

**A COMPARITIVE EVALUATION OF THE RELEASE OF
METAL IONS FROM ROUND AND RECTANGULAR
SUPERELASTIC NITI WIRES OF THREE
DIFFERENT MANUFACTURERS
- AN IN VITRO STUDY**

DISSERTATION

Submitted to The Tamil Nadu Dr. M.G.R Medical University
in partial fulfillment of the requirement for the degree of

MASTER OF DENTAL SURGERY



BRANCH V

ORTHODONTICS AND DENTOFACIAL ORTHOPEDICS

2015 - 2019

CERTIFICATE

This is to certify that the dissertation entitled “**A comparative evaluation of the release of metal ions from round and rectangular superelastic NiTi wires of three different manufacturers - An in vitro study**” is a bonafide record of the work done by **Dr. Chandana L.R.**, a Post graduate student during the period 2015-2019 under my guidance and supervision. This dissertation is submitted in partial fulfilment of the requirements for the award of **Master of Dental Surgery** on Branch V (Orthodontics and Dentofacial Orthopedics) under **The Tamil Nadu Dr. M.G.R Medical University, Guindy, Chennai**. It has not been submitted (partial or full) for the award of any other degree or diploma.

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I hereby declare that this dissertation **“A comparative evaluation of the release of metal ions from round and rectangular superelastic NiTi wires of three different manufacturers - An in vitro study”** is a bonafide record of work undertaken by me during the period 2015-2019 as a part of post graduate study. This dissertation, either in partial or in full, has not been submitted earlier for the award of any degree, diploma, fellowship or similar title of recognition.

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6	0.016 3M WIRES
7	19 X 25 3M WIRES
8	GROUP IA
9	GROUP IB
10	GROUP IIA
11	GROUP IIB
12	GROUP IIIA
13	GROUP IIIB
14	ICP-MS
15	SAMPLE INTRODUCTION SYSTEM
16	INCUBATOR
17	DIGESTER
19	SHAKER

LIST OF ABBREVIATIONS

NiTi	Nickel Titanium
Ni	Nickel
Ti	Titanium
Mn	Manganese
Cr	Chromium
Co	Cobalt
NiO ₂	Nickel Oxide
TiO ₂	Titanium Oxide
NaF	Sodium Fluoride
TMA	Titanium Molybdenum Alloy
TiAl ₆ V ₄	Titanium Alloy
ICP MS	Inductively Coupled Plasma Mass Spectrometry

ABSTRACT

ABSTRACT

Nickel titanium wires are the routinely using archwires in orthodontics. The two wires which are used in the initial and final stages are round and rectangular wires respectively. Biocompatibility of any dental material is now a fundamental requirement of successful clinical behavior in oral cavity. NiTi is a universal wire, Nickel in NiTi is capable of eliciting toxic and allergic responses and can produce more allergic reactions than any other metal elements. So, nickel titanium orthodontic arch wire with a good properties including corrosion resistance is essential to its biocompatibility. The metal ions leaching from orthodontic wires cannot be fully evaded; but it is possible to use materials with lower amounts of ions leaching in the mouth. It was proven that the amount of ions, leached from orthodontic wires in saliva was less than the toxic concentrations, which is below the critical value necessary to induce allergy and less than the daily dietary intake levels. The purpose of this study is to determine the amount of Ni and Ti ion release from NiTi wires of three different manufactures and to check whether the leached metal ions is lower than the daily dietary intake.

Methods

The study was performed by immersion of the samples in artificial saliva at various time intervals and Ni and Ti release was quantified with the use of an inductively coupled plasma mass spectrometry. In this study superelastic NiTi Archwires of three different manufactures which is in two shapes Round and Rectangular of commonly using dimensions 0.016 and 19x25 respectively of 7 inches long are used in this study. The testing solution used in the study is artificial saliva buffer solutions.

Procedure

Each wires separately dipped into 126 polypropylene beakers containing 50 ml of buffer solution. Then incubate and quantify the ions leached at T1=1 hour, T2=24 hour, T3=1 week, T4=3 week using the software Thermoscientific Qtegra™ Intelligent Scientific Data Solution Software (ISDS) which is attached to the ICP MS instrument. This shared software approach provides control and data processing for a range of elemental and isotopic analysis technologies The output is numerical, and provided in counts per second i.e., how much Nickel and Titanium (mass-ion ratio) is released per second.

Results

From the findings of present study, revealed that Round wire shows least metal ion leaching than rectangular wire. This may differ according to the manufacturers choice. Least immersion time shows greatest release of metal ions and Group Ia is better than all other groups.

Conclusion

When comparing three manufactures, Group I shows least Ni and Ti ion leaching among other two groups. When comparing round and rectangular wires; round wires shows less ion release than rectangular wires. The least Ni and Ti ion release is shown by Group Ia at all time periods. The highest Ni and Ti ion release is shown by Group IIIb at all time periods .At each immersion time, T1 shows least Ni and Ti ion release than other time periods, which gradually increases with immersion period. The average amount of ions leached per day from round and rectangular of three manufactures was well below the tolerable daily dietary intake level.

Key words: Superelastic, Leaching, Artificial Saliva, Nickel Titanium Wire, Inductively coupled plasma mass spectrometry.

INTRODUCTION

INTRODUCTION

Nickel titanium wires are the inevitable archwires in orthodontics, for its good mechanical and clinical properties¹. The major property of nickel titanium wires are their springback, which enables a wide deflection and activation range².

Traditionally, Round Nickel titanium (NiTi) wires are used in orthodontics in the initial stages of treatment in the oral environment due to their excellent shape memory and superelasticity, and rectangular wires can be used during final stage of orthodontic treatment³.

Additionally, Nickel in NiTi wires is capable of eliciting toxic and allergic responses and can produce more allergic reactions than any other metal elements⁴.

When using nickel titanium arch wire, there is a possibility of arch wire corrosion results in biologically harmful effects due to the Ni ion release. So, nickel titanium orthodontic arch wire with a good properties including corrosion resistance is essential to its biocompatibility.

It could be noted that the surface corrosion of NiTi arch wires may increase the friction which appears at the interface between the arch wire and bracket, reducing the free sliding action during orthodontic treatment^{5,6,7}.

Nickel-titanium (NiTi) archwires contain 47%–50% Ni, which are the common source of Ni in the intraoral environment of an orthodontic patient. It has been found that several cytotoxic, allergenic and mutagenic actions to Ni in various forms and compounds are also present⁸. Studies also shown that Ni is attributed with different carcinogenic problems. There is a trend for state laws to create awareness and emphasize the necessity for patient awareness about the harmful effects of NiTi⁹.

The metal ions leaching from orthodontic wires cannot be fully evaded; but it is possible to use materials with lower amounts of ions leaching in the mouth. It was proven that the amount of ions, leached from orthodontic wires in saliva was less than the toxic concentrations, which is below the critical value necessary to induce allergy and less than the daily dietary intake levels of 200–300 µg/day¹⁰.

Ni and Ti are much less toxic than other heavy metals. Animal research has shown that a relatively high concentration of Ni is needed to produce toxic effects but that low concentrations of this metal can provoke allergic reactions^{11,12}. Reports in the literature indicate that approximately 10% of the general population is sensitized to Ni and that the prevalence is much higher in female individuals¹³, average daily intake through food has been estimated at 3.52 mg/day for Mn, 200 to 300 mg/day for Ni, 11 280 mg/day for Cr, and 300 to 2000 mg/day for Ti¹⁴.

Most inevitable characteristics of a material to be considered as ideal in dentistry are the Biological safety, functionality, adequate tissue response, and resistance to corrosion etc. As such, metal alloys have been extensively used in orthodontics because of their elasticity, shape memory, hardness, and stress resistance. The biocompatibility of orthodontic materials is widely discussed in recent scientific literature¹⁵.

In 2009 Kuhta et al studied the influence of wire type on the leaching of metal ions and found that leaching of ions was depended on composition of the arch wire, but it was not related to the content of metal. However, there is a lack of data in the relevant literature to support an increased prevalence of clinical adverse effects of Ni, still the studies is been continued to prove the biocompatibility of nickel containing orthodontic appliances¹⁶.

Biocompatibility of any dental material is now a fundamental requirement of successful clinical behavior in oral cavity. Depending on the characteristics and solubility of the products, nickel ions may be released in different places and at different levels, so introduction of these metal ions into the human body is an additional risk to health. Nickel containing alloys has become an integral part of almost every routine orthodontic intervention. The majority of dental allergies, including responses to nickel containing dental alloys, which are mediated through the immune system, comprise type IV hypersensitivity reactions¹⁷.

The purpose of this study is to determine the amount of Ni and Ti ion release from NiTi wires of three different manufactures. The study will be performed by immersion of the samples in artificial saliva at various time intervals and Ni and Ti release was quantified with the use of an inductively coupled plasma mass spectrometry.

AIMS & OBJECTIVES

AIMS

The aim of this study is

- To compare the release of Ni and Ti ions from NiTi wires of three different manufactures-Dentaram,Ormco, 3M Unitek
- To compare the release of Ni and Ti ions from round(0.016), and rectangular (19x25) wires of three different manufactures.
- To compare the release of Ni and Ti ions at different immersion time (T1, T2, T3, T4) in artificial saliva.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

John W Edie et al (1981)²² compared corrosion of the two metals under clinical conditions and found that Nitinol has more susceptible to electrolytic dissolution than stainless steel.

Bernhard Schwaninger et al (1982)³² investigated that failure of nitinol orthodontic wire is because of the presence of surface defects which occurred during manufacturing and not due to the effects of corrosion and also they evaluated the flexural properties of both control and corroded samples of nitinol wires and not found to be statistically different.

HY Park D et al (1983)⁵⁸ estimate the amount of nickel and chromium leached from a orthodontic wire incubated in 0.05 percent sodium chloride solution and they found that the average release of metals was 40 µg nickel and 36 µg chromium per day for a full-mouth appliance, which is in acceptable levels.

Toms AP et al (1988)⁶³ investigated corrosion of orthodontic wire using the electrochemical technique of polarization resistance. They calculated polarization resistance from the slope of the plot, and the corrosion current and rate determined. Problems associated with the technique are non-linear polarization plots and a prior knowledge of Tafel coefficients. In the present work a non-linear optimization procedure was developed to solve the corrosion equations directly for corrosion current and Tafel coefficients.

Edward F Harris et al (1988)⁶⁶ studied that the changes in the mechanical properties of a nickel-titanium in a simulated oral environment, at various levels of

acidity, they found that, mechanical properties were decreased when compared with the group which is kept dry and unstressed.

Dunlap CL et al (1989)⁴³ stated that nickel hypersensitivity is commonly seen in women, because of frequent contact with ornaments which contains nickel. Some other metal are also allergenic but less than nickel such as mercury, beryllium, chromium, cobalt, and gold. Nickel has high possibility of cutaneous allergy but only few cases of oral allergic reactions are reported in dental literature. This prove that high concentration of nickel is needed to provoke oral mucosal lesions compared with skin lesions which is generally regarded as a type IV cell mediated reaction.

Miura F et al (1990)² stated that Nickel titanium (NiTi) wires are now routinely used in dentistry due to their distinctive shape memory and superelasticity. In the initial phase of treatment the new superelastic NiTi can be used, this gives a three dimensional control as well as a horizontal and vertical leveling.

Bishara SE et al (1993)⁴¹ aimed to find the concentration of nickel in blood in orthodontic patients during their initial course of orthodontic therapy. They concluded that there was no consistent increase in nickel level in blood during 4-5 months after appliance insertion. This indicate that orthodontic appliances used, in their "as-received" condition, may corrode in the oral environment, in amounts very below the average dietary intake level.

Justin K. Bass et al (1993)¹ found that nickel sensitivity is more in females than males. (28% in females, 0% in males.) Nickel containing appliance had only little effects on the gingiva and allergy may usually occurs due to cutaneous nickel contact.

Kalimo K et al (1993)⁴⁰ stated that development of nickel allergy was significantly associated with skin piercing (54% compared with 12%). There was a slight but non-significant difference in the prevalence of nickel allergy between students who had been subjects for orthodontic treatment (49%) compared with non-treated ones (58%) if they had pierced skin and there were no significant differences in the development of nickel allergy among students who had had permanent dental braces before (50%) or after skin piercing (48%). They concluded that orthodontic treatment may not lead to tolerance induction on all occasions, and sensitization through permanent devices seems to be possible.

Veien NK et al (1994)⁴⁴ done a study on 5 patients with dermatitis or stomatitis related to the use of appliances were selected. All the patients were undergone patch test. 3 of the patients had recurrent vesicular hand eczema, which flared after oral challenge with 1 of the metals used in their orthodontic appliances. The dermatitis of 4 of the 5 patients cleared completely upon the removal of their metal orthodontic appliances or their replacement with appliances made of acrylics.

Dietmar Segner Dagmar Ib el et al (1995)²⁹ investigated that many materials either did not show any pseudoelastic properties or that the wire parameters were such that they did not give any benefit over traditional work hardened NiTi materials. In many archwires the beginning of the plateau and desired characteristics began only when the archwire was displaced 1 mm or more.

Waheidi EM (1995)⁴⁵ stated that nickel is the most common allergen. Orthodontic appliances with nickel content are main cause of allergic reactions, which results in type IV delayed hypersensitivity immune response. Allergic reactions may involve intra- and extraoral clinical signs, comprising diffuse

erythema, edema, eczema, fissuring, desquamation, and symptoms such as itching and soreness. He aimed to provide orthodontists with the necessary knowledge about the biologic mechanisms, diagnostic tools, and clinical signs, as well as the treatment alternatives to nickel-induced allergic reactions.

DJ Wever et al (1997)²³ aimed to evaluate the short-term biological safety of the NiTi alloy. He concluded that NiTi alloy showed no cytotoxic, allergic or genotoxic activity, similar to the clinical reference control material. This biological behaviour was most likely due to a minimal release of ions and in that way a reflection of the good corrosion resistance of the NiTi alloy.

Janson GR et al (1998)⁷¹ compared the prevalence of nickel hypersensitivity reaction before, during, and after orthodontic therapy with conventional stainless steel brackets and wires. Nickel may induce delayed hypersensitivity reaction (type IV immune response) as it is a strong biological sensitizer and he concluded that nickel hypersensitivity reactions only occurs in individuals with previous allergic history to metals as well as daily use of metal objects. The result was not statistically significant in the prevalence of contact dermatitis. He also suggest that orthodontic therapy with nickel containing appliances does not initiate or aggravate a nickel hypersensitivity reaction.

Torrisi L et al (1998)³⁵ compared NiTi with stainless steel and given Special interest to the use of NiTi alloy as orthodontic wires and endodontic instruments. The superelastic properties of orthodontic wires and endodontic files are measured in accordance with stress, strain, and temperature. Both are subjected to surface analysis to control the interface with the biological environment in which they will be immersed.

Kim H et al (1999)⁵⁵ stated that orthodontic wires containing nickel is the main cause of allergic reactions. The purpose of this study was to determine if there is a significant difference in the corrosive potential of stainless steel, nickel titanium, nitride-coated nickel titanium, epoxy-coated nickel titanium, and titanium orthodontic wires. Variability in breakdown potential of nickel titanium alloy wires differed with different manufactures. Least corrosive potential shown by Titanium wires and epoxy-coated nickel titanium wires. The use of titanium or epoxy-coated wires during orthodontic treatment is recommended for patients allergic to nickel.

