Comparison of metal release from fixed orthodontic appliances in oral mucosa cells in patients with and without fixed orthodontic appliances- An in vivo study

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Abstract

Introduction: A large variety of metallic alloys are routinely used in dentistry. Orthodontic appliances are made of stainless steel containing Nickel and Chromium. Nickel is added to maintain the steels face - centered cubic structure, and is created when heated at 9120°C or higher. Nickel also increases the strength, ductility, and resistance to general, crevice and erosion corrosion. This study aims to compare the release of Nickel and Cobalt in the oral mucosa cells of patients with and without fixed orthodontic appliance.

Materials and Method: A total of 60 subjects were selected. Test group comprised of 30 Orthodontic patients who had fixed orthodontic appliances in both arches. Control group included 30 subjects without any type of fixed orthodontic appliances or metal restorations in the mouth. The oral mucosal cell sample was collected and the metal content was determined using Atomic Absorption Spectrophotometery.

Results: The Nickel content in mucosa samples of test group was significantly higher than that in the control group. The content of Cobalt in the buccal mucosa cells of the test group was significantly higher in the test groups compared to the control group. There was no significant increase in the content of the Chromium in the buccal mucosa cells of the test group.

Conclusion: To conclude, it was seen that there is significant increase in the concentration of nickel and cobalt in the buccal mucosa in both the groups, but there was no significant change in the chromium concentration between the groups.

Keywords: Orthodontic Appliances, Nickel, Cobalt, Chromium, Concentration, Buccal Mucosa.

Introduction

A large variety of metallic alloys are routinely used in dentistry. Gold was used in orthodontics for fabrication of the accessories until the 1930s and 40s. In 1929, stainless steel was used for the first time to replace gold. Orthodontic bands, brackets and wires are universally made up of austenitic stainless steel (302 or 304) containing approximately 8-12% Nickel and 17-22% Chromium. These elements give stainless steel its strength ductility and corrosion resistance. Nickel-Titanium alloys were introduced for use as orthodontic wires in the 1970s and these alloys introduced another potential source of metallic corrosion products that could result in patient exposure. Nickel is added to maintain the steels face - centered cubic structure, and is created when heated at 9120°C or higher. Nickel also increases the strength, ductility, and resistance to general, crevice and erosion corrosion.

There is little information regarding the corrosion of orthodontic appliances in the oral cavity during treatment. Discoloration of underlying tooth surface during orthodontic treatment has been regarded as the consequence of crevice corrosion of the bracket bases.

It has been observed that the warm and moist condition in the mouth offers an ideal environment for the biodegradation of metals, consequently facilitating the release of metals ions that can cause adverse effects. Biocompatibility is strongly related to ionic release and therefore the public may express concern about possible leakage of metal ions from an orthodontic appliance.

The major corrosion products are ion, Chromium, and Nickel for stainless steel, and Ti and Ni for Nickel-Titanium alloys. Among stainless steel and Nickel-Titanium corrosion products Nickel and Chromium have received the most attention because of their reported adverse effects. Nickel is a known allergen[(1)] with carcinogenic[(2)] and mutagenic effects.[(3)]

However, the cause and effect relationship between intra-oral use of Nickel alloys and carcinogenicity has never been demonstrated.[(4)] Nickel is a component of certain enzyme systems in humans and it is considered an essential trace element. Daily intake of Nickel is estimated to be 100-600 μgm/day.[(5)]

Nickel is one of the most common causes of allergic contact dermatitis, and the incidence of such contact dermatitis is as high as approximately 20-30%.[(6)] Adverse reactions related to Nickel containing orthodontic devices such as arch wires, brackets, and soldered stainless steel face-bows have been reported.[(7)] Surprisingly Nickel sensitivity has been reported to be lower in subjects who have received orthodontic treatment. It seems that treatment with Nickel-containing metallic orthodontic appliances before sensitization to Nickel (ear piercing) may have reduced the frequency of Nickel hypersensitivity[(8)] and patients developed immunologic tolerance over a long period of treatment.[(9)] Allergic response to Nickel-containing alloys is mainly type IV hypersensitivity reaction, cell mediated by T-lymphocytes.[(10)] It has been suggested that long-term exposure to Nickel- containing dental

materials may adversely affect both human monocytes and oral mucosal cells.[11]

