive and adhesives which contain BPA, showed that the BPA-free resin based adhesive can be a better alternative in clinical practice.[22]

Glass ionomer cements:

GICs are primarily used for bonding of molar bands as the luting cement and for giving posterior disoccluding bite blocks frequently used during orthodontic therapy.

A number of studies have been done to assess the cytotoxic behaviour glass ionomer cements.[23-25] The cytotoxic behaviour of cements depend on their setting conditions.. Non-set materials exhibit high cytotoxicity, whereas set specimens have low to no cytotoxicity.

A number of studies have shown that toxicity varies among products. The cytotoxicity of freshly mixed glass ionomeric cements is related to the acidity and release of fluoride and other ions. including aluminium. The leached aluminium ions, however, were too low to cause cytotoxicity. [26,27] Air humidity has also been associated with toxicity. Incomplete setting of

GIC occurs in presence of low air humidity.(Values < 60%). This can in turn lead to cytotoxic reactions.[28,29] Resin-modified GICs are responsible for causing a variety of reactions, which can be toxic especially in the uncured state. After setting these products have shown decrease in the levels of toxicity.[30] According to certain cytotoxicity studies, inflammatory reactions were seen in rats soon after conventional GIC was implanted subcutaneously but after a longer duration of time the inflammation subsided.[31-33] Another implantation study showed that more pronounced tissue reactions were seen when a thin mix of GIC was used as compared to thicker mixes.[34]

Alloys and implants:

Alloys are commonly used in dentistry. They are materials that are in contact with the oral tissues for prolonged time. Hence it is of utmost importance to understand and investigate the biocompatibility of, especially in the field of orthodontics where the entire system is metallic. Several studies that have been done on dental and orthopaedic alloys have shown that release of elements into the adjacent tissues from these implanted materials occurs.[35] Among the different types of alloys used in orthodontics, titanium alloys usually release relatively less amount of elements into the neighbouring tissues. Whereas, a larger amount of elements are released cobalt and nickel based alloys, or stainless especially over prolonged periods of time. These elements can be easily can be identified in the neighbouring gingival or mucosal tissues as well as the saliva.[36]

Titanium and gold when added to alloys are known to increase the corrosion resistance. But high concentrated fluoride solutions can increase the corrosion of titanium.[37,38]

Alloys which contain less gold showed similar corrosion resistance as alloys with higher content of gold in electrochemical tests. But the corrosion resistance of alloys with low gold content was reduced in immersion tests.[39]

Alloys made up of Cobalt-Chromium show better corrosion resistance to Nickel alloys. Beryllium is known to reduce the corrosion stability of Nickel alloys. Hence beryllium should be avoided as much as possible in alloys.[40]

Element release from alloys is closely related to the pH of the environment. An acidic pH will accelerate the release rate of elements from a metallic alloy. This is frequently seen in alloys made up of Nickel. Clinically the plaque that is formed around these alloys in the oral cavity, produces an acidic environment and can lead to increased release rate of elements.

Stainless Steel alloys: They contain chromium to provide excellent corrosion resistance. Chromium forms thin, adherent, passive surface oxide layer. The chromium oxide surface layer passivate and re-passivate in air. Oxygen is necessary for formation of film. Whereas presence of chloride ions and a low pH can be detrimental to its stability. The chromium oxide passive film are not as stable as their titanium oxide counterparts in titanium containing alloys, contributing to inferior corrosion resistance of stainless steel relative to titanium alloys.

Silver solder: It consists of metallic ions such as cadmium, copper, silver, and zinc. According to the International Register of Potentially toxic Chemicals of the United Nations Environment program, these ions may possess a potential for harmful effect.

Toxicity studies have shown that the silver solder which is used in orthodontics can exhibit severe toxicity of cells and can have inhibiting effects on the growing and proliferating cells that are being analysed. A study was done to check if toxic metallic ions were released from the silver solder into saliva. The results showed that a substantial amount metallic ions such as cadmium, copper, zinc, and silver ions were released into the saliva.

