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COMPARATIVE EVALUATION OF METAL IONS RELEASED FROM NICKEL-TITANIUM AND TITANIUM MOLYBDENUM ALLOY ORTHODONTIC ARCH WIRES PROTOCOL FOR IN VITRO STUDY.

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ABSTRACT

INTRODUCTION: Oral environment promotes corrosion of metallic components of orthodontic appliance, which leads to release of metals from metallic component causing relevant health threat.

AIM: This study is planned to evaluate and compare the release of metal ions from NiTi and Titanium Molybdenum Alloy (TMA) Orthodontic arch wire.

MATERIAL AND METHOD: 10 wires each of NiTi and TMA will be incubated in simulated oral environment in artificial saliva for 90 days. After incubation, artificial saliva will be collected and subjected to analytical procedure using Atomic Absorption Spectrophotometer. Data will be subjected to statistical analysis.

RESULTS: The metals released from both orthodontic arch wires will be assessed. As the

nickel content in NiTi orthodontic arch wire is more, release of nickel may be more causing toxic effects if the patient is allergic to Nickel.

CONCLUSION: Metals released from both NiTi and TMA orthodontic arch wires will be accessed and the safer wire will be preferred.

Introduction: Orthodontic components can release trace elements like metals and metalloids which can cause relevant health hazard. Oral environment promotes various factors which lead to corrosion of metallic component of orthodontic appliances. These are saliva pH, temperature fluoride, bacterial flora, enzyme activity, and proteins.[1] Cytotoxic effects result when tissues are exposed to a sufficient concentration of a primary irritant for a sufficient period of time.

Corrosion releases metal ions into the oral cavity that are ingested into the gastrointestinal system. A previous cell culture study found that stainless steel brackets incubated in cell culture medium for 30 days released high concentrations of metal ions, such as titanium, chromium, manganese, nickel, and molybdenum[2].Release of metallic ions can cause adverse local and systemic adverse biological effects on patients' health. Locally, the released ions may adversely affect the oral tissues by inhibiting enzyme or mitochondrial activity and damaging DNA, as has been demonstrated in vitro.[3, 4, 5]

Nickel-titanium (NiTi) wire is one of the most commonly used orthodontic wires clinically due to its good working and mechanical properties. However, nickel-titanium wires may contain in excess of 50% nickel and stainless-steel wires contain 8% nickel. In some individuals Nickel intake can develop a cutaneous reaction known as systemic contact dermatitis (SCD). Apart from SCD, Chronic oral exposure to Nickel can lead to other adverse effect, like Hematotoxicity and Nephrotoxicity, and may induce type IV hypersensitivity.[5, 6, 7]

The use of TMA (beta titanium) wires i increasing its popularity over NiTi wires due to its improved properties. Further there is no documented case of allergies towards TMA reported in the literature. Due consideration regarding release of toxic material before insertion of wires in oral cavity is essential to avoid toxic effects and health risk. Orthodontic appliances used for a prolonged period of time with exposure to various intraoral conditions can lead to toxic effects caused by the release of various elements from components of orthodontic appliances. Hence with this observation in view, this study is planned to evaluate and compare the release of metal ions from NiTi and Titanium Molybdenum Alloy (TMA) orthodontic arch wire.

Primary Research Question: What are the metal ions released from NiTi and TMA orthodontic arch wire?

Primary Hypothesis: There is release of metal ions from NiTi and TMA orthodontic arch wire.