As mentioned earlier Nickel is ingested with the intake of foods in the range of 300 to 600 μgm per day. Approximately 10% of the general population exhibits hypersensitivity reactions to Nickel. Peltonen reported that women were 10 times more sensitive to Nickel than men. In 1977, Moffa et al. did a study to determine the intraoral response to Nickel-based alloy by patients known to be allergic to Nickel. All of the sensitized patients reacted positively to Nickel sulfate, and eight showed a positive extra oral response to Nickel alloy. However, after intraoral exposure, only one of the Nickel-sensitized patients showed evidence of allergy to Nickel alloy. Perhaps differences between the skin and the oral cavity may account for the lack of reported mucosal reactions to Nickel. The rapid and complete formation of salivary glycoprotein films may act as diffusion barriers.

The readiness with which Nickel may induce carcinogenicity depends on its solubility in the form in which it involves the tissue. Nickel carbonyl, Nickel subsulfide, and Nickel sulfide have been shown to produce carcinogenesis. As yet, no one has detected or reported a relationship between carcinogenicity in humans and the wearing of Nickel-chrome dental restorations. Nickel accumulates in the skin, central nervous system, lungs, and kidneys.

Chromium in the form of chromate salts in concentration of 885 ppm were sufficient to cause positive patch tests in patients. Chromium powder and hexavalent Chromium compound have produced local sarcomas in rodents.[12]

This study aims to compare the release of Nickel and Cobalt in the oral mucosa cells of patients with and without fixed orthodontic patients.

Materials and Method

Subjects were selected from the patients of Department of Orthodontics, at our institution. A total sample of 60 were selected. Test group comprised of 30 Orthodontic patients who had fixed orthodontic appliances in both arches. Control group included 30 subjects without any type of fixed orthodontic appliances or metal restorations in the mouth.

The exclusion criteria in both groups were:

1. Smoking
2. Pre-existing systemic diseases or medications
3. Associated with oral mucosa changes
4. Intra-oral piercing or metal restorations

The objectives of the study was fully explained and informed consent was obtained.

In total, 20 females and 10 males, from 16-20yrs (mean age 18.2year) agreed to participate in the test group. For the patients in the test group, average period since appliance insertion was 16 months in both the upper and the lower arches at the time of sample collection. Patients wearing different fixed orthodontic appliances were chosen randomly from the department of orthodontics, at our institution. 20 females and 10 males aged 16-20 years formed the control group.

Sample collection: The participants were asked to rinse their mouth for one minute to remove the exfoliated dead cells. Mucosa samples were collected by gentle brushing of the internal part of right and left buccal mucosa with interdental brush (Fig. 1).

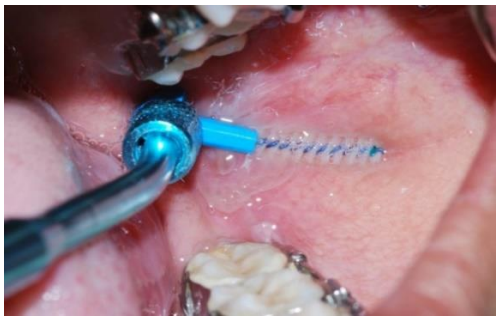

Fig. 1: Sample collection with Interdental brush

The brushes were transferred to polypropylene tubes and stirred in 5 ml of phosphate buffer saline solution (Fig. 2).