Nickel Titanium Alloys**:** Corrosion resistance of Nickel Titanium wires is primarily due to titanium. Titanium forms several oxides, of which titanium oxide is the most commonly formed and is most stable. Titanium oxide is much more stable and hence more corrosion resistant than chromium oxide.

Intra oral aging of orthodontic alloys: Orthodontic alloys get corroded intraorally when the appliances made from these alloys are placed in the oral cavity for prolonged duration of time. Different types of corrosion is seen in the alloys. Pitting corrosion occurs in brackets and wires. Crevice corrosion is seen on the surfaces that are exposed to corrosive environment, commonly in the region where elastomeric ligatures are applied. Another reason could be the decrease in oxygen levels because of the plaque formation in that area and other by-products of the microbial flora. The third type of corrosion seen is fretting corrosion. This is seen when a metallic wire slides on the bracket slot. It occurs because of the cold welding mechanism acting at the bracket wire interface under pressure.

Action of microbiota colonization: Some bacterial species can uptake metallic elements from alloys and metabolize them. Other species like Bacteroides corrodens, bacillinum ferrroxidans and acid producing Streptococcus mutans adversely affects the surface characteristics of dental alloys. Bacterial metabolism particularly bacteria that reduces sulphates and nitrates which are known to be aggressive, and have effect on the corrosion process of various alloys .

Fatigue fracture: Another important process for aging of orthodontic alloys is tendency of metals to undergo fatigue fracture under repeated cyclic stressing. This process is accelerated by exposing the alloy to a corrosive medium like saliva and appears frequently in wires which are under tension for a prolonged duration of time in the oral environment. For example, archwires engaged to brackets bonded to crowded teeth, and fracture incidents of headgear facebows especially innerbows.

In a study done by Eliades et al, the elements released from orthodontic brackets made of stainless steel and NiTi wires were characterized. They also assessed the cytotoxicity of the ions released from these alloys. It was seen that for the stainless steel group, measurable amounts nickel and low concentration of chromium was detected. Whereas for the Nickel-Titanium group no substances were released. No effect was seen on the survival and DNA synthesis of the culture cells in both the groups. Huang et al conducted a study where they assessed the release of metallic ions in artificial saliva at different pH levels over an immersion period of 12 months from two group of brackets: New and Recycled. It was seen that there was more metallic ion release in the recycled brackets groups when compared to the new brackets. The amount of ion release increased with time. At the end of the 12 week immersion period, the amount of total ion release when averaged was within the recommended daily intake.

Biological effects of nickel: Major corrosion products of stainless steel are iron, nickel and chromium which are reported as potential for producing allergic, toxic, or carcinogenic effects.[41] Nickel ions may affect chemotaxis of leukocytes, they inhibit calcium mediated contractile activity in neutrophils. Nickel induces T lymphocytes to produce several cytokines, including interferon, interleukin IL-2, IL-5, and IL-10, and stimulates cellular proliferation.

Nickel appliances can induce an immune response, a type IV hypersensitivity reaction which manifests in the form of contact dermatitis. One of the most common causes of contact dermatitis is nickel allergy. It’s prevalence is more in women . Nickel alone created more number of cases of contact dermatitis than all other metals combined. Contact dermatitis can occur after coming in direct contact of nickel containing materials such as jewellery, watches, etc.

Clinical features of Nickel allergy generally include contact dermatitis that appears as eczema clinically. In some cases the mucous membrane reactions, such as stomatitis are also seen. Gingival hyperplasias, cheilitis, labial desquamation, and multiform erythemas are frequently observed. Another common symptom is a burning sensation. In chronic cases, the affected mucosa is stays in contact with the causal agent , that is Nickel, for a longer duration of time and can appear from erythematous or hyperkeratotic to ulcerated. Other symptoms can also be present, such as perioral dermatitis and, in some cases orolingual paresthesia is also observed.