Review of Literature:

N. Staffolani et al. (1999) evaluated release of nickel, chromium, copper, silver and palladium as the microbiological and enzymatic characteristics of the oral cavity would provide a suitable environment for the corrosion of metals. In the study an orthodontic appliance was constructed containing two molar bands, ten brackets and one NiTi memory arch-wire. Corrosion of metals was determined using an atomic absorption spectrophotometer Varian AA 10. The daily release of Ni, Cu and Cr by an orthodontic appliance in acid pH, particularly favourable to corrosion, was found to be well below that ingested with a normal daily diet. It was therefore concluded that the quantities of metal ions released in their experimental conditions should not be cause for concern in utilizing the appliance.[9]

Chung-Ju Hwang et al. (2001) measured metal ions released from the fixed orthodontic appliances. Four simulated fixed orthodontic appliances were fabricated that corresponded to half of the maxillary arch and were soaked in 50 mL of artificial saliva for 3 months. Appliance were divided into four groups. Group A and B both were of Stainless Steel and group C and D were of NiTi. Study concluded that there was decrease in metal ion released as immersion time increased during three months immersion period.[10]

Fiorenzo Faccioni et al. (2003) investigated the biocompatibility of fixed orthodontic appliances.The study evaluated the presence of metal ions in oral mucosa cells, their cytotoxicity, and possible genotoxic effects. The total 5. The biologic effects indicated that both metals induced DNA damage. There were significant positive correlations between cobalt levels and the number of comets and apoptotic cells, nickel levels and number of comet cells, and cobalt levels and comet tails. This study corroborates that nickel and cobalt released from fixed orthodontic appliances can induce DNA damage in oral mucosa cells.[11]

Luciane M. Menezes et al. (2004) evaluated the incidence of hypersensitivity to orthodontic metals. The tested substances were cobalt chloride, copper sulphate, potassium dichromate, iron sulphate, manganese chloride, molybdenum salt, nickel sulphate, and titanium oxide. Total sample size was 38. Statistically significant positive reactions were observed for nickel sulphate, potassium dichromate, and manganese chloride. Reactions to nickel sulphate had the greatest intensity. No differences were observed between the reactions before and after placement of the orthodontic appliances indicating that the materials did not sensitize the patients or affect their tolerance to these metals during the study period. No statistical difference was observed regarding sex for any evaluated substance, although a greater positivity to nickel sulphate was observed among female patients and to potassium dichromate in male patients.[12]

Maja Kuhta et al. (2009) conducted a study, to examine the effects of three different parameters-pH value, type of archwire, and length of immersion on release of metal ions from orthodontic appliances. A total of 18 simulated fixed appliances were used. Results showed that the appliances released measurable quantities of all ions examined. Change in pH had a very strong effect on the release of ions; and the release of ions was dependent on wire composition, but not proportional to the content of metal in the wire. The largest number of ions were released during the first week of appliance immersion. The study concluded that Levels of released ions were sufficient to cause delayed allergic reactions. This must be taken into account when type of arch wire is selected, especially in patients with hypersensitivity or compromised oral hygiene.[13]

The aim of the study conducted by Antonio Jose Ortiz et al was to determine the amounts of metallic ions of stainless steel, nickel free, and titanium alloys release into a culture medium, and to evaluate the cellular viability and DNA damage of cultivated human fibroblasts with those mediums. The metals were extracted from 10 samples. The nickel-free alloy released lower amounts of ions to the medium. The greatest damage in the cellular DNA, measured as the olive moment, was also produced by the stainless -steel alloy followed by the nickel-free alloy. Conversely, the titanium alloy had an increased cellular viability and did not damage the cellular DNA, as compared with the control values. The titanium brackets and tubes were the most biocompatible of the three alloys studied.[2]

Srinivas Kumar Karnam et al. (2012) evaluated the nickel, chromium, copper, cobalt and iron ions released from simulated orthodontic appliance made from new arch-wires and brackets. Sixty sets of new archwire, band material, brackets and ligature wires were prepared simulating fixed orthodontic appliance. Results showed that high levels of nickel ions were released from all four groups, compared to all other ions, followed by release of iron ion levels. There is no significant difference in the levels of all metal ions released in the different groups. They concluded that use of newer brackets and newer arch-wires confirms the negligible release of metal ions from the orthodontic appliance. The measurable amount of metals, released from orthodontic appliances in artificial saliva, was significantly below the average dietary intake and did not reach toxic concentrations.[14]