Fig. 2: Polypropylene tubes with 5 ml of phosphate buffer saline solution

Metal content determination: Mucosa samples were diluted in water and acidified in nitric acid, kept at 600^{o}C for 10 minutes to dissolve the metal content before analysis. The concentration of Nickel, Chromium and Cobalt ions was quantified using atomic absorption spectrophotometery. (Perkin Elmes Analyst 300 AAS with a graphite furnace). Results were given as ng/ml (Fig. 3).

All metal content determinations were performed at the Indian Institute of Chemical Technology, (Council of Scientific and Industrial Research) Hyderabad, India.

Fig. 3: Atomic absorption spectrophotometery Unit

Statistical analysis: The Student's t-test was applied to assess differences in Nickel, Chromium, and Cobalt mucosa cell contents between orthodontic patients (test group) and the control group. All analyses were carried out using SPSS 12 (Statistical Package for Social sciences: SPSS Inc. Gulbarga, INDIA). Statistical significance was determined at the 0.05 level throughout.

Results

The Nickel content in the buccal mucosa cells was given as mean and standard deviations and are shown in Table 1.

Table 1: Nickel content in the buccal mucosa cells (ng/ml)

Orthodontic treatment	Mean ± SD	t – value	p – value
With braces	9.51 ± 10.82	4.19	p <0.001
Without braces	1.21 ± 0.86		Highly significant

The mean levels of Nickel in control and test group were 1.21 ± 0.86 and 9.51 ± 10.82, respectively.

Examining the content of Nickel in the buccal mucosa cells of orthodontic patient (test group) and controls group the Nickel content in mucosa samples of test group was significantly higher (P<0.001) than that in the controls (Table 1).

When compared the content of Cobalt in the buccal mucosa cells of the test group (Table 2) there was a significant increase in the test groups compared to the control group 1.91±3.06 and 0.5±1.13 respectively.

Table 2: Cobalt content in the buccal mucosa cells (ng/ml)

Orthodontic treatment	Mean ± SD	t – value	p – value
With braces	1.91 ± 3.06	2.25	p <0.05
Without braces	0.57±1.13		Significant

There was no significant increase in the content of the Chromium in the buccal mucosa cells of the test group (Table 3). The mean levels in the control group were 0.64±1.07 whereas in the test group 0.26±0.33, respectively.

Table 3: Chromium content in the buccal mucosa cells (ng/ml)

Orthodontic treatment	Mean ± SD	t – value	p – value
With braces	0.64 ± 1.07	1.86	p <0.05
Without braces	0.26±0.33		Not significant

Discussion

The present study investigated the presence of metal ions in oral mucosa cells in orthodontic patients wearing fixed appliances. Orthodontic appliances are mostly made of stainless steel and Nickel—Titanium alloys. The orthodontic alloy constituents are mostly iron, cobalt, chromium, and nickel. Because the corrosion products from orthodontic appliances can be harmful to the surrounding structure or body, we decided to evaluate the buccal mucosa cell content of three main possibly harmful constituents of orthodontic fixed appliances. Variety of factors can affect the amount of metal released from orthodontic appliances including the corrosion resistance of the material, the brazing or welding effects on the metal, galvanic corrosion of dissimilar metals, the surface of the appliance.

Oral cavity provides an environment that makes aqueous corrosion in metals and alloys more favorable. Saliva as an electrolyte and medium for chemical reactions between metals can cause corrosion. The organic acids and enzymes that microbes produce or the bacteria existing within the mouth can also cause corrosion. The present study used atomic absorption spectrophotometry with a graphite furnace for analysis of metal content in oral tissues. This is a common method used for trace element analysis in the literature.[13]

Metal corrosion may be altered by the use of passivating alloys elements or by coating with another metal. Passivation of steel is obtained by adding 20% Chromium, which forms a surface layer of Chromium oxide.[14]