Jia W et al (1999)⁴² compare and measure the amount of nickel released from three types of nickel-containing arch wires into a synthetic saliva, and determine if the concentrations were sufficient to elicit either and the result was the maximum amount of nickel released from all tested arch wires was 700 times lower than the concentrations necessary to elicit cytotoxic reactions in human.

Widu F et al (1999)⁵⁰ stated that the corrosion behavior of orthodontic wires is an important factor which determining their biocompatibility. They classified the wires into two groups, one with a high and a second group with a low tendency towards corrosion and analyses of wires after clinical usage indicate that changes of wire surfaces show the same characteristics under in vitro conditions.

Nakagawa M et al (1999)³⁸ clarified the effects of fluoride concentration and pH on the corrosion behavior of Ti, he conducted anodic polarization and immersion tests in NaF solution of various concentrations and pH values. Then the concentrations of dissolved Ti in the test solutions were analyzed. Results showed that obvious limits of fluoride concentration and the pH value at which the corrosion behavior of Ti changed. The results of this study revealed a relation between the

fluoride concentrations and pH values at which Ti corrosion occurred and provided data on such corrosion in environments where the fluoride concentration and pH value are known.

NP Hunt et al (1999)²⁷ investigate the effect of surface roughness on the corrosion rates of SS, NiTi, Co Cr, and TMA wires. Wires in first group were commercially polished for an equal surface finish; wires in the second group were kept to compare as “as received”. The relative corrosion rates (expressed in terms of corrosion current density) were estimated, in that NiTi wires shows the greatest dissolution in the as-received state. Corrosion rate of NiTi decreases with increased polishing.

Chung-Ju Hwang et al (2001)³³ divided the patients with orthodontic appliance into 4 groups. In his 3-month-long investigation, they found that there was no change in the amount of nickel released in all four groups Group A, Group B, Group C, and Group D after 14 days, 3 days, 7 days, and 1 month respectively; they observed a decrease in metal ion leaching when immersion time increased.

Nicolas Schiff et al (2002)³⁷ aimed to compare the NiTi, NiTiCo and TiAl6V4 alloys with the titanium, regarding the corrosion resistance in artificial saliva with different pH and fluoride contents. The corrosion potential was measured over time. Their results have shown that TiAl6V4 alloys have a good corrosion resistance as good as that of titanium except NiTi and NiTiCo alloys.

Rahilly G et al (2003)⁴⁶ stated that Nickel is the most common metal to cause contact dermatitis in orthodontics. Nickel-titanium alloys may have nickel content in excess of 50 per cent and can thus potentially release enough nickel in the oral environment to elicit manifestations of an allergic reaction. Stainless steel has a

lower nickel content (8 per cent Stainless steel orthodontic components are therefore very unlikely to cause nickel hypersensitivity. This article diagnosed the nickel allergy in orthodontics and describes alternative products that are nickel free or have a very low nickel content, which is appropriate in patients diagnosed with a nickel allergy.

Faccioni F et al (2003)⁶⁴ aimed to investigate the biocompatibility in vivo of fixed orthodontic appliances, evaluating the presence of metal ions in oral mucosa cells, their cytotoxicity, and their possible genotoxic effects. The results indicate that nickel and cobalt concentrations were higher, respectively, in the patients than in the controls. The biologic effects, evaluated, results, indicated that both metals induced DNA damage. This study corroborates that nickel released from fixed orthodontic appliances can induce DNA damage in oral mucosa cells.

Shin JS et al (2003)¹⁸ determined surface corrosion of the archwires macroscopically, with scanning electron microscopy, and with spectrophotometry for that Simulated fixed orthodontic appliances were constructed, immersed and then incubated in artificial saliva for three months. Two types of stainless steel archwires and two types of NiTi wires were used. Uniform corrosion was observed on stainless steel wires, and a slight colour change was detected on the NiTi wires beneath stainless steel ligatures. But the NiTi archwires did not corrode, and there was no significant difference in surface morphology. NiTi archwires are significantly more stable and resistant to corrosion than stainless steel archwires.

Theodore Eliades et al (2004)⁵⁶ found that orthodontic alloys occurs in the intraoral environment, regardless of the alloys metallurgic structure, and it is also known that the extent of manufacturing defects may accelerate the corrosion

process. Ni at nontoxic concentrations induces DNA damage by base damage and DNA strand scission (single strand breaks).

Kazuyuki Kaneko et al (2004)³⁰ tried to investigate the degradation in performance of four orthodontic alloys nickel-titanium, beta titanium, stainless steel, and cobalt-chromium nickel, which is dipped in an acid fluoride solutions. Results shows that degradation of orthodontic wires of titanium alloys occurs faster.

Yong Hoon KWON et al (2004)⁵⁷ examined the effect of acidic fluoride solution on NiTi arch wires and element release from wire under four different test solutions after 1 or 3d immersion. Increased tensile strength showed by 3M wires whereas G&H and Ormco wires showed decreased strength. Element release in the test solution increased as NaF concentration and the period of immersion increased, and as pH valued decreased. The factors affecting these properties were NaF concentration, pH value, and the period of immersion

Genelhu MC et al (2005)⁶⁵ found that dissolution of orthodontic appliances and leaching of nickel ions results in an inflammatory response in some patients. They concluded that females with a history of allergic reactions had a greater predisposition to some steroids. A prior allergy to be taken in account and should be recorded in the patient's medical history.

Cioffi M et al (2005)²⁵ investigate the alloys based on Ni–Ti intermetallics generally exhibit special shape memory and pseudoelastic properties, which make them desirable for use in the dental field as orthodontic wires. Because the allergenicity of Nickel the possibility of metal ion release from these materials is of high concern.

Her-Hsiung Huang et al (2005)³¹ investigated the surface characterizations and corrosion resistance of as-received commercial nickel–titanium (NiTi) dental orthodontic archwires from different manufacturers using a cyclic potentiodynamic test in artificial saliva with various acidities. The results showed that, because of presence of TiO₂ and small amounts of NiO, the surface structure of the passive film on the tested NiTi. Various manufacturers shows a different surface topography on the NiTi wires. Corrosion potential, corrosion rate, passive current, breakdown potential, and crevice-corrosion susceptibility these all depends on the wire manufacturer and solution pH. They concluded that the difference in the corrosion resistance of orthodontic archwires did not correspond with the surface roughness and pre-existing defects.

Her-Hsiung Huang et al (2005)¹⁵ investigate the disparity in corrosion resistance of NiTi wires from different manufacturers. Nickel-titanium (NiTi) wires produced by various manufacturers may have different corrosion resistance in acidic oral environment. The results showed that NiTi wires from different manufacturers had a statistically significant difference in Rp (P, .001).

Hyung-SooAhn et al (2006)³⁴ evaluated the effect of pH and temperature on orthodontic NiTi arch wires after immersed in an acidic fluoride solution. Each as-received wire exhibited quite different microhardness values. The reduction of microhardness, 1.2-5.7%, occurred after immersion, pH and temperature influence the volumetric weight change, concentration of the released elements, and surface morphology were influenced by. At pH 3.5 of 60°C solution, the greatest weight loss, release of elements, and corrosion of surface occurred from the wires. At pH 6, no such loss or release occurred regardless of temperature. At 5°C solution, the surface exhibited minor corrosion regardless of pH value.

Peitsch T et al (2007)⁶⁷ investigated the effect of mechanical loading and of surface treatment on the leaching of nickel from nickel-titanium orthodontic wires. They monitored the release of nickel by atomic absorption spectroscopy and found that mechanically loaded wires released significantly more nickel than nonloaded wires. The amount of released nickel was little in all cases.

Wang J et al (2007)³⁹ aimed to study the mechanism of the cracking of orthodontic NiTi wire. The results showed that there were three areas at the fracture surface of NiTi orthodontic wire. Area '1' was a tool-made notch. Crack initiated from the root of this notch and propagated to form Area '2', which was perpendicular to the wire axis and covered by surface film. The pH of saliva and applied stress influence cracking process of NiTi alloy. They found that at high stress and low pH, this NiTi alloy was most sensitive to cracking.

Olga-Elpis Kolokitha et al (2008)¹³ investigated the effect of orthodontic therapy on the prevalence of nickel hypersensitivity and compare it with the prevalence in the general population, they found orthodontic treatment is not associated with an increase in the prevalence of Ni hypersensitivity, unless subjects have a history of Ni exposure from cutaneous piercing

Rodrigo Matos de Souzaa et al (2008)¹⁴ investigated variability in ion concentration among individuals is a common finding when examining the release of metal ions in saliva. This variation may be related to several factors since saliva does not present a constant composition and may be different among individuals or even among periods for the same individual. The physical properties, amount, and composition of saliva are influenced by factors such as diet, period of the day, and psychic conditions.

Noble J et al (2008)⁶ studied two cases and found that oral exposure to nickel may induce immunologic tolerance to nickel and thereby reduce the incidence of nickel exposure causes formation of free radicals in various tissues in both human and animals. After effects of nickel intoxications are various modifications to DNA bases, enhanced lipid peroxidation, and altered calcium and sulphhydryl homeostasis.

Das KK et al (2008)⁹ investigate common harmful health effect of nickel in humans is an allergic skin reaction in those who are sensitive to nickel. Nickel is the most common reason of immediate and delayed hypersensitivity noticed in occupationally exposed as well in the general population. The metal is not just an allergen but also a potential immunomodulatory and immunotoxic agent in humans. Nausea, vomiting, abdominal pain, diarrhea, headache, cough, shortness of breath, and giddiness were reported for workers of an electroplating plant who drank water contaminated with nickel chloride and nickel sulphate (1.63 g/l). Signs and symptoms of toxicity lasted for up to 2 days. Most chronic inhalation exposures involve occupational exposure to nickel dust or nickel vapors resulting from welding nickel alloys which may lead to respiratory disorders such as asthma, bronchitis, rhinitis, sinusitis, and pneumoconiosis.

Svetlana A Shabalovskaya et al (2009)²⁴ found that the patterns of Ni release from Nitinol vary depending on the type of material (Ni–Ti alloys with low or no processing versus commercial wires or sheets). The present study of Nitinol wires with surface oxides resulting from production was conducted to identify the sources of Ni release and its distribution in the surface sublayers. Orthodontic wires in the as-received state showed low breakdown potentials. Nitinol wires with the thick TiO₂ layer showed the highest Ni release.

Maja Kuhtaa et al (2009)¹⁶ found that leaching of metal ions depends on the composition of the archwire, not the components of amount of metal in a particular arch wire. Amount of all released metal ions were well below toxic levels and did not exceed the daily dietary intake. However, these levels are sufficient to cause an allergic reaction because of the high haptenic potential of released elements.

Evangelia Petoumenou et al (2009)²⁸ tried to estimate the chances of risk to use nickel-titanium wires in patients who have nickel hypersensitivity to examine whether these wires can change nickel concentration in the saliva. By using mass spectrometry, there is no statistically significant differences were found in the nickel concentrations in the samples taken without appliances and in those obtained 2 weeks after engaging the bands and brackets. Saliva taken immediately after placement, associated with an increase of the ion concentration in the patient's saliva.

Tzu-Hsin Lee et al (2010)⁴ found that the archwire manufacturer and NaF concentration had a statistically significant influence on the corrosion resistance, of the four different kinds of commercial NiTi orthodontic archwires in acidic fluoride-containing artificial saliva. The surface topography of the commercial NiTi archwires with identical surface chemical structures, TiO₂, and small amounts of NiO, did not correspond to the variation in corrosion resistance in fluoride containing artificial saliva.

Luciane Macedo de Menezes et al (2010)³⁶ stated that several metallic alloys used in orthodontics have nickel and chromium as their components. These metal ions are known to be essential elements but are considered one of the most

common causes of allergic contact dermatitis. The allergic reactions are caused by a direct relationship with the presence of nickel in the environment and may be caused by ingestion or direct contact with the skin and/or mucosa.

Abalos C et al (2011)⁶⁸ investigate several topographical features and their influence upon fluoride corrosion were studied. Four topographies (smooth, dimple, scratch, and crack) according to the main surface defect were characterized. An increase in the surface defects and/or roughness was observed on the cracked and scratched surfaces. Thus there is an increase in corrosion behavior. Smooth and dimpled surfaces are the best surfaces for the orthodontic archwires to reduce corrosion. The increase in defects was independent of roughness.

Fariborz Amini et al (2012)⁶¹ concluded that the corrosion of orthodontic appliances and their subsequent metal ion release in the oral environment is governed by two main factors. The first is the manufacturing process, which includes characteristics of the metals used. Second is environmental factors, such as mechanical stress, diet, time of the day, salivary flow rate, and health and psychosomatic condition of the individual.

Elisa J Kassab et al (2013)⁵¹ compared resistance of NiTi and TMA to leaching with and without addition of fluoride, thus assess the influence of fluoride content on the corrosive behavior of orthodontic wires. Concluded that in the presence of fluoride NiTi corrosion resistance decreases.

Luca Lombardoa et al (2013)⁵³ stated NiTi archwires have become increasingly popular in recent years because of their ability to release light forces,

which are considered to improve the efficiency and efficacy of treatment, especially during initial alignment and leveling phases.

Roberto Rongoet al (2014)¹² studied the clinical use of wires altered their surface properties and increased surface roughness and level of friction. After clinical use, surface roughness increased considerably. As-received superelastic Titanium Memory Wire shows lowest levels of friction. All the wires shows an increase in friction when tested with ceramic brackets.

Visnja Katica et al (2014)⁵² tried to assess the effect of various surface coating and their influence on leaching of nickel-titanium orthodontic. They concluded that uncoated NiTi wires showed anticorrosive properties, than coated NiTi wires.

Manu Krishnan et al (2014)⁵⁴ tried to evaluate and compare the association of corrosion behavior of commercially available surface coated nickel titanium wires conventional NiTi wires. He compared the Surface modification of NiTi wires may improve its corrosion resistance and decreasing surface roughness. They concluded that, anticorrosive features depends on nature of coating material and surface roughness.

Senkutvan RS et al (2014)⁴⁷ investigated to analyze and evaluate the amount of leaching of Ni ions from various manufactures arch wires. They found that the daily release of NiTi, SS, Cu NiTi and ion implanted NiTi by an orthodontic appliance in acid pH, particularly favorable to corrosion, was well below with a normal daily diet.

Sandhya M et al (2015)⁴⁸ aimed to give an awareness to doctors, as the need for orthodontic treatment is increasing, reported number of patients with allergy has been also increased. Therefore, it is imperative to take thorough history from patients. The unexplained occurrence of gingival or mucosal inflammation, erythema, or the report of a burning sensation in the patient's oral tissues should be investigated as a potential allergic response to their orthodontic appliances.