Marcin Mikulewicz et al. (2012) conducted an in vitro experiment on the release of metal ions from orthodontic appliances composed of alloys containing iron, chromium, nickel, silicon, and molybdenum into artificial saliva. In relation to the maximum acceptable concentrations of metal ions in drinking water and to recommended daily doses, two elements of concern were nickel and chromium. The elevated levels of metals in saliva are thought to occur by corrosion of the chemical elements in the alloys or welding materials. The concentrations of some groups of dissolved elements appear to be interrelated.[15]

Barat Ali Ramazanzadeh et al. (2014) investigated release of nickel ion from three types of nickel-titanium-based wires in the actual received state and after immersion in a simulated oral environment. The single-strand NiTi released the highest quantity of nickel ion in the as-received state and the multi-strand NiTi showed the highest ion release after oral simulation. The quantity of nickel ion released from Damon Copper NiTi was the lowest in both conditions. Oral simulation followed by sterilization did not have a significant influence on nickel ion release from multistrand NiTi and Damon Copper NiTi wires, but single-strand NiTi released statistically lower quantities of nickel ion after oral simulation. The multi-strand nature of the Super-cable did not enhance the potential of corrosion after immersion in the simulated oral environment. In vitro use of nickel-titanium-based arch-wires followed by sterilization did not significantly increase the amount of nickel ion released from these wires.[16]

Rabindra S Nayak et al. (2015) evaluated the release of nickel and chromium ions from orthodontic appliances in the oral cavity using Inductively Coupled Plasma Mass Spectrometer (ICP-MS). The ionic concentration at the end of 10-12 months of treatment showed a statistically significant increase in of 17.92 ppb for chromium and a statistically insignificant decrease in nickel ion concentration by 1.58 ppb. Pearson's correlation coefficient showed a positive correlation for an increase in nickel concentration after aligning, but not at the end of 10-12 months. A positive correlation was seen for an increase in chromium ion concentration at both time intervals. Nickel and chromium ion concentration in saliva even though below the recommended values per day should not be ignored in light of the new knowledge regarding effects of these ions at the molecular level and the allergic potential. Careful and detailed medical history of allergy is essential. Nickel free alternatives should form an essential part of an orthodontist's inventory.[17]

R. S. Senkutvan et al. (2015) analysed and evaluated the rate of Ni ion released from different types of arch wires used in orthodontics. Results showed significant statistical influence on the released amount of Ni and Ti ions. The amount of Ni ions released in all test solutions diminished with time and was below the critical value necessary to induce allergy and below daily dietary intake level. The daily release of NiTi, SS, Cu NiTi and ion implanted

NiTi by an orthodontic appliance in acid pH, particularly favourable to corrosion was well below the amount ingested with a normal daily diet. It is therefore concluded that the quantities of metal ions released in our experimental conditions should not be cause for concern in utilizing the appliance.[18]

A study was conducted by Marcin Mikulewicz et al. (2015) to evaluate metal ion accumulation in hair of patients undergoing orthodontic treatment with fixed appliances. Hair sampling was performed at the beginning and in the 4th, 8th, and 12th month of the treatment. The mean content of Cr was higher than the 90th percentile value for this element. The upper limit of literature reference ranges for Cr, Ni, and Fe in hair were not exceeded. The value of exposure of orthodontic patients to metal ions released from orthodontic appliances can be assessed by hair mineral analysis. The content of Cr was statistically significantly higher during the treatment than before the beginning of therapy.[19]

Huma D. Hussain et al. (2016) conducted a study to evaluate and compare the nickel released from stainless steel and nickel titanium arch-wires in artificial saliva over three months with the use of simulated fixed orthodontic appliances. Significant release of nickel was seen in all groups up to the end of first month. On comparing the nickel release between all groups at each time interval, the results were not significant. The highest amount of nickel was released from nickel titanium arch-wires, however, the quantity of nickel released from both NiTi and stainless- steel arch-wires were not significant. The rate of nickel released was high within the first week and continued up to the first month after which the nickel content was stable in all the groups.[20]