In our study, the Nickel content in buccal mucosa cells of orthodontic patients (test group) was found to be significantly higher than in controls. This in vivo observation is in line with previous study by Amini et al.[15] in which the presence of Nickel has been shown in oral mucosa cells of orthodontic patients. Contrary to the work of Amini et al.[15] we did not find a significant difference in Chromium cell contents in patients with orthodontic appliances compared with their non-appliance controls. In the groups the amount of metal leached was seen to be more in females then in males. The failure to reach statistical significance was

probably due to the wide variation in metal contents and larger number may be required to demonstrate significant difference. Nickel is known allergen. In the study of Finnish adolescents, the prevalence of Nickel allergy was found to be 30% in girls and 3% in boys. This was thought to be related to sensitization to Nickel by ear piercing as the prevalence in adolescents with ear piercing was 31% and only 2% otherwise.

Nickel is very important as it resists corrosion even at high temperature. Nickel compounds are ubiquitous and are consumed as part of a normal diet from foods such as vegetables, with the daily intake estimated to be 100-600 µg/day. Nickel is component of certain enzyme system is in humans and is considered as essential trace element. Nickel is a known hapten, which can bind to proteins and form complete antigens. However, Nickel has to be released from the alloys to be able to act as a hapten.(14)

Allergic responses are mediated through the immune system. In a sensitized individual, allergic responses can be entailed by relatively small amounts of the allergen, for example, if Nickel ions are released from a Nickel – plated material following direct and prolonged contact with the skin.

The majority of dental allergies including allergic responses to Nickel containing dental alloys, comprise type IV hypersensitivity reactions, cell mediated by T-lymphocytes.

These toxic reactions are dose dependent. The effects primarily depend on the nature of leach ants from the material. Some toxic effects may be initiated by a onetime large dose above threshold or by repeated small doses, provided that the doses are cumulative to above threshold levels. It is not known if this is true for induction or elicitation of Nickel sensitization. Although the dose effect is in disputable toxic reactions, it is important to note that the thresholds for reactions vary from endpoint to endpoint and to some extent from individual to individual. Recent, publications have suggested that 'long-term' exposure to Nickel - containing dental materials may adversely affect both human monocytes and oral mucosal cells.(16)

Data on the prevalence of allergic reactions and positive test results e.g., from skin testing for metal salts are available. The possible causes of oral tissue reactions alleged by related to dental alloys are bacterial adhesion, toxicity, sub toxic effects and allergy.(17)

Nickel is released primarily as a soluble compound, while Chromium is released primarily in an insoluble form with corrosion of the simulated orthodontic appliance.(18)

The general mechanism for the corrosion and subsequent release of metal ions from stainless steel involves the loss of the passivated layer consisting of Chromium oxides and Chromium hydroxide which forms on the surface of stainless steel upon contact with oxygen. A number of factors facilitate the corrosion of stainless steel. Crevice corrosion is an intensive local attack which occurs in shielded areas on a metallic surface. Stainless steel is especially susceptible to this form of corrosion and has been implicated as the mechanism involves in the orthodontic brackets.

Halide ions, especially chloride causes pitting corrosion. Mechanical distortion and excessive cold working promote corrosion by making the distorted portion of the wire or band more anodic. The alloy then behaves electrochemically as if two alloys were present.

The presence of dissimilar metals or alloys such as silver solder amalgams, or gold may lead to galvanic corrosion. Heating between 400^0 and 900^0 C makes stainless steel more susceptible to intergranular corrosion because of loss of Chromium carbide at the grain boundaries.

In the oral cavity such factors as temperature quantity and quality of saliva, plaque pH, protein, physical and chemical properties of food and liquids and general and oral health may influence corrosion by a combination of the mechanism discussed above.