Sepideh Arab et al (2015)⁴⁹ concluded that the daily body intake of nickel and chromium via food is approximately 300-500 and 5-100 microgram, which is obviously less than daily intake which is not capable of inducing systemic toxicity. The average concentration of nickel in drinkable water is less than 20 microgram per liter. The results of the present study revealed that the nickel ion release decreases in the second 28 days, it is possible to use the biomechanical advantages of these arch wires without biological concerns.

Arash Azizi et al (2016)⁷ found the amount of nickel and titanium ions released from two wires with different shapes and a similar surface area were evaluated in this study. Their results clearly revealed that rectangular group shows higher leaching than round wires. They concluded that shape of the wire and increase of time control the release of metal ions.

Wendl B et al (2017)⁷⁰ evaluated the leaching of Mn, and Ni from orthodontic appliance during treatment. They found that Mn, and Ni, release is more in bands after that brackets and archwires. After six months of treatment some corrosion products may developed on bands in artificial saliva because of some metals were welded to the bands. They concluded that leaching of metals by orthodontic materials are at very low levels, below typical dietary intake.

MATERIALS & METHODS

MATERIALS AND METHODS

This study has been conducted at sophisticated instrumentation and computation center (SICC), Thiruvananthapuram, Kerala.

Materials and Instruments

Archwires

- 0.016 NiTi (Dentaram, Ormco, 3M Unitek) round Orthodontic Archwires
- 19x25NiTi (Dentaram, Ormco, 3MUnitek) rectangular Orthodontic Archwires

Equipment used

- Inductively coupled plasma mass spectrometry (Thermo scientific, ICPA Q, Bremen, Germany)
- Digester(Anton parrmultiwave 3000 Microwave digester)
- Incubator (HEPA,Forma series II, Thermo electron Corporation)
- Shaker

Materials

- Artificial saliva (Biotene Glaxo Smith Kline, Consumer, Healthcare, L.P Moon Township, USA)
- Sodium Hydroxide (Medilisechemicals, Kannur)
- pH paper (INDICATORS India ,Product No:74026A)
- 6 ml syringe (Dispovan, Hindustan syringes and Medical Devices Ltd, Lour Mount)
- Polypropylene beakers

Inclusion criteria: Most commonly using 7 inch long round and rectangular Niti wires of three different manufactures

- (0.016) and (19x25) NiTiarchwires of Dentaaurum
- (0.016) and (19x25) NiTiarchwires of Ormco
- (0.016) and (19x25 NiTiarchwires of 3M Unitek

Exclusion: Damaged archwire

CONSTITUTION OF TEST GROUPS OF THE ARCHWIRES

Sample size: 126

Groups: Three main groups, Six subgroups

Sample size of each group: 21

Detailed description:

Six groups and subgroups

Groups	Subgroups	Colour Code	Sample Code	Size	Manufacturer
Group I	Group I	Red	IA	0.016	Dentaaurum
	Group II	Green	IB	19x25	Dentaaurum
Group II	Group III	Blue	IIA	0.016	Ormco
	Group IV	Orange	IIB	19x25	Ormco
Group III	Group V	Violet	IIIA	0.016	3M Unitek
	Group VI	Black	IIIB	19x25	3M Unitek

METHODOLOGY**Procedure in detail**

In this study superelastic NiTi Archwires of three different manufactures which is in two shapes Round and Rectangular of commonly using dimensions 0.016 and 19x25 respectively of 7 inches long are used in this study. Wires are divided into three groups

Groups	Subgroups	Colour Code	Sample Code	Size	Manufacturer
Group I	Group I	Red	IA	0.016	Dentaurum NiTi wires
	Group II	Green	IB	19x25	Dentaurum NiTi wires
Group II	Group III	Blue	IIA	0.016	Ormco NiTi wires
	Group IV	Orange	IIB	19x25	Ormco NiTi wires
Group III	Group V	Violet	IIIA	0.016	3M NiTi wires
	Group VI	Black	IIIB	19x25	3M NiTi wires

In this study inductively coupled plasma mass spectrometry (Thermo scientific, ICPA Q, Bremen, Germany) is used for the quantification of Nickel and Titanium from Artificial Saliva (Biotene GlaxoSmithKline Consumer Healthcare, L.P Moon Township, USA)

Sample Preparation

The testing solution used in the study is artificial saliva (Biotene GlaxoSmithKline Consumer Healthcare, L.P Moon Township, USA), components are Magnesium chloride 6 H₂O - 0.052, Sorbitol solution (70%) - 60, xanthan gum

Sodium benzoate - 0.844, Calcium chloride anhydrous - 0.146, Potassium phosphate dibasic - 0.34 Methyl paraben - 2, Hydroxyethyl cellulose 3.5, Potassium sorbate - 1.2 Purified water, sunflower oil; which will be adjusted to pH 6.75 ± 0.15 (neutral PH, which is seen in normal oral environment) buffer solutions (which can resist changes in pH when acid or alkali is added to it) by adding 1 mMNaOH (Sodium Hydroxide).

Procedure

Each wires separately dipped into 126 polypropylene beakers containing 50 ml of buffer solution. Then incubate at 37°C (as in normal oral environment) to promote cell growth and place on a shaker for mild agitation to simulate in vivo situation.

After each immersion period(At T1=1 hour, T2=24 hour, T3=1 week, T4=3 week) Whole 126 polypropalene beakers were taken out from the incubator and 5ml of eluent (Fluid used to elute a substance) is removed from each beakers using a syringe with a plastic tip and acid digestion has been done.

This solution contains 2% nitric acid, and 0.5% hydrochloric acid added to stabilize the elements. Then the samples are digested (Anton parr multiwave 3000 Microwave digester) for few minutes (To dissolve the metals, at high temperature and pressure upto 260⁰ C) ,then make up the solution by adding 1 drop of Hcl and place into the auto sampler tray of ICP MS.

Sample introduction is achieved via analytical nebulizers. Nebulizer converts liquids into an aerosol, and that aerosol can then be swept into the plasma to create the metal ions. Then the aerosol moves into the bottom of the torch body of the machine.

ICP-MS Interface

The ionisation occurs at atmospheric pressure, the interface between the Inductively Coupled Plasma and Mass Spectrometry components becomes essential in creating a vacuum environment for the MS system. Ions flow through a small orifice, into a pumped vacuum system, and the sample ions are passed into the MS system at high speeds. The entire mass spectrometer should be kept in a vacuum so that the ions are free to move without collisions with air molecules, meanwhile the ICP should be maintained at atmospheric pressure.

Plasma torch

The ICP torch comprised of a copper induction coil wrapped around a concentric quartz Structure. The plasma used here is partially ionizing argon gas. Argon gas is continuously flowing throughout the quartz torch, and a radio-frequency generator provides power to the RF coil at oscillating frequencies.

The required energy is obtained by pulsing an electrical current that surround the argon gas. After the sample is injected, atomization occurs and thus the plasma's extreme temperature causes the sample to separate into individual atoms then the plasma ionizes these atoms so that they can be detected by the mass spectrometer.

The end of this torch is placed inside an induction coil supplied with an electric current. When ample argon supplied, the plasma will reach equilibrium and remain at a constant temperature of about 6,000°C for the duration of analysis. To introduce free electrons into the gas stream argon gas is introduced between the two outermost tubes of the torch. These electrons interact with the magnetic field of the induction coil. These ions are then separated and detected by the mass spectrometer⁵⁹.

Interface

The interface serves to allow the ICP and MS portions to be coupled. The first component in the sample matrix of ICP torch is the sampler cone. This is a water cooled cone allowing the hot plasma gas to enter a depressurizing chamber. A fraction of this gas then passes through a skimmer cone, and into a chamber that is maintained at a vacuum of that of the MS. This two-step pressure reduction allows the ionic gas to enter the MS at proper temperature and pressure.

Mass Spectrometry

In the first stage, ions are removed from the plasma by a pumped extraction system. After passing through the sample and skimmer cones, the ion stream is focused into the quadrupole region by single ion lenses. Then these ions pass through a charged metallic cylinder which keeps the ion beam from diverging. The suitable type of reaction gas and pressure is set, and adjustments can be done in the computer software.

Energetic electrons collide with analyst molecules and fragmentation occurs to produce ions of lower masses. Ions are dispersed in the mass analyser based on their mass-to-charge ratio (m/z).

Four parallel cylindrical rods (approximately 1 cm in diameter and 15-20 cm long) serve as electrodes; opposite rods are connected electrically with one pair connected to the positive side of direct current (dc) source, and second one to the negative terminal.

Detector

The ion detector used is the channeltron electron multiplier. This cone or horn shaped tube has a high voltage applied to it opposite in charge to that of the

ions being detected. When they strike the surface, additional secondary electrons are emitted, which move farther into the tube. As the process continues, even more electrons are formed, resulting in as many as 10⁸ electrons at the other end of the tube after one ion strikes at the entrance of the cone.⁶⁰

After the analysis of the first sample, the instrument can be switched to work on the next sample. A series of such sample measurements requires the instrument to have plasma ignited. Then measure the amount of ion released at four immersion times: T₁=1 hour, T₂=24 hours, T₃=1 week, T₄=3 weeks using the software Thermoscientific Qtegra™ Intelligent Scientific Data Solution Software (ISDS) which is attached to the ICP MS instrument. This shared software approach provides control and data processing for a range of elemental and isotopic analysis technologies. The output is numerical, and provided in counts per second, i.e., how much Nickel and Titanium (mass-ion ratio) is released per second. Levels are recorded and displayed in computer monitor in parts per million (ppm).

Statistical Analysis

The data obtained was analysed using Statistical Package for Social Sciences (16.0) version software used for analysis. One-way ANOVA (Post hoc) followed by Dunnett t test applied to find the statistical significant between the groups. Unpaired t test applied to find the statistical significant within the groups. P value less than 0.05 considered statistically significant at 95% confidence interval.