Arash Azizi et al. (2016) evaluated the amount of nickel and titanium ions released from two wires with different shapes and a similar surface area. Wires were separately dipped into polypropylene tubes containing 50 ml of buffer solution and were incubated and maintained at 37 °C. Inductively coupled plasma atomic emission spectrometry (ICP-AES) was used to measure the amount of ions released after exposure lengths of 1 h, 24 h, 1 week, and 3 weeks. The results indicated that the amount of nickel and titanium concentration was significantly higher in the rectangular wire group. The most significant release of all metals was measured after the first hour of immersion. Release of metal ions was influenced by the shape of the wire and increase of time.[21]

Tipanan Yanisarapan et al. (2018) conducted a study. The aim of this study was to determine the cytotoxicity, metal ion release and surface roughness of metal orthodontic appliances after immersion in different fluoride product solutions. The four metal ion levels and surface roughness of the brackets and arch-wires significantly increased, while cell viability significantly decreased, especially in the TMA subgroup. The SEM results showed that the brackets and wires in the APF groups demonstrated more lines and grooves compared with the other groups. Using APF gel during orthodontic treatment with fixed metal appliances should be avoided.[4]

Aneta Olszewska et al. (2020) experimentally tested whether these appliances may contribute to exposure to toxic elements. In this study, elastomeric ligatures were incubated in artificial human saliva for 1 month. For comparison, stainless steel ligatures were incubated for 1, 3, and 6 months under similar conditions. The determined metal levels were compared to the corresponding safety limits for human exposure. The results in vitro study high-light that the use of ligatures in orthodontic treatment can be considered safe in terms of metal exposure although elastic ligatures replaced on a monthly basis appear to be advantageous in comparison to the prolonged use of stainless- steel appliances.[8]

There are many reports and articles referring to allergies, intoxication and other adverse effects due to release of different elements or metal ions from metallic components used in medical science. Limited research is documented with reference to studies relating to metal ions release with the said wires. Thus, an attempt to evaluate and compare metal ions released from NiTi and TMA arch wires is made in this study.

Primary Objectives: To assess release of metal ions from orthodontic NiTi and TMA arch wire.

Other Objectives: To quantify and compare metal ions released from NiTi and TMA Orthodontic arch wires.

Methodology:

STUDY DESIGN: An in vitro experimental study.

STUDY SETTING: The study will be carried out in the Department of Orthodontics and Dentofacial Orthopedics.

STUDY POPULATION: In vitro, NiTi and TMA orthodontic arch wire.

SAMPLE SIZE ESTIMATION:

Level of significance = 5%, Power = 80%, Type of test = two-sided

Formula of calculating sample size is: Sample size for clinical trial (outcome variable on ratio scale and testing null hypothesis

$$N = \frac{2\,S^2(Z1+Z2)^2}{(M1-M2)^2}$$

$$N = \frac{2\,(1.644+0.0841)^2}{1.2} = 20$$

t tests - Means: Difference between two independent means (two groups)

Analysis: A priori: Compute required sample size

Input:

Tail(s) = One

Effect size d = 1.2
α err prob = 0.05
Power (1-β err prob) = 0.8
Allocation ratio N2/N1 = 1
Output:
Non-centrality parameter δ = 2.6832816
Critical t = 1.7340636
Df = 18
Sample size group 1 = 10
Sample size group 2 = 10
Total sample size = 20
Actual power = 0.8252225

A power analysis was established by G*Power,Sversion 3.0.1(Franz Faul universitat , Kiel, Germany). Total minimum sample size of 20 units (10 units per wire type/group; 2 study groups) would yield 80% power to detect significant differences, with effect size of 1.2 and significance level at 0.05.

SAMPLING TECHNIQUE: Purposive sampling
OPERATIONAL DEFINITION:
ATOMIC ABSORPTION SPECTROMETRY (AAS): It detects elements in either liquid or solid
samples through the application of characteristic wavelengths of electromagnetic radiation from a light source. Individual elements will absorb wavelengths differently, and the absorbance is measured against standards.