The amount of daily intake of metals from orthodontic appliances over time is a matter of great importance. Metal is released into the oral cavity with saliva as the medium, and this release could be influenced by a high chloride mixture in the saliva or the intake of various organic substances or foods with low pH such as fruit juices and soft drinks. The physical characteristics of saliva are changed according to food intake, health and time of the day. The most important factors in corrosion is the flow rate of saliva.(19) The release of Nickel was seen more due to galvanic corrosion between the brackets and the bands rather than to corrosion of the NiTi arch wire itself, Nickel was released because of the difference in electromotive force (driving force).(5)

Characteristics lesions of contact stomatitis vary from barely visible, mild erythemia to a fiery red color with or without edema, symptoms may include loss of taste, numbness, burning sensation and soreness of the involved area, often accompanied by angular cheilitis. Itching is not a frequent symptom. Although it is more difficult to provoke contact stomatitis than contact dermatitis, severe gingivitis associated with orthodontic therapy may be manifestation not only of poor oral hygiene but also of a contact hypersensitivity reaction to Nickel and/or Chromium ions released during the corrosion of stainless steel.(20)

Nickel can be taken up into cells by diffusion via the Mg^{+2} transport system,(21) or via the calcium and iron channels.(22) The most effective way of Nickel uptake into cells is by phagocytosis of metallic Nickel or Nickel compound dust which has been seen in cultured cells, the efficiency of which depends on the size and surface charge of the Nickel particles.(1) Of the two environmentally available forms of Chromium, bexavalent and trivalent, the hexavalent form has been demonstrated to be associated with the toxic parameters and classified as human, carcinogen and mutagen. Several studies have shown that the cellular uptake of chromate is several fold greater than that of the trivalent

ion, because trivalent Chromium is predominantly octahedral and diffuses slowly. The tetrahedral hexavalent ion has been shown to enter the cell through general anion channels and bind to cellular components, causing disruptions in biochemical pathways.

Reductive metabolism of Chromium within the cell by the cell's redox system leads to the formation of various intermediate forms, Cr (V), Cr (IV), and Cr (III).[23] While there is overwhelming evidence to show that Cr (VI) complexes are mutagenic in bacterial and mammalian cells, most of the Cr (III) complexes are shown to be non-mutagenic. Entry of Cr (III) into cells has also been shown to be diffusion controlled and macrophage mediated, Chromium has been recognized as an essential trace element.[24] Interpretation of the metal content of buccal mucosa cells is hampered by inherent limitations of the atomic absorption spectrophotometry to differentiate between oxidation levels of the metal contents. However, the valence of a metal affects its biologic activity, e.g., being mutagenic, hexavalent Cr crossed the cell membrane in contrast to trivalent Cr during in vitro studies There have been many studies on the amount of metal released from orthodontic appliances under various physical and chemical conditions.[20] These studies demonstrated that these metals were released and absorbed by patients during the early stages of orthodontic therapy. They concluded that Nickel ions, released from orthodontic appliances in saliva or blood samples was significantly below the average dietary intake and did not reach toxic concentration. However, a review of the literature reveals that prolonged in vitro exposure to low levels of Nickel ions can alter cellular metabolic activity, Furthermore studies taking oral mucosal cell brushings in orthodontic patients compared with control subjects concluded that Nickel release from fixed orthodontic appliances could induce DNA damage in oral mucosal cells.[22]

Therefore, to ensure the safety of patients, further research and continued follow up would be needed to determine the long term significance of Nickel release and other corrosion products.

Conclusion

According to the result attained from this study, it has been shown that there is significant increase in the concentration of nickel and cobalt in the buccal mucosa cells of the patients of test group compared to the control group. Although this change was significant clinically for cobalt and nickel, it was seen that this increase in their levels is quite low to cause any carcinogenic or mutagenic effect.

Currently, there is a lack of data linking directly the prevalence of nickel induced side effects in non-sensitized individuals with the insertion of orthodontic materials. It has not been established what is the threshold amount of nickel necessary to elicit skin or oral mucosal reactions. However, further research should be conducted on the tolerance to these metals and the recurrence of metal allergies after continous contact with the oral mucosa cells. More studies are required concerning the amount of metal leached during orthodontic treatment in buccal mucosa cells and their cytotoxic effects.

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