COLOR PLATES

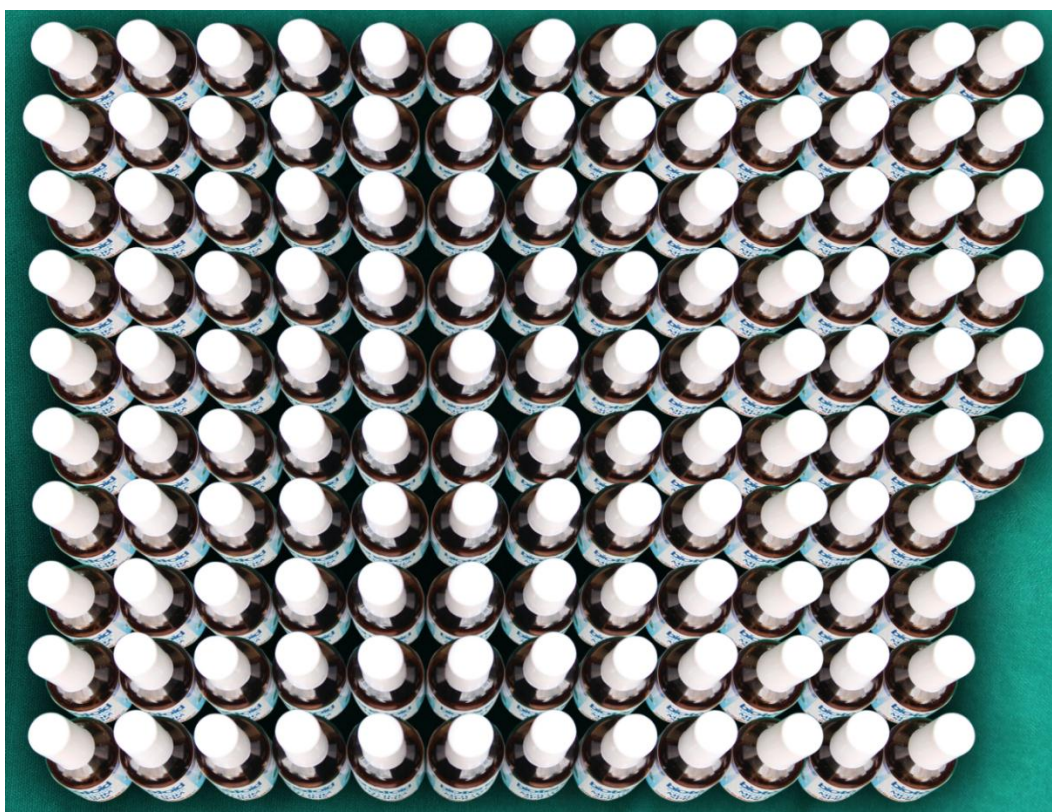


Fig 1. Testing sample - artificial saliva



Fig 2. 0.016 Dentaurum Wires



Fig 3. 19x25 Dentaurum Wires



Fig 4. 0.016 Ormco Wires



Fig 5. 19 x 25 Ormco Wires

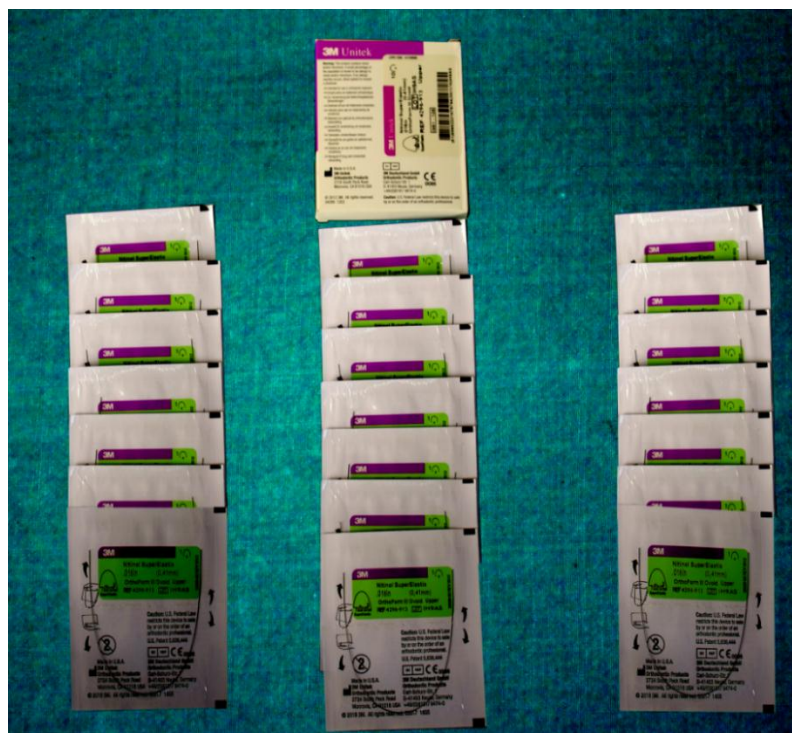


Fig 6. 0.016 3M Wires

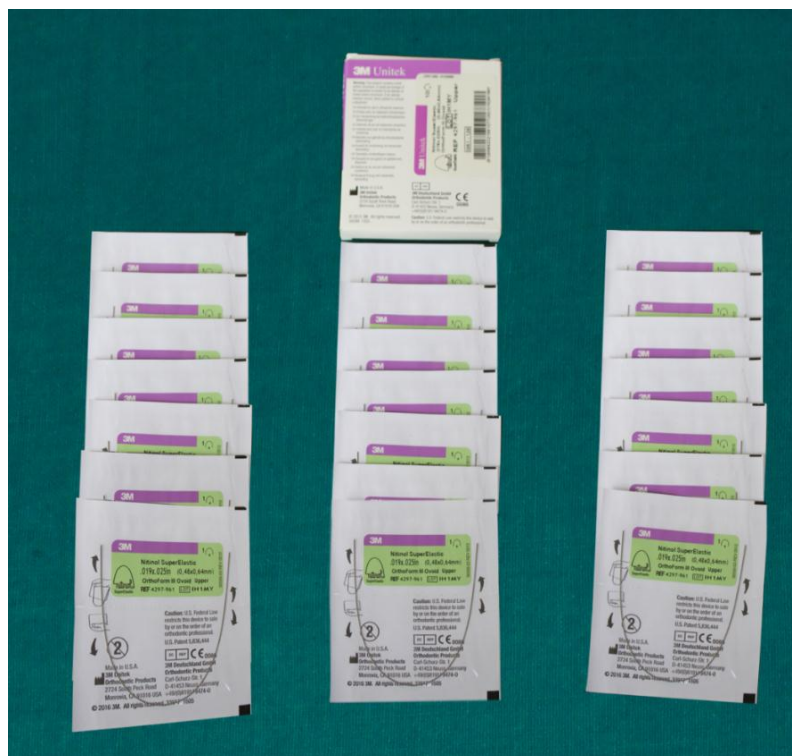


Fig 7. 19 x 25 3M Wires

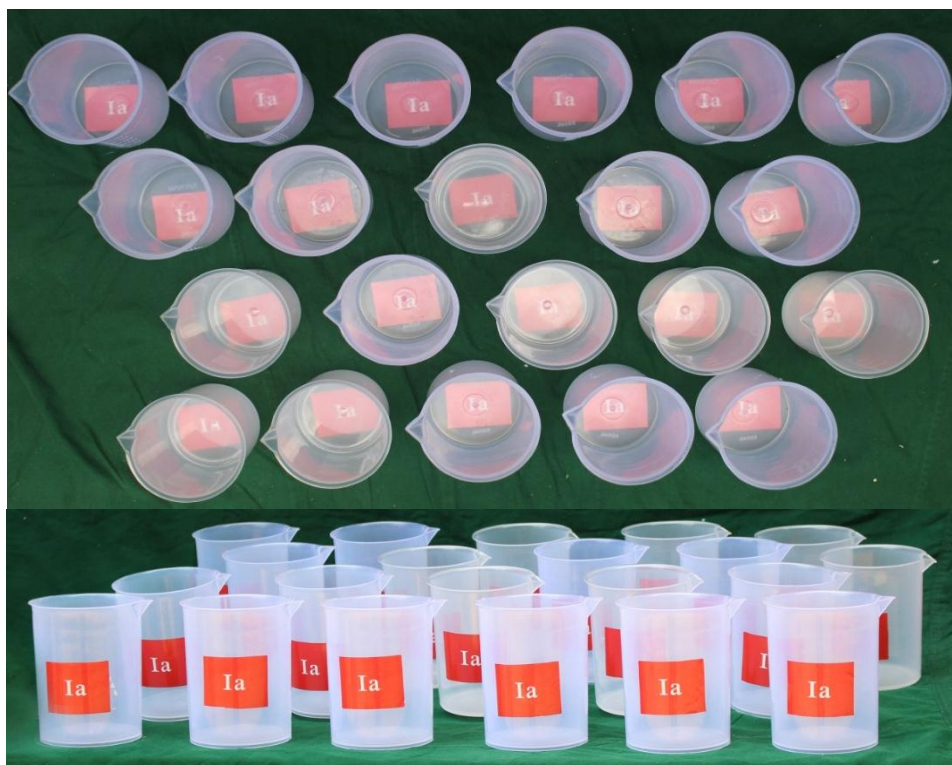


Fig 8. Group Ia



Fig 9. Group Ib



Fig10. Group IIa



Fig 11. Group IIb



Fig 12. Group IIIa

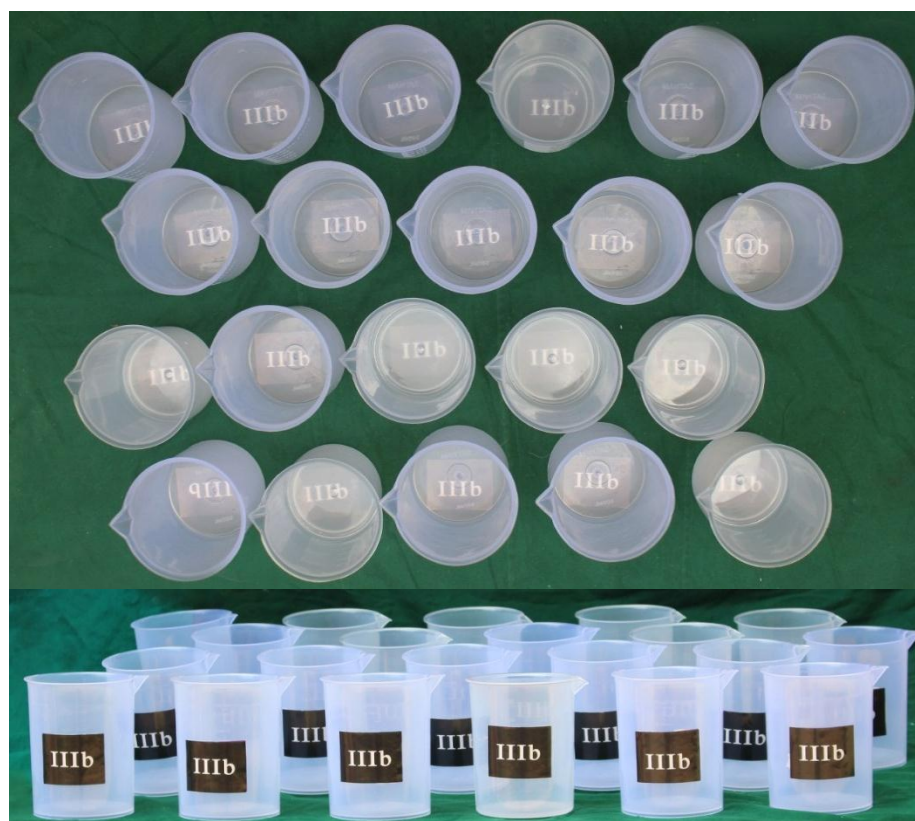


Fig 13. Group IIIb

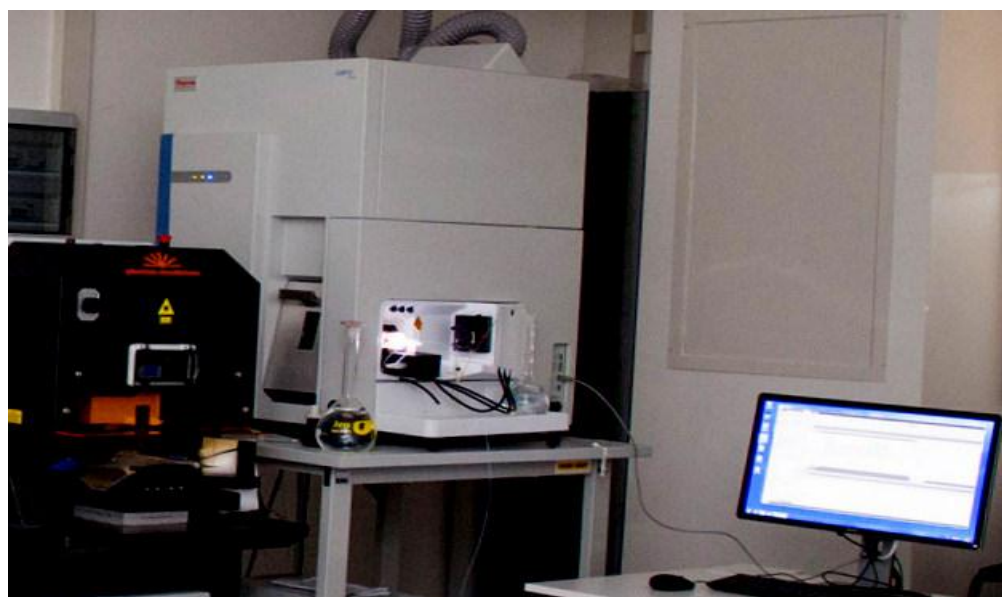


Fig 14. ICP-MS



Fig 15. Sample Introduction System



Fig 16. Incubator



Fig 17. Digester



Fig 18. Shaker

RESULTS & OBSERVATIONS

RESULTS AND OBSERVATION

Statistical analysis: The data was expressed in mean and standard deviation. Statistical Package for Social Sciences (16.0) version used for analysis. One way ANOVA (Post hoc) followed by Dunnet t test applied to find the statistical significant between the groups. Unpaired t test applied to find the statistical significant within the groups. P value less than 0.05 considered statically significant at 95% confidence interval.

Table-1: Mean Ni and Ti release values of Group-I

Time	Group-IA (MEAN±SD)		Group-IB (MEAN±SD)	
	Ni	Ti	Ni	Ti
T1 (1 hour)	0.21±0.004	0.11±0.006	0.21±0.004	0.13±0.008
T2 (24 hours)	0.25±0.005	0.13±0.008	0.25±0.009	0.14±0.008
T3 (1 week)	0.28±0.006	0.14±0.005	0.28±0.006	0.15±0.005
T4 (3 weeks)	0.31±0.008	0.16±0.007	0.32±0.009	0.17±0.008

Table-2: Mean Ni and Ti release values of Group-II

Time	Group-IIA (MEAN±SD)		Group-IIB (MEAN±SD)	
	Ni	Ti	Ni	Ti
T1 (1 hour)	0.24±0.005	0.14±0.007	0.25±0.009	0.15±0.007
T2 (24 hours)	0.28±0.006	0.16±0.007	0.31±0.005	0.17±0.007
T3 (1 week)	0.31±0.005	0.17±0.008	0.34±0.001	0.18±0.008
T4 (3 weeks)	0.34±0.007	0.20±0.007	0.37±0.008	0.21±0.007

Table-3: Mean Ni and Ti release values of Group-III

Time	Group-III A (MEAN±SD)		Group-III B (MEAN±SD)	
	Ni	Ti	Ni	Ti
T1 (1 hour)	0.25±0.009	0.18±0.008	0.28±0.006	0.21±0.10
T2 (24 hours)	0.31±0.005	0.21±0.009	0.37±0.005	0.23±0.001
T3 (1 week)	0.37±0.005	0.28±0.009	0.44±0.10	0.29±0.006
T4 (3 weeks)	0.44±0.001	0.30±0.007	0.50±0.10	0.32±0.10

Table-4: Comparison of mean Ti and Ni release values of three different round wires at T1 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.21±0.004	0.12	0.11±0.006	0.04*
Group-IIA	0.24±0.005	0.04*	0.14±0.007	0.03*
Group-IIIA	0.25±0.009	0.03*	0.18±0.008	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three round wires at T1, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-5: Comparison of mean Ti and Ni release values of three different rectangular wires at T1 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IB	0.21±0.004	0.13	0.13±0.008	0.04*
Group-IIB	0.25±0.009	0.03*	0.15±0.007	0.03*
Group-IIIB	0.28±0.006	0.02*	0.28±0.006	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three rectangular wires at T1, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.03 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-6: Comparison of mean Ti and Ni release values of three different round wires at T2 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.25±0.005	0.14	0.13±0.008	0.17
Group-IIA	0.28±0.006	0.04*	0.16±0.007	0.04*
Group-IIIA	0.31±0.005	0.03*	0.21±0.009	0.03*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three round wires at T2, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.04 and 0.03 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-7: Comparison of mean Ti and Ni release values of three different rectangular wires at T2 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IB	0.25±0.009	0.17	0.14±0.008	0.19
Group-IIB	0.31±0.005	0.03*	0.17±0.007	0.04*
Group-IIIB	0.37±0.005	0.02*	0.23±0.001	0.03*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three rectangular wires at T2, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.03 and 0.02 respectively for Ni and 0.04 and 0.03 for Ti, shows there is significant difference compared with other groups.

Table-8: Comparison of mean Ti and Ni release values of three different round wires at T3 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.28±0.006	0.11	0.14±0.005	0.13
Group-IIA	0.31±0.005	0.04*	0.17±0.008	0.04*
Group-IIIA	0.37±0.005	0.03*	0.28±0.009	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three round wires at T3, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.04 and 0.03 respectively for Ni and 0.04 and 0.02 for Ti, shows there is significant difference compared with other groups.

Table-9: Comparison of mean Ti and Ni release values of three different rectangular wires at T3 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IB	0.28±0.006	0.13	0.15±0.005	0.14
Group-IIB	0.34±0.001	0.04*	0.18±0.008	0.04*
Group-IIIB	0.44±0.10	0.02*	0.29±0.006	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three rectangular wires at T3, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-10: Comparison of mean Ti and Ni release values of three different round wires at T4 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.31±0.008	0.11	0.16±0.007	0.13
Group-IIA	0.34±0.007	0.04*	0.20±0.007	0.04*
Group-IIIA	0.44±0.001	0.02*	0.30±0.007	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three round wires at T4, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-11: Comparison of mean Ti and Ni release values of three different rectangular wires at T4 Period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IB	0.32±0.009	0.12	0.17±0.008	0.14
Group-IIB	0.37±0.008	0.03*	0.21±0.007	0.03*
Group-IIIB	0.50±0.10	0.02*	0.32±0.10	0.02*

(*p<0.05 significant compared)

On comparison of Ni and Ti release of three round wires at T4, Group Ia compared with Iia and IIIa shows there is no significant difference compared with other groups. P value of Group Iia and IIIa is 0.03 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

Table-12: Comparison of mean Ti and Ni release values between the groups

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.21±0.004	0.14	0.11±0.006	0.04*
Group-IB	0.21±0.004	0.13	0.13±0.008	0.04*
Group-IIA	0.24±0.005	0.04*	0.14±0.007	0.03*
Group-IIB	0.25±0.009	0.03*	0.15±0.007	0.03*
Group-IIIA	0.25±0.009	0.03*	0.18±0.008	0.02*
Group-IIIB	0.28±0.006	0.02*	0.28±0.006	0.02*

Comparison of mean Ni release between the groups at T1, P value of Group Ia is 0.14 and P value 0.13 of Group Ib shows there is no significant difference compared Group Ia and Group Ib with other groups. P value of 0.04, 0.03, 0.03, 0.02 of Group Iia, Group Iib, Group IIIa, Group III b respectively shows there is no significant difference between each other.

Comparison of mean Ti release of T1, on comparison with P value of 0.04 shows there is statistically significant difference when compared Group Ia with all other groups; P value of 0.04 shows there is statistically significant difference when compared Group Ib with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIa with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIb with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIa with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIb with all other groups.