Diagnosis of nickel allergy causing contact stomatitis is more difficult in the oral mucosa than on the skin. These lesions can be confused with mechanical injury, autoimmune lesions, aphthous stomatitis, or poor oral hygiene.

Several studies have shown that in a general populations nickel allergy affects 10%-30% of females and 1-3% of males. Incidence of adverse patient reactions in orthodontic practice is approximately 0.3%-0.4%. People with cutaneous piercing were considered a significant risk factor for Ni allergy.[42,43]

Orthodontic patients who show mild symptoms of nickel allergic contact stomatitis, the appliances should be removed immediately. Patients who exhibit more severe reactions should also be treated with antihistamines, anesthetics, or topical corticoids in addition to removing the appliance.

Kolokitha OE et al.(2008) conducted a study to assess the effects of orthodontic treatment on the prevalence of nickel hypersensitivity and compare it with the prevalence in the general population. The results of study showed that orthodontic therapy has no association with an increase in the prevalence of nickel hypersensitivity unless subjects have a history of cutaneous piercing.

Marigo et al (AJODO 2003) conducted a study on 35 patients undergoing orthodontic treatment. These patients were divided into 2 groups: the nickel sensitive and non–nickel sensitive groups. Lymphoproliferation assay was used in vitro to diagnose nickel sensitivity. The results showed that exposure to nickel alloy castings for more than 24 months resulted in lower cell proliferation indices, suggesting that the development of oral tolerance mechanisms might play a role in modulating the cellular response to nickel.

Conclusion:

Biocompatibility is a dynamic and rapidly evolving theory and depends on the communication between the host, the material itself and the expected functioning of the material. The recent years have seen the introduction of a variety of new orthodontic products necessitating the need for an all-inclusive training in the field of dental materials. It is important for the orthodontist to have the basic knowledge about composition of these materials, their structure and properties. One of the main concerns of any dentist is to make sure that the patient is not harmed in any way. This becomes especially important in orthodontics where majority of the patients are children or young adults. Harmful reactions can occur among the various types of materials that are commonly used in orthodontic clinical practice which include alloys, resin based and other cements. Two major biocompatibility issues faced in today's scenario include corrosion and estrogenicity of dental resins. Biocompatibility of materials is not just necessary for the patient but also pertinent to the practitioner from the view point of health of orthodontic team because of the chronic exposure to these materials. The main area of concern for the dental team in orthodontics seems to be contact with latex products and resin-based materials. As no dental material can be proved 100% safe, the choice of using the material should be based on a balance between risks and benefits and what is best for the patient and the dental team.

2. REFERENCES:

[1] Brantley WA, Eliades T. Orthodontic materials: scientific and clinical aspects. AMERICAN JOURNAL OF ORTHODONTICS AND DENTOFACIAL ORTHOPEDICS. 2001;119(6):672-3.

[2] Goldstein RS, Schnellmann RG. Casarett and Doull's Toxicology–The Basic Science of Poisons.

[3] Burke FJ, Wilson MA, McCord JF. Allergy to latex gloves in clinical practice. Quintessence International. 1995 Dec 1;26(12).