MATERIALS:
1.Artificial Saliva:
The composition of commercially available artificial saliva is (amount: mg/100ml)
Sodium chloride 0.84
Potassium chloride 1.2
Magnesium chloride 0.052
Calcium chloride 0.146
Potassium di-hydrogen phosphate 0.34
Sorbitol Solution (70%) 60ml
Hydroxyethyl cellulose 3.5
2.17x25 NiTi arch wire. Composition of NiTi arch wire
Nickel (Ni) 55%
Titanium (Ti) 45%
3.17x25 TMA arch wire. Composition of TMA arch wire
Titanium (Ti) 77.8%
Molybdenum (Mo) 11.3%
Zirconium (Zr) 6.6%
Tin (Sn) 4.3%

INCLUSION CRITERIA:
New sterile orthodontic arch wires.
EXCLUSION CRITERIA:
Used wires
Broken wires

DATA ANALYSIS PLAN AND METHOD:
Statistical analysis will be performed using Statistical Package for Social science (SPSS) version
21 for Windows (SPSSInc, Chicago, IL).
Descriptive quantitative data will be expressed in mean and standard deviation respectively.
Data normality will be checked
It is an experimental in vitro study.
0.017x 0.025 NiTi and 0.017x 0.025 TMA (6 inch) prefabricated Orthodontic arch wires of will be selected for study and it will be divided into two groups respectively (n=10)
Group A: NiTi arch wire
Group B: TMA arch wire
These wires will be immersed in 50ml commercially available artificial saliva for 90 days in 10
different glass containers (petri dish) for each group. Each container will have same amount of saliva, which will be incubated in temperature simulating oral environment. All 20 samples
will be coded. After incubation, artificial saliva will be collected and subjected to analytical procedure-Atomic Absorption Spectrophotometer (AAS, Model: Shimadzu AA-6800, Optics: Double beam with background correction, Wavelength range: 190 to 900nm) at Department of Chemistry, University Laboratory to evaluate and quantify the release metal ions in saliva. Data obtained will be subjected to Statistical Analysis.

DATA COLLECTION CHART/RESULTS:

Group A sample	Metal released							
	1	2	3	4	5	6	7	8
1								
2								
3								
4								
5								

6								
7								
8								
9								
10								

Group B sample	Metal released							
	1	2	3	4	5	6	7	8
1								
2								
3								
4								
5								
6								
7								
8								
9								
10								

DISSCUSION: Orthodontic components can release trace elements like metals and metalloids which can cause relevant health hazard. [1] Cytotoxic effect result when tissues are exposed to a sufficient concentration of a primary irritant for a sufficient period of time. Corrosion releases metal ions into the oral cavity that are ingested into the gastrointestinal system. Release of metallic ions can cause adverse local and systemic adverse biological effects on patients' health. Locally, the released ions may adversely affect the oral tissues by inhibiting enzyme or mitochondrial activity and damaging DNA, as has been demonstrated in vitro.[3, 4, 5]

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CONCLUSION: Evaluation and comparison of metal ions released from NiTi and TMA arch wires made in this study will enable the orthodontist in precise selection of arch-wires customised for a given case.

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GANTT CHART:

ACTIVITIES	YEAR 2020-2021						YEAR 2021-2022						YEAR 2022-2023					
	Sept / Oct	Nov / Dec	Jan / Feb	Mar / Apr	May / June	July / Aug	Sept / Oct	Nov / Dec	Jan / Feb	Mar / Apr	May / June	July / Aug	Sept / Oct	Nov / Dec	Jan / Feb	Mar / Apr	May / June	July / Aug
Identification of research problems	■																	
IEC clearance		■																
Formulation and synopsis submission			■															
Synopsis approval				■														
Literature search					■	■	■	■	■	■								
Data Collection							■	■	■	■								
Data analysis and interpretation											■	■						
Results and conclusions												■	■					
Thesis write up and submission													■	■				