Table-13: Comparison of mean Ti and Ni release values between the groups

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-Ia	0.25±0.005		0.13±0.008	
Group-Ib	0.25±0.009	0.17	0.14±0.008	0.19
Group-IIa	0.28±0.006*.#	0.04	0.16±0.007*.#	0.04
Group-IIb	0.31±0.005*.#,\$	0.03	0.17±0.007*.#,\$	0.04
Group-IIIa	0.31±0.005*.#,\$	0.03	0.21±0.0095*.#,\$,&	0.03
Group-IIIb	0.37±0.005*.#,\$,&,	0.02	0.23±0.001*.#,\$,&,	0.03

On comparison of Ni release at T2, P value of Group Ia and Group Ib is 0.14 and 0.17 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T2, P value of Group Ia and Group Ib is 0.21 and 0.19 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.03 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

Table-14: Comparison of mean Ti and Ni release values between the groups

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-Ia	0.28±0.006		0.14±0.005	
Group-Ib	0.28±0.006	0.13	0.15±0.005	0.14
Group-IIa	0.31±0.005* [#]	0.04	0.17±0.008* [#]	0.04
Group-IIb	0.34±0.001* ^{#, \$}	0.04	0.18±0.008* [#]	0.04
Group-IIIa	0.37±0.005* ^{#, \$, &}	0.03	0.28±0.009* ^{#, \$, &}	0.02
Group-IIIb	0.44±0.10* ^{#, \$, &, I}	0.02	0.29±0.006* ^{#, \$, &}	0.02

On comparison of Ni release at T3, P value of Group Ia and Group Ib is 0.14 and 0.13 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T3, P value of Group Ia and Group Ib is 0.16 and 0.19 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

Table-15: Comparison of mean Ti and Ni release values between the groups

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.31±0.008	0.11	0.16±0.007	0.16
Group-IB	0.32±0.009	0.12	0.17±0.008	0.14
Group-IIA	0.34±0.007	0.04*	0.20±0.007	0.04*
Group-IIB	0.37±0.008	0.03*	0.21±0.007	0.03*
Group-IIIA	0.44±0.001	0.02*	0.30±0.007	0.02*
Group-IIIB	0.50±0.10	0.02*	0.32±0.10	0.02*

(*p<0.05 significant)

On comparison of Ni release at T4, P value of Group Ia and Group Ib is 0.11 and 0.12 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T4, P value of Group Ia and Group Ib is 0.16 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

Table-16: Comparison of mean Ni release values between the groups at same time period of Group-IA and Ib

Time	Ni		p value
	Group-IA (MEAN±SD)	Group-IB (MEAN±SD)	
T1 (1 hour)	0.21±0.004	0.21±0.004	0.34
T2 (24 hours)	0.25±0.005	0.25±0.009	0.48
T3 (1 week)	0.28±0.006	0.28±0.006	0.12
T4 (3 weeks)	0.31±0.008	0.32±0.009	0.45

When comparing Ni release of Group Ia and Group Ib, P value at T1 is 0.34, T2 is 0.48, T3 is 0.12, T4 is 0.45 respectively shows there is not statistically significant difference between groups.

Table-17: Comparison of mean Ni release values between the groups at same time period of Group-IIA and IIB

Time	Ni		p value
	Group-IIA (MEAN±SD)	Group-IIB (MEAN±SD)	
T1 (1 hour)	0.24±0.005	0.25±0.009	0.17
T2 (24 hours)	0.28±0.006	0.31±0.005	0.04*
T3 (1 week)	0.31±0.005	0.34±0.001	0.04*
T4 (3 weeks)	0.34±0.007	0.37±0.008	0.04*

(*p>0.05 significant)

When comparing Ni release of Group IIA and Group IIB, P value at T1 is 0.17, shows there is not statistically significant difference between groups. T2 is 0.04, T3 is 0.04, T4 is 0.04 respectively shows there is statistically significant difference between groups

Table-18: Comparison of mean Ni release values between the groups at same time period of Group-IIIa and IIIb

Time	Ni		p value
	Group-IIIa (MEAN±SD)	Group-IIIb (MEAN±SD)	
T1 (1 hour)	0.25±0.009	0.28±0.006	0.04*
T2 (24 hours)	0.31±0.005	0.37±0.005	0.03*
T3 (1 week)	0.37±0.005	0.44±0.10	0.03*
T4 (3 weeks)	0.44±0.001	0.50±0.10	0.03*

(*p>0.05 significant)

When comparing Ni release of Group IIIa and Group IIIb, P value at T1 is 0.4, T2 is 0.3, T3 is 0.3, T4 is 0.3 respectively shows there is statistically significant difference between groups.

Table-19: Comparison of mean Ti release values between the groups at same time period of Group-IA and Ib

Time	Ti		p value
	Group-IA (MEAN±SD)	Group-IB (MEAN±SD)	
T1 (1 hour)	0.11±0.006	0.13±0.008	0.14
T2 (24 hours)	0.13±0.008	0.14±0.008	0.12
T3 (1 week)	0.14±0.005	0.15±0.005	0.26
T4 (3 weeks)	0.16±0.007	0.17±0.008	0.31

When comparing Ti release of Group Ia and Group Ib, P value at T1 is 0.14, T2 is 0.12, T3 is 0.26, T4 is 0.31 respectively shows there is no statistically significant difference between groups.

Table-20: Comparison of mean Ti release values between the groups at same time period of Group-IIA and IIB

Time	Ti		p value
	Group-IIA (MEAN±SD)	Group-IIB (MEAN±SD)	
T1 (1 hour)	0.14±0.007	0.15±0.007	0.34
T2 (24 hours)	0.16±0.007	0.17±0.007	0.28
T3 (1 week)	0.17±0.008	0.18±0.008	0.17
T4 (3 weeks)	0.20±0.007	0.21±0.007	0.16

When comparing Ti release of Group IIA and Group IIB, P value at T1 is 0.34, T2 is 0.28, T3 is 0.17, T4 is 0.16 respectively shows there is no statistically significant difference between groups.

Table-21: Comparison of mean Ti release values between the groups at same time period of Group-IIIA and IIIB

Time	Ti		p value
	Group-IIIA (MEAN±SD)	Group-IIIB (MEAN±SD)	
T1 (1 hour)	0.18±0.008	0.21±0.10	0.12
T2 (24 hours)	0.21±0.009	0.23±0.001	0.16
T3 (1 week)	0.28±0.009	0.29±0.006	0.18
T4 (3 weeks)	0.30±0.007	0.32±0.10	0.32

When comparing Ti release of Group IIIa and Group IIIb, P value at T1 is 0.12, T2 is 0.16, T3 is 0.18, T4 is 0.32 respectively shows there is no statistically significant difference between groups

Table-22: Comparison of mean Ti and Ni release values between the groups at T1 time period

Groups	Ni	p value	Ti	p value
Group-IA	0.21±0.004	0.12	0.11±0.006	0.04*
Group-IB	0.21±0.004	0.13	0.13±0.008	0.04*
Group-IIA	0.24±0.005	0.04*	0.14±0.007	0.03*
Group-IIB	0.25±0.009	0.03*	0.15±0.007	0.03*
Group-IIIA	0.25±0.009	0.03*	0.18±0.008	0.02*
Group-IIIB	0.28±0.006	0.02*	0.28±0.006	0.02*

(*p<0.05 significant compared)

When comparing mean Ni and Ti release at different immersion time at T1, on comparison of P value 0.12 of Group Ia and P value 0.13 of Group Ib shows there is no significant difference compared Group Ia and Group Ib with other groups. P value of 0.04, 0.03, 0.03, 0.02 of Group Iia, Group Iib, Group IIIa, Group III b respectively shows there is no significant difference between each other

Comparison of mean Ti release of T1 ,on comparison with P value of 0.04 shows there is statistically significant difference when compared Group Ia with all other groups; P value of 0.04 shows there is statistically significant difference when compared Group Ib with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group Iia with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group Iib with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIa with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIb with all other groups.

Table-23: Comparison of mean Ti and Ni release values between the groups at T2 time period

Groups	Ni	p value	Ti	p value
Group-IA	0.25±0.005	0.14	0.13±0.008	0.17
Group-IB	0.25±0.009	0.17	0.14±0.008	0.19
Group-IIA	0.28±0.006	0.04*	0.16±0.007	0.04*
Group-IIB	0.31±0.005	0.03*	0.17±0.007	0.04*
Group-IIIA	0.31±0.005	0.03*	0.21±0.009	0.03*
Group-IIIB	0.37±0.005	0.02*	0.23±0.001	0.03*

(*p<0.05 significant compared)

On comparison of Ni release at T2, P value of Group Ia and Group Ib is 0.14 and 0.17 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.03, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T2, P value of Group Ia and Group Ib is 0.17 and 0.19 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.03 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

Table-24: Comparison of mean Ti and Ni release values between the groups at T3 time period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.28±0.006	0.11	0.14±0.005	0.13
Group-IB	0.28±0.006	0.13	0.15±0.005	0.14
Group-IIA	0.31±0.005	0.04*	0.17±0.008	0.04*
Group-IIB	0.34±0.001	0.04*	0.18±0.008	0.04*
Group-IIIA	0.37±0.005	0.03*	0.28±0.009	0.02*
Group-IIIB	0.44±0.10	0.02*	0.29±0.006	0.02*

(*p<0.05 significant)

On comparison of Ni release at T3, P value of Group Ia and Group Ib is 0.11 and 0.13 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T3, P value of Group Ia and Group Ib is 0.13 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.02, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

Table-25: Comparison of mean Ti and Ni release values between the groups at T4 time period

Groups	Ni (Mean±SD)	p value	Ti (Mean±SD)	p value
Group-IA	0.31±0.008	0.11	0.16±0.007	0.13
Group-IB	0.32±0.009	0.12	0.17±0.008	0.14
Group-IIA	0.34±0.007	0.04*	0.20±0.007	0.04*
Group-IIB	0.37±0.008	0.03*	0.21±0.007	0.03*
Group-IIIA	0.44±0.001	0.02*	0.30±0.007	0.02*
Group-IIIB	0.50±0.10	0.02*	0.32±0.10	0.02*

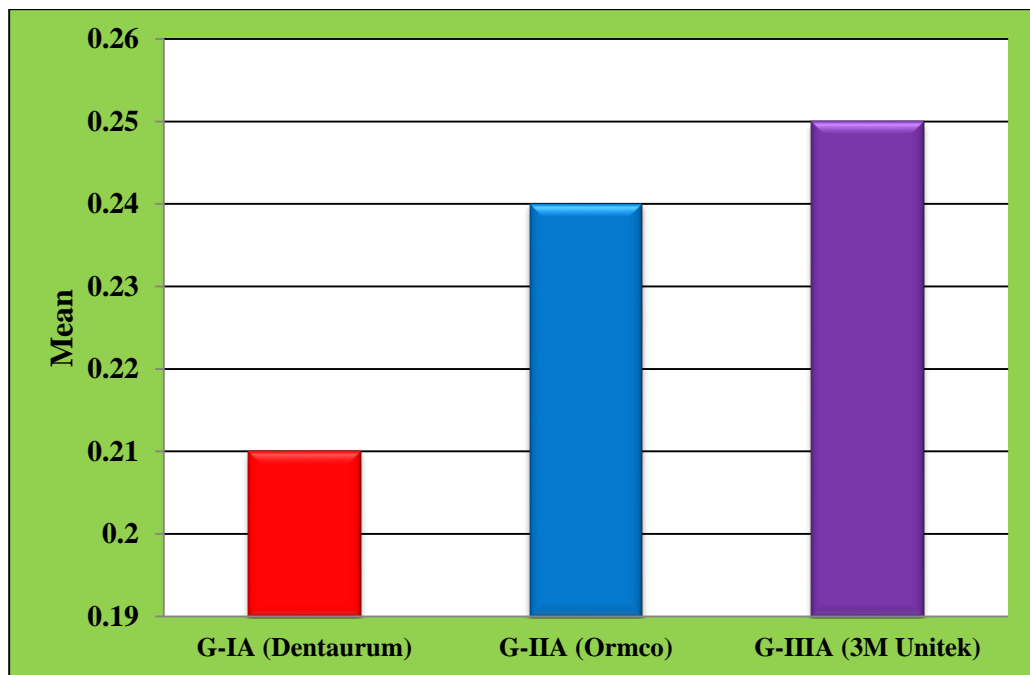
(*p<0.05 significant)

On comparison of Ni release at T4, P value of Group Ia and Group Ib is 0.11 and 0.12 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

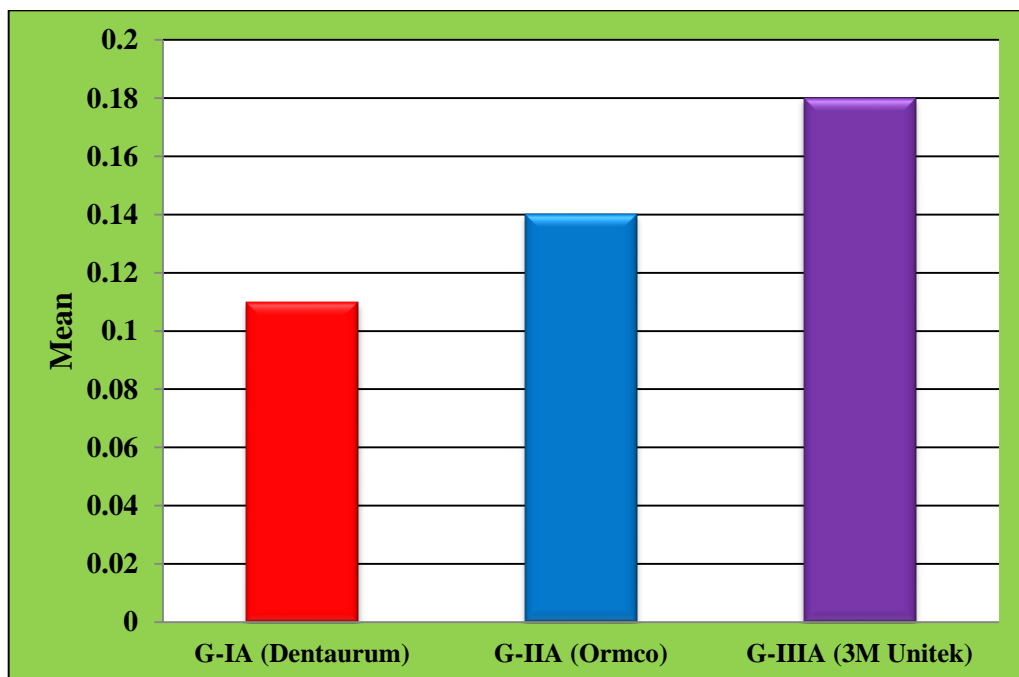
On comparison of Ti release at T4, P value of Group Ia and Group Ib is 0.13 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group Iia, Group Iib, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