[4] Heese A, Peters KP, Stahl J, Koch HU, Hornstein OP. Incidence and increase in type I allergies to rubber gloves in dental medicine students. Der Hautarzt; Zeitschrift fur Dermatologie, Venerologie, und Verwandte Gebiete. 1995 Jan 1;46(1):15-21.
[5] Tarlo SM, Sussman GL, Holness DL. Latex sensitivity in dental students and staff: a cross-sectional study. Journal of allergy and clinical immunology. 1997 Mar 1;99(3):396-400.
[6] Brehler R. Contact urticaria caused by latex-free nitrile gloves. Contact Dermatitis. 1996;34(4).
[7] Horn HM, Aldridge RD. Contact urticaria due to nitrile gloves. Contact dermatitis. 2003 Sep;49(3):163-4.
[8] Hamann CP, Kick SA. Allergies associated with medical gloves: manufacturing issues. Dermatologic clinics. 1994 Jul 1;12(3):547-59.
[9] Polydorou O, König A, Hellwig E, Kümmerer K. Long-term release of monomers from modern dental-composite materials. European journal of oral sciences. 2009 Feb;117(1):68-75.
[10] Gaintantzopoulou M, Rahiotis C, Eliades G. Molecular characterization of one-step self-etching adhesives placed on dentin and inert substrate. Journal of Adhesive Dentistry. 2008 Feb 1;10(2):83.
[11] Vrochari AD, Eliades G, Hellwig E, Wrbas KT. Curing efficiency of four self-etching, self-adhesive resin cements. Dental Materials. 2009 Sep 1;25(9):1104-8.
[12] Emmler J, Seiss M, Kreppel H, Reichl FX, Hickel R, Kehe K. Cytotoxicity of the dental composite component TEGDMA and selected metabolic by-products in human pulmonary cells. Dental Materials. 2008 Dec 1;24(12):1670-5.
[13] IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, International Agency for Research on Cancer. Wood dust and formaldehyde. World Health Organization; 1995.
[14] Amirouche-Korichi A, Mouzali M, Watts DC. Effects of monomer ratios and highly radiopaque fillers on degree of conversion and shrinkage-strain of dental resin composites. Dental Materials. 2009 Nov 1;25(11):1411-8.
[15] Eliades T, Elidades G, Bradley TG, Watts DC. Degree of cure of orthodontic adhesives with various polymerization initiation modes. The European Journal of Orthodontics. 2000 Aug 1;22(4):395-9.
[16] Caughman WF, Caughman GB, Shiflett RA, Rueggeberg F, Schuster GS. Correlation of cytotoxicity, filler loading and curing time of dental composites. Biomaterials. 1991 Oct 1;12(8):737-40.
[17] Gioka C, Bourauel C, Hiskia A, Kletsas D, Eliades T, Eliades G. Light-cured or chemically cured orthodontic adhesive resins? A selection based on the degree of cure, monomer leaching, and cytotoxicity. American Journal of Orthodontics and Dentofacial Orthopedics. 2005 Apr 1;127(4):413-9.
[18] Eliades T, Hiskia A, Eliades G, Athanasiou AE. Assessment of bisphenol-A release from orthodontic adhesives. American journal of orthodontics and dentofacial orthopedics. 2007 Jan 1;131(1):72-5.
[19] Eliades T, Voutsa D, Sifakakis I, Makou M, Katsaros C. Release of bisphenol-A from a light-cured adhesive bonded to lingual fixed retainers. American journal of orthodontics and dentofacial orthopedics. 2011 Feb 1;139(2):192-5.
[20] Kloukos D, Pandis N, Eliades T. Bisphenol-A and residual monomer leaching from orthodontic adhesive resins and polycarbonate brackets: a systematic review. American journal of orthodontics and dentofacial orthopedics. 2013 Apr 1;143(4):S104-12.