GRAPHS

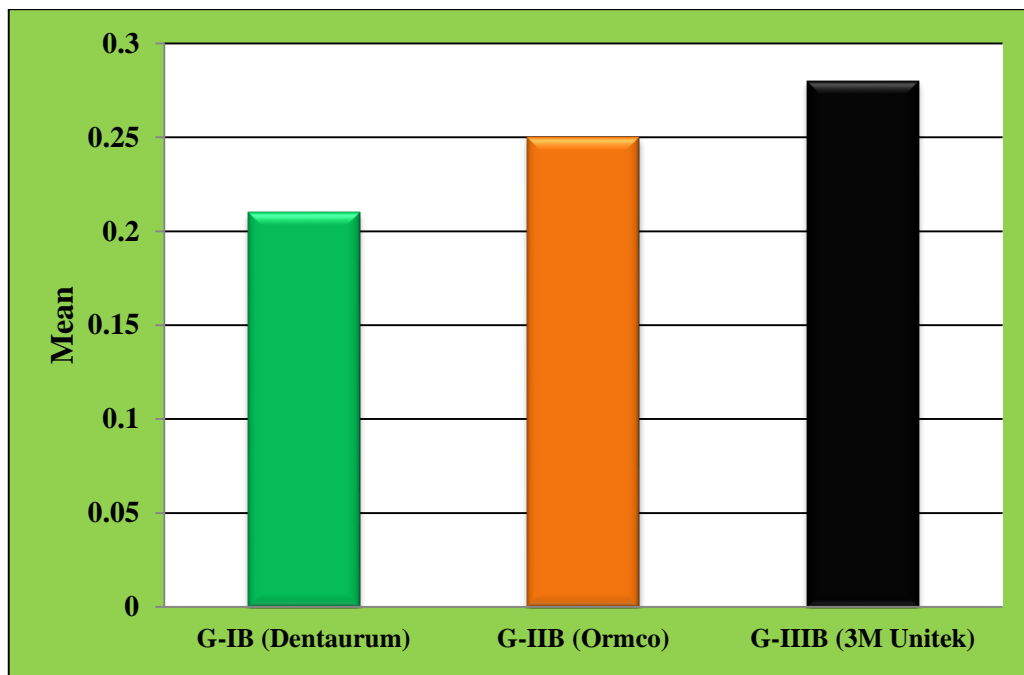
Graph-1: Comparison of mean Ni and Ti release values of three different round wires at T1 Period



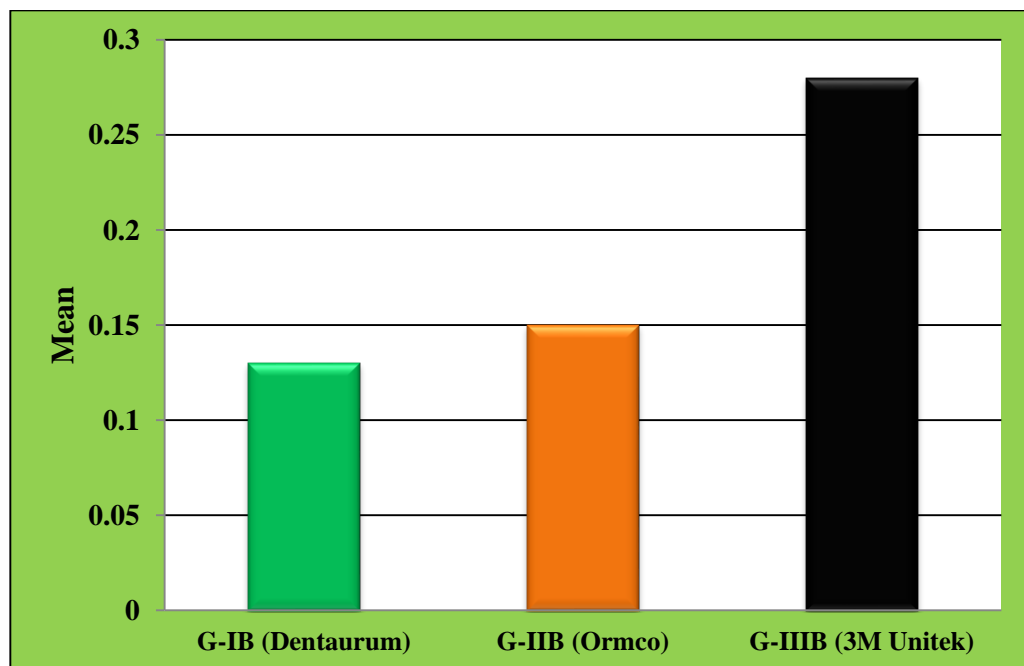
Graph-2: Comparison of mean Ni release values of three different round wires at T1 Period



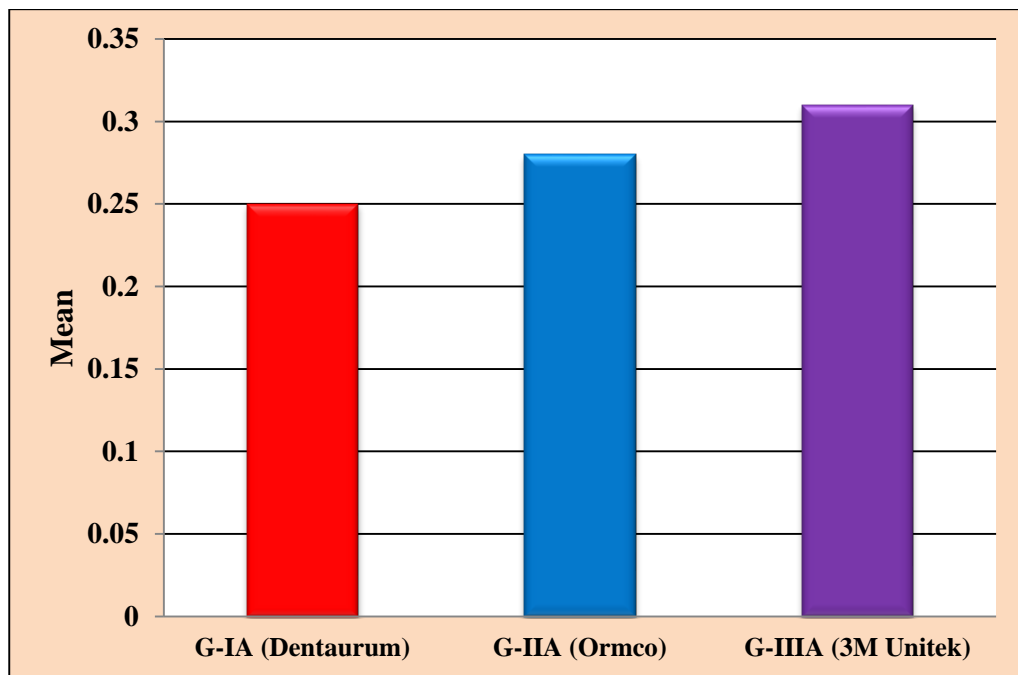
Graph-3: Comparison of mean Ni release values of three different rectangular wires at T1 Period



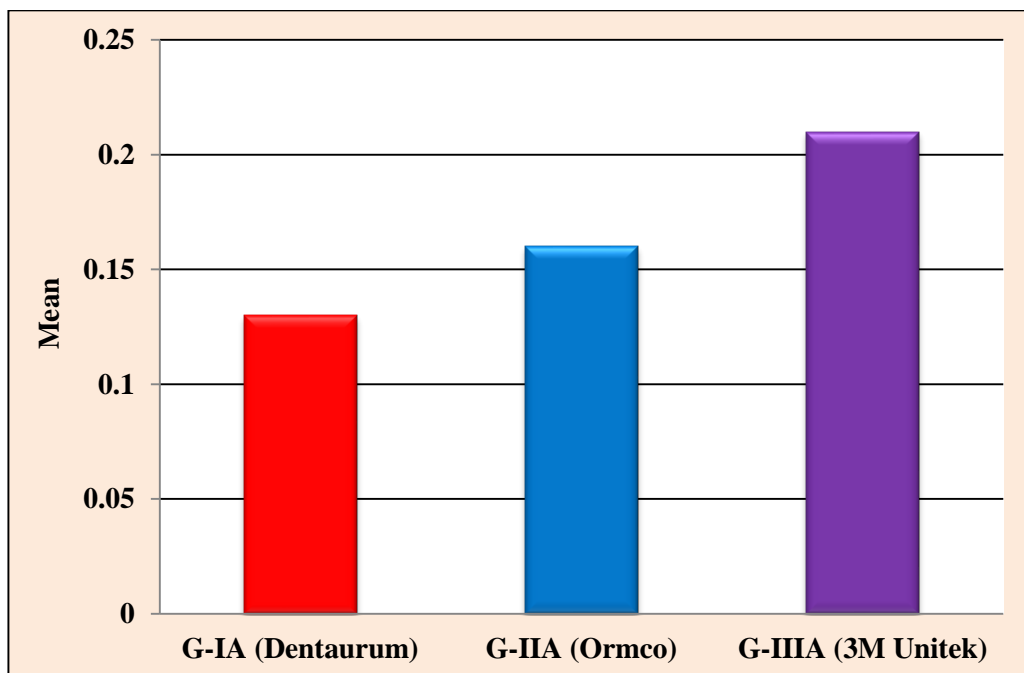
Graph-4: Comparison of mean Ti release values of three different rectangular wires at T1 Period



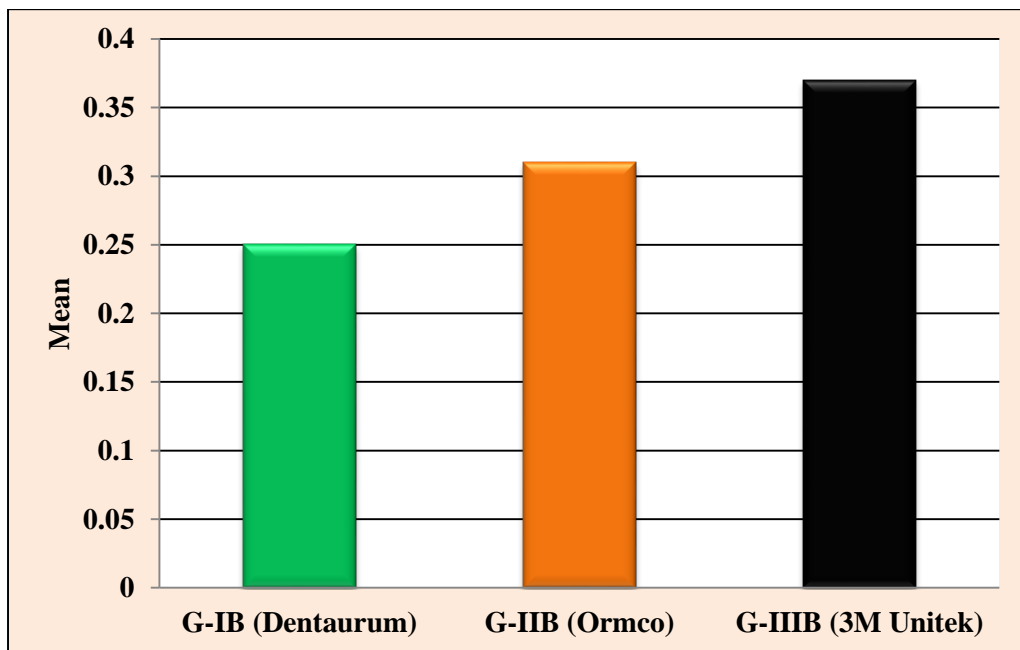
Graph-5: Comparison of mean Ni release values of three different round wires at T2 Period



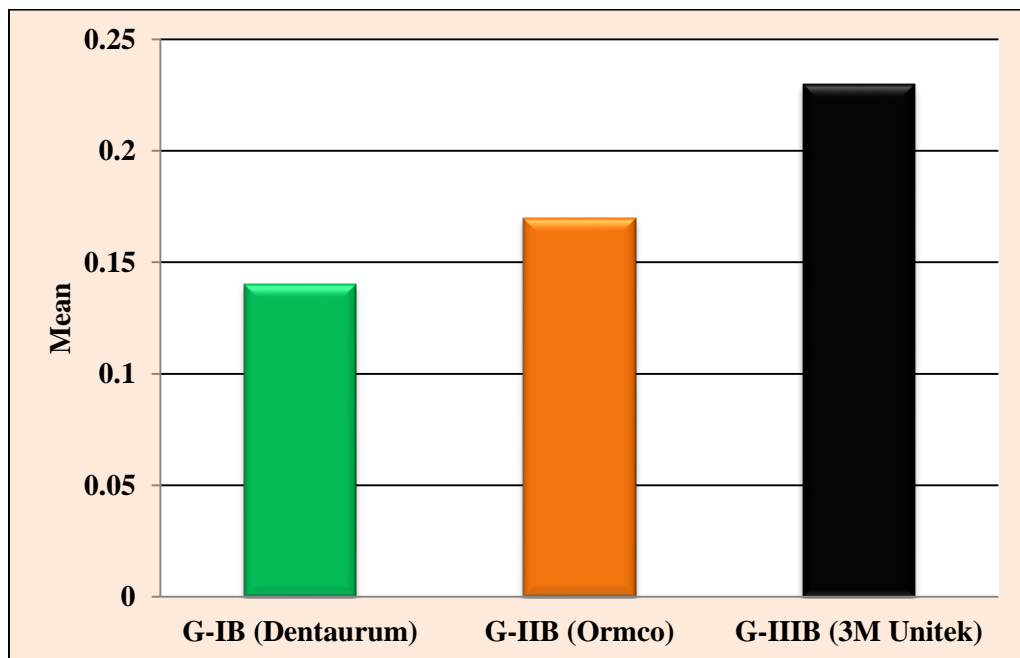
Graph-6: Comparison of mean Ti release values of three different round wires at T2 Period



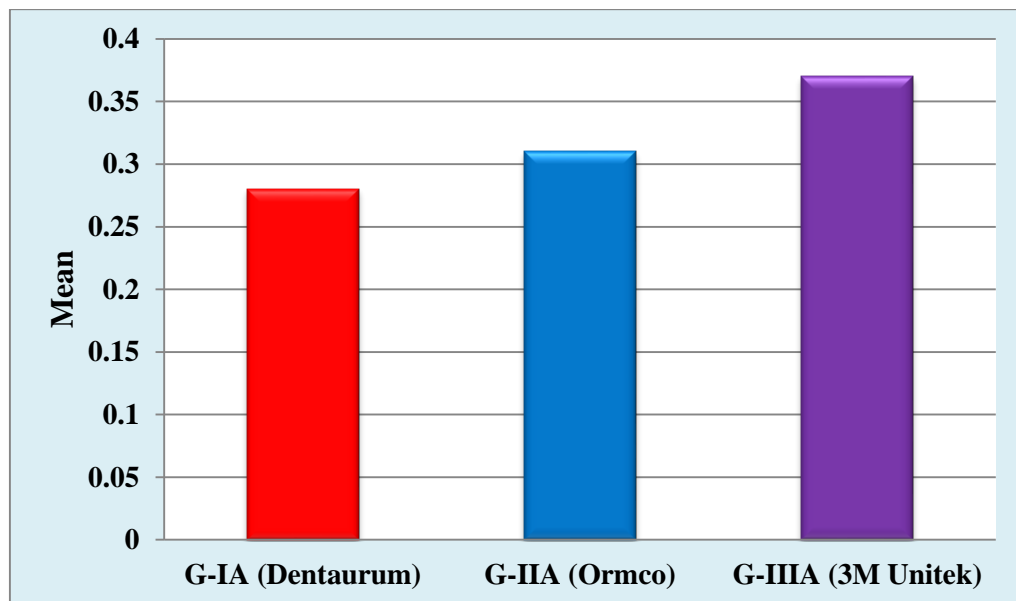
Graph-7: Comparison of mean Ti release values of three different rectangular wires at T2 Period



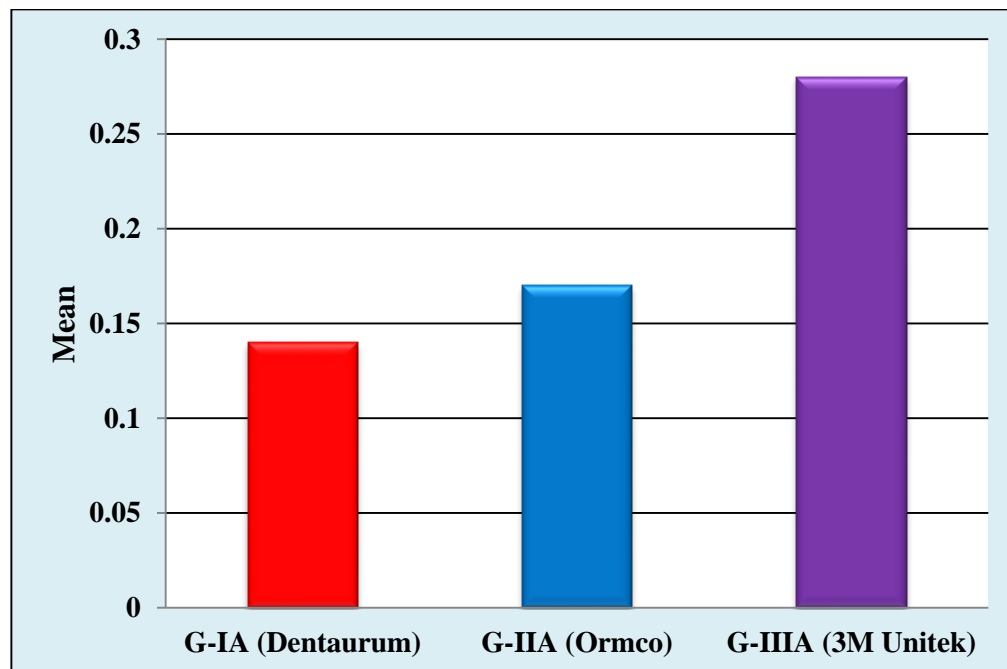
Graph-8: Comparison of mean Ni release values of three different rectangular wires at T2 Period



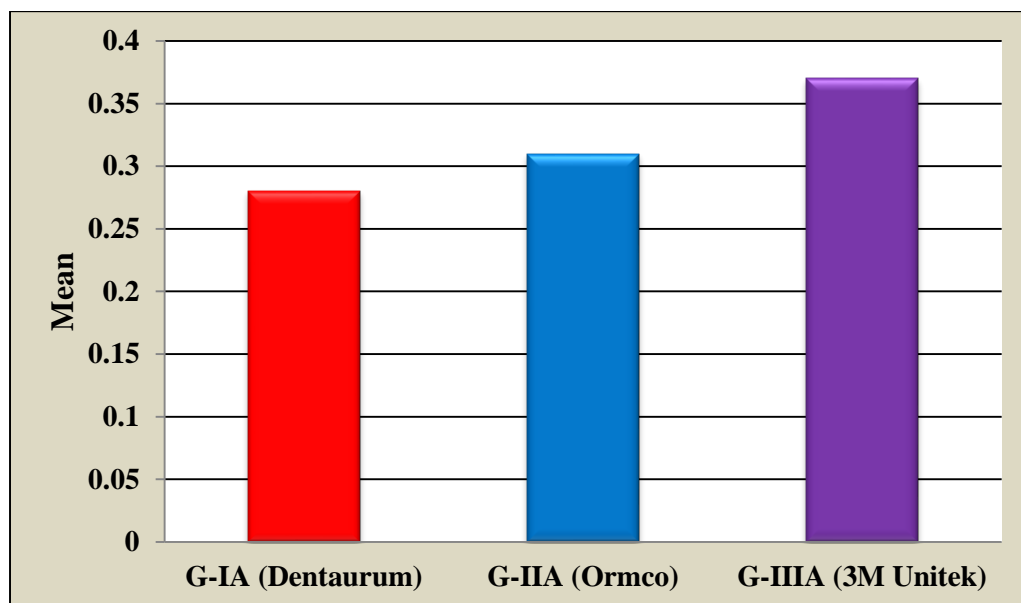
Graph-9: Comparison of mean Ni release values of three different round wires at T3 Period



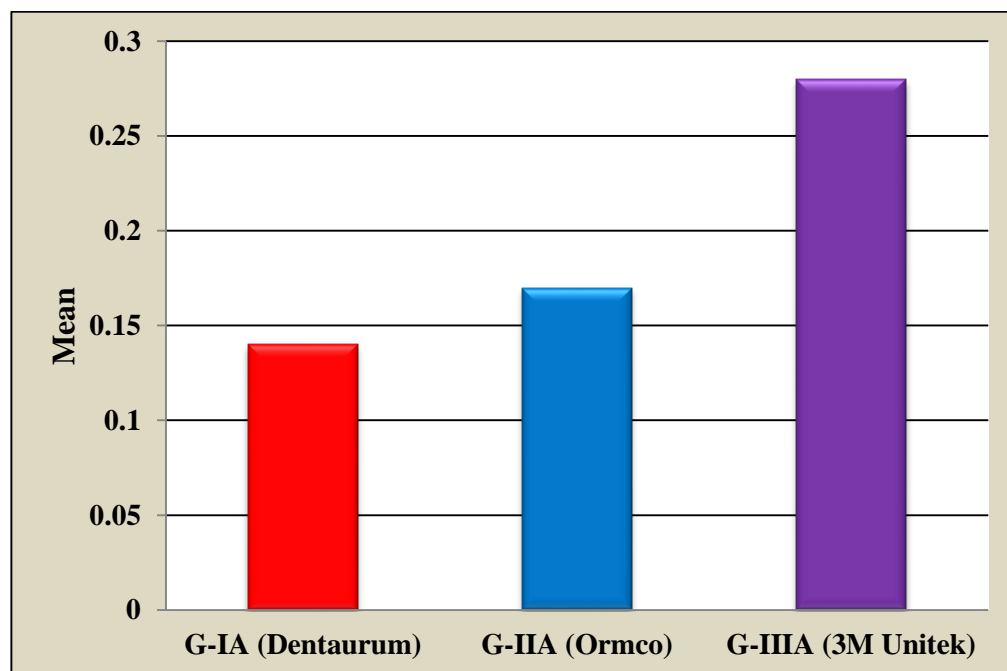
Graph-10: Comparison of mean Ni release values of three different round wires at T3 Period



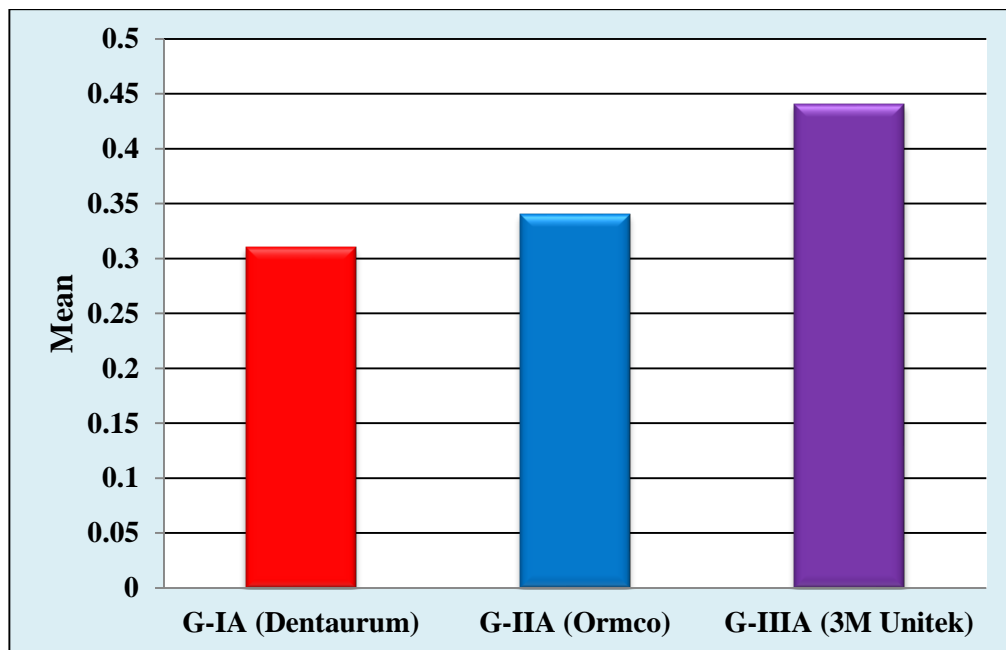
Graph-11: Comparison of mean Ti release values of three different rectangular wires at T3 Period



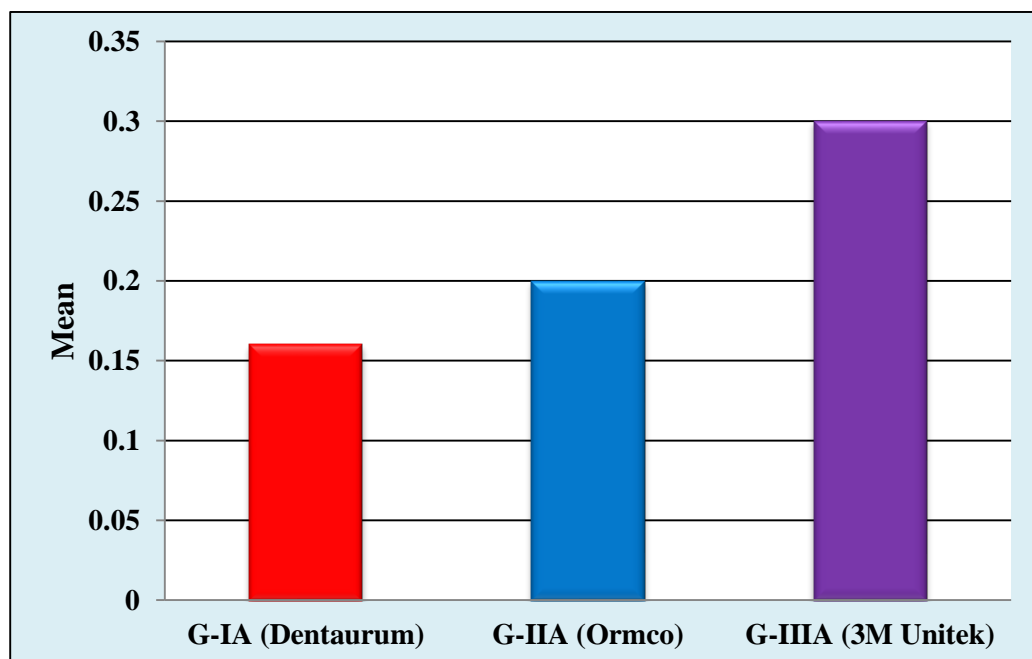
Graph-12: Comparison of mean Ni release values of three different rectangular wires at T3 Period



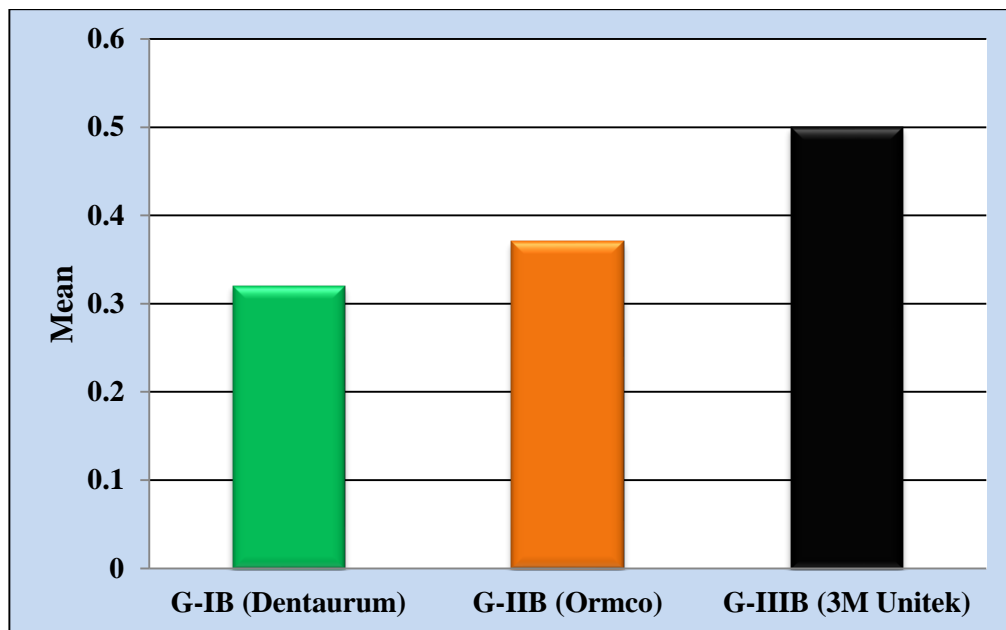
Graph-13: Comparison of mean Ti release values of three different round wires at T4 Period



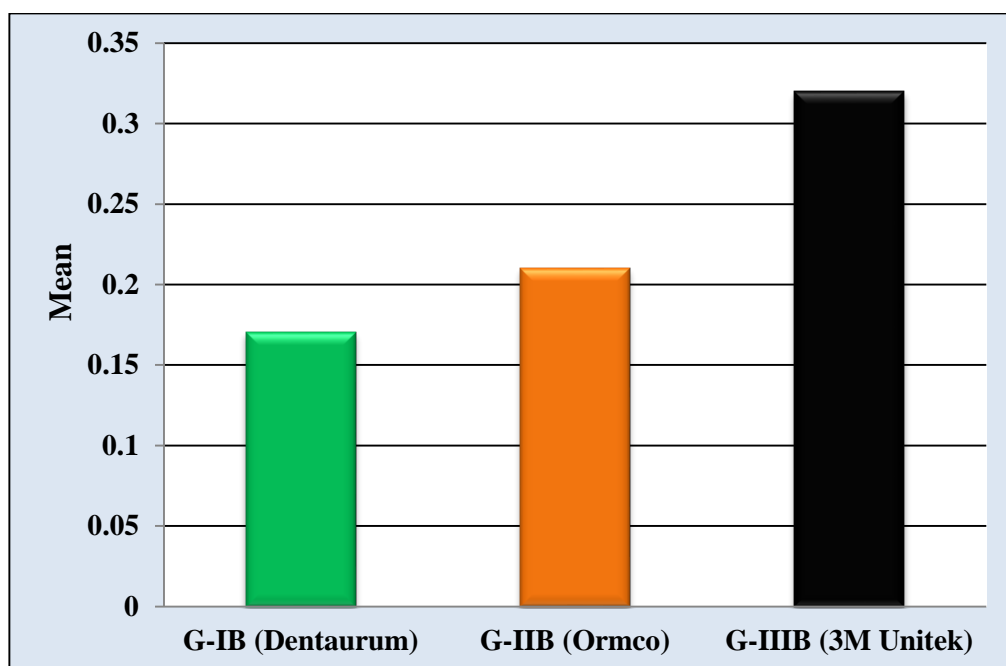
Graph-14: Comparison of mean Ni release values of three different round wires at T4 Period