[21] Kloukos D, Sifakakis I, Voutsa D, Doulis I, Eliades G, Katsaros C, Eliades T. BPA qualtitative and quantitative assessment associated with orthodontic bonding in vivo. Dental materials. 2015 Aug 1;31(8):887-94.
[22] Iliadi A, Eliades T, Silikas N, Eliades G. Development and testing of novel bisphenol A-free adhesives for lingual fixed retainer bonding. European journal of orthodontics. 2017 Feb 1;39(1):1-8.
[23] Geurtsen W, Spahl W, Leyhausen G. Residual monomer/additive release and variability in cytotoxicity of light-curing glass-ionomer cements and compomers. Journal of dental research. 1998 Dec;77(12):2012-9.
[24] Müller J, Bruckner G, Kraft E, Hörz W. Reaction of cultured pulp cells to eight different cements based on glass ionomers. Dental Materials. 1990 Jul 1;6(3):172-7.
[25] Schmalz G, Thonemann B, Riedel M, Elderton RJ. Biological and clinical investigations of a glass ionomer base material. Dental Materials. 1994 Sep 1;10(5):304-13.
[26] Kawase T, Suzuki A. Studies on the transmembrane migration of fluoride and its effects on proliferation of L-929 fibroblasts (L cells) in vitro. Archives of oral biology. 1989 Jan 1;34(2):103-7.
[27] Müller J, Hörz W, Bruckner G, Kraft E. An experimental study on the biocompatibility of lining cements based on glass ionomer as compared with calcium hydroxide. Dental Materials. 1990 Jan 1;6(1):35-40.
[28] Schmalz G, Hiller KA, Dörter-Aslan F. New developments in the filter test system for cytotoxicity testing. Journal of Materials Science: Materials in Medicine. 1994 Jan 1;5(1):43-51.
[29] Barry TI, Clinton DJ, Wilson AD. The structure of a glass-io nomer cement and its relationship to the setting process. Journal of Dental Research. 1979 Mar;58(3):1072-9.
[30] Beetke E, Bening G, Sobkowiak EA, Bienengräber V. Zur Frage der Gewebsverträglichkeit von Sanal und Calcinat.[Biocompatibility of Sanal and Calcinat] Dtsch Zahn. Mund-u Kieferheilkd. 1974;62:243-51.
[31] Bauer JG, Al-Rubayi A. Tissue response to direct filling materials. Journal of Prosthetic Dentistry. 1987 Nov 1;58(5):584-9.
[32] Blackman R, Gross M, Seltzer S. An evaluation of the biocompatibility of a glass lonomer-silver cement in rat connective tissue. Journal of Endodontics. 1989 Feb 1;15(2):76-9.
[33] Duque C, de Cássia Negrini T, Sacono NT, Spolidorio DM, de Souza Costa CA, Hebling J. Clinical and microbiological performance of resin-modified glass-ionomer liners after incomplete dentine caries removal. Clinical oral investigations. 2009 Dec 1;13(4):465-71.
[34] Steinbrunner RL, Setcos JC, Kafrawy AH. Connective tissue reactions to glass ionomer cements and resin composites. American journal of dentistry. 1991 Dec;4(6):281.
[35] Wataha JC, Craig RG, Hanks CT. Element release and cytotoxicity of Pd-Cu binary alloys. International Journal of Prosthodontics. 1995 May 1;8(3).
[36] Michel R. Trace-metal analysis in biocompatibility testing. CRC Critical Reviews in Biocompatibility. 1987 Jan 1;3(3):235-317.
[37] Kappert HF. Metallegierungen in der Zahnheilkunde. Zahnarztl Mitt. 1992;7:46-54.
[38] Hosch A, Strietzel R. Corrosion of titanium in synthetic salivas containing thiocyanate, chloride and fluoride. Deutsche Zahnarztliche Zeitschrift. 1994;49(10):767-9.
[39] International Organization for Standardization: ISO 1562 - Casting gold alloys. International Organization for Standardization, Geneva 2004
[40] International Organization for Standardization: ISO 8891 - Dental casting alloys with noble metal content of at least 25% but less than 75%. International Organization for Standardization, Geneva 1998

[41] Wataha JC, Lockwood PE, Frazier KB, Khajotia SS. Effect of toothbrushing on elemental release from dental casting alloys. Journal of Prosthodontics. 1999 Dec;8(4):245-51.
[42] Gölz L, Papageorgiou SN, Jäger A. Nickel hypersensitivity and orthodontic treatment: a systematic review and meta-analysis. Contact Dermatitis. 2015 Jul;73(1):1-4.
[43] Pazzini CA, Marques LS, Ramos-Jorge ML, Júnior GO, Pereira LJ, Paiva SM. Longitudinal assessment of periodontal status in patients with nickel allergy treated with conventional and nickel-free braces. The Angle Orthodontist. 2012 Jul;82(4):653-7.