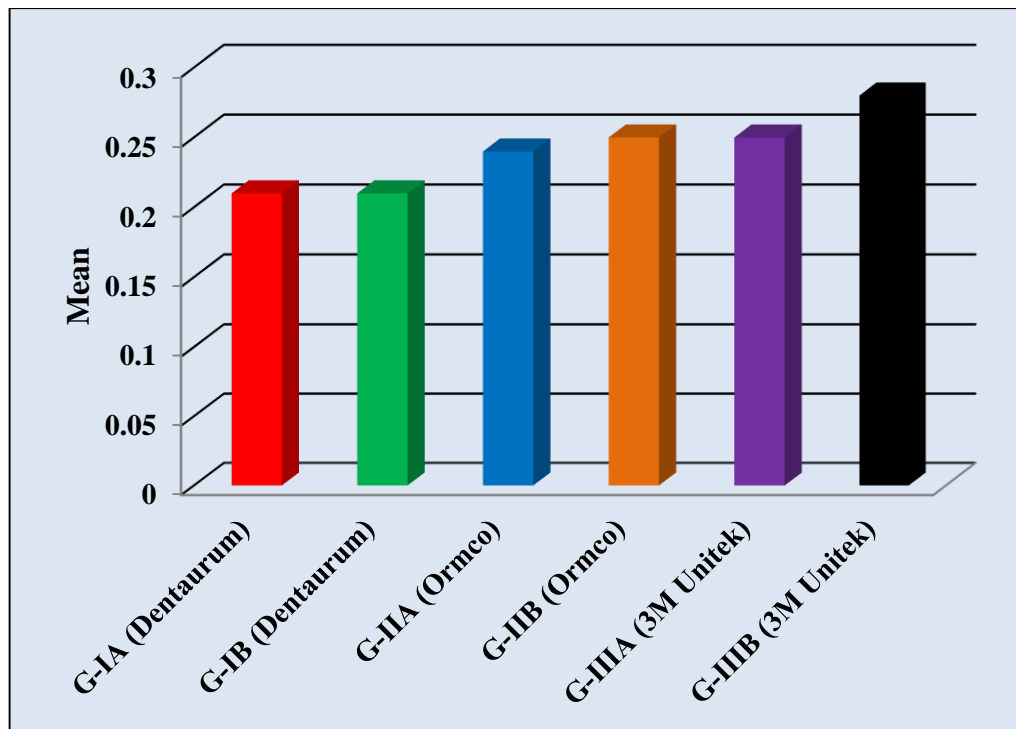
Graph-15: Comparison of mean Ti release values of three different rectangular wires at T4 Period



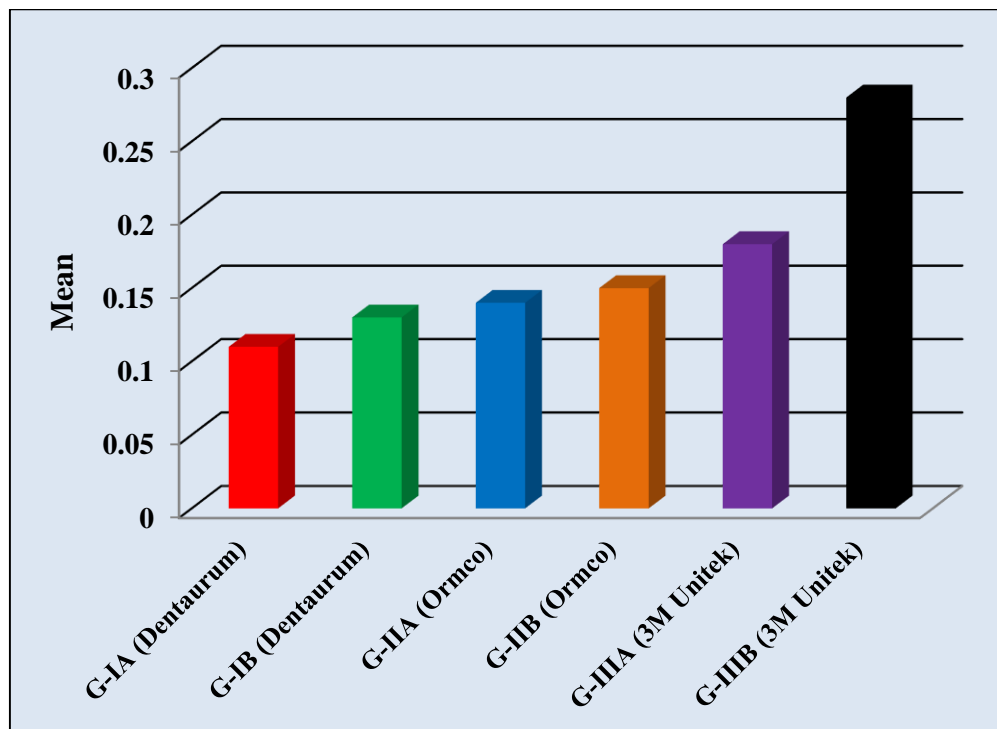
Graph-16: Comparison of mean Ni release values of three different rectangular wires at T4 Period



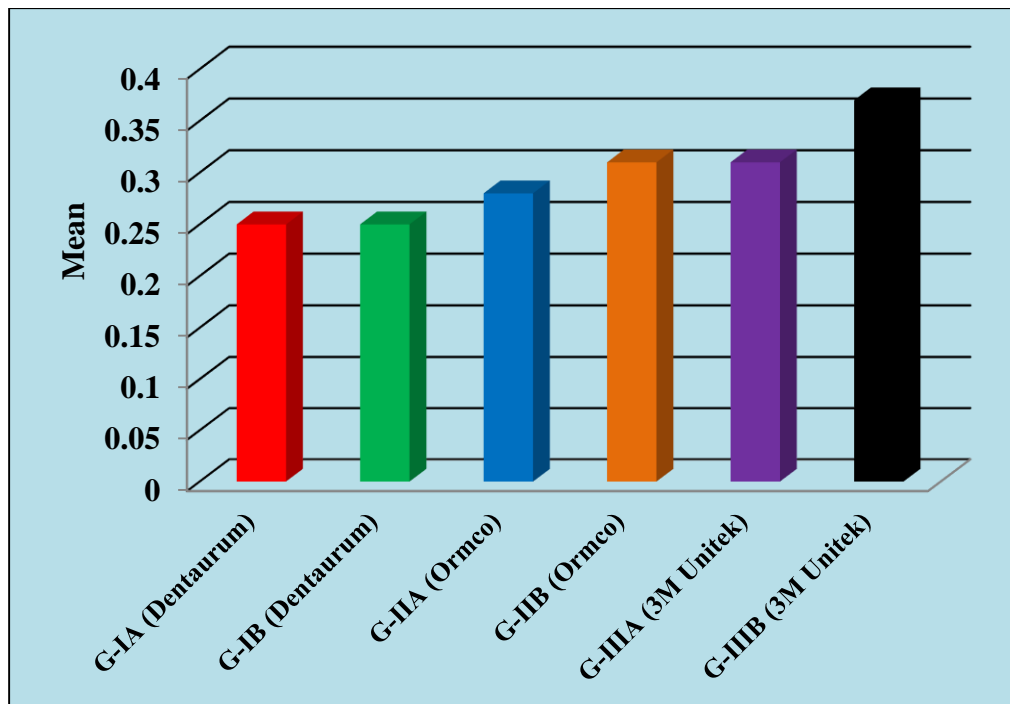
Graph-17: Comparison of mean Ni release values between the groups



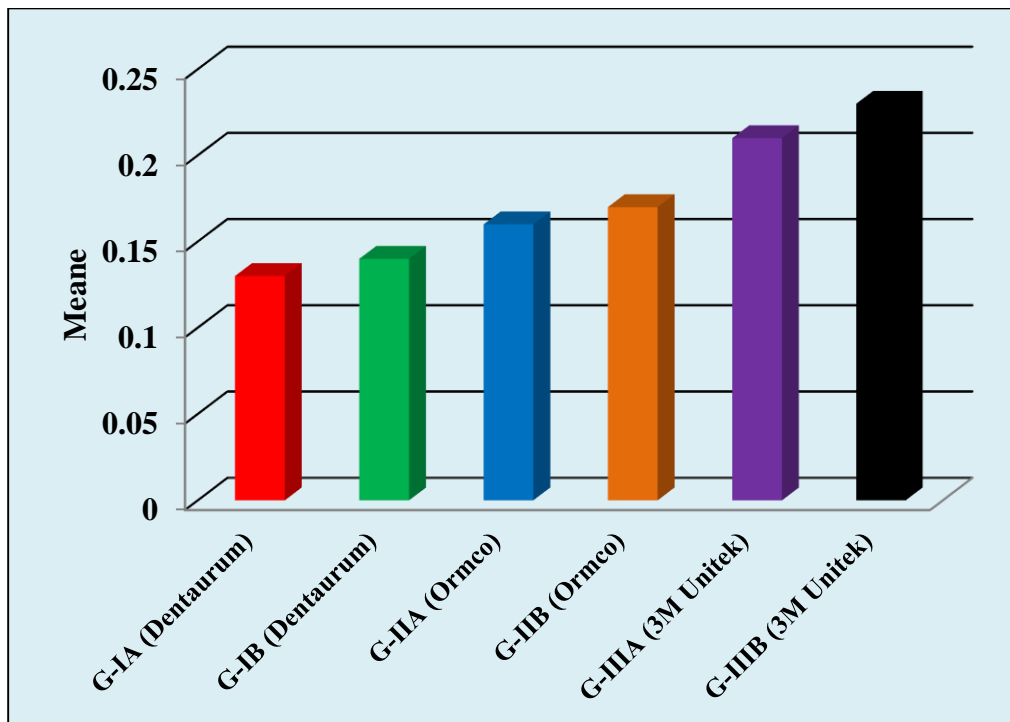
Graph-18: Comparison of mean Ti release values between the groups



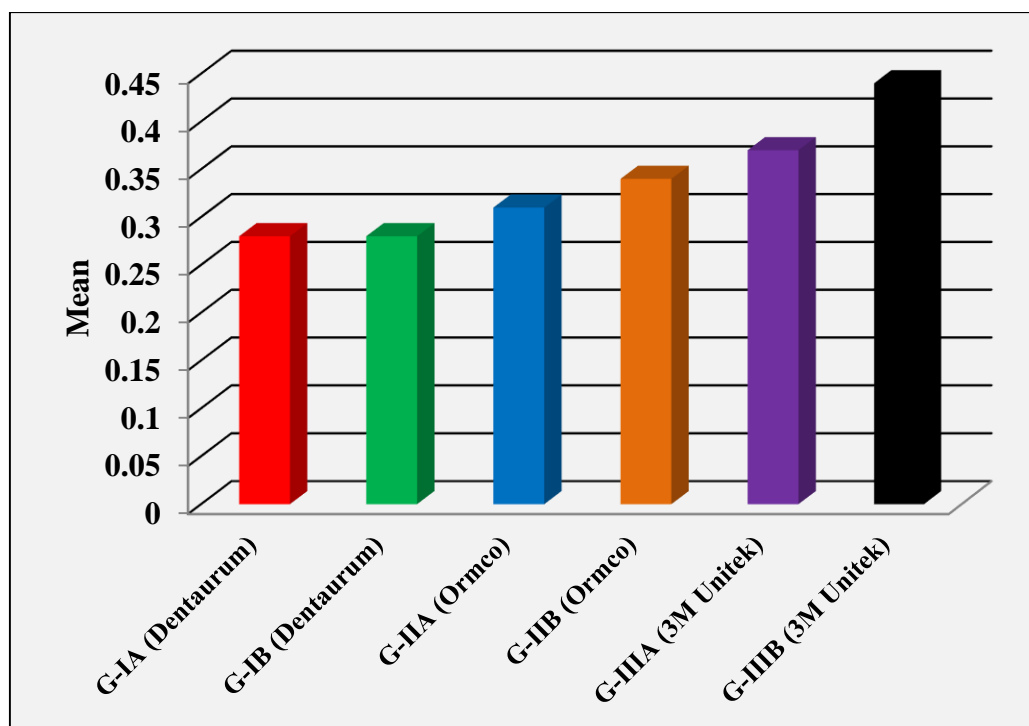
Graph-19: Comparison of mean Ni release values between the groups



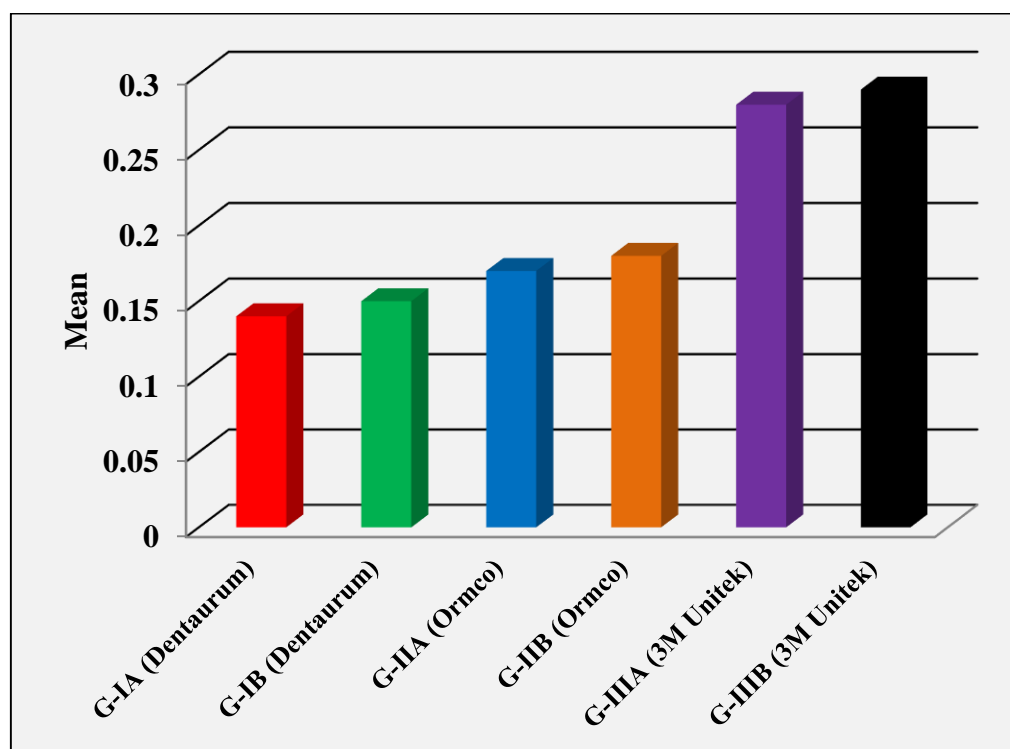
Graph-20: Comparison of mean Ti release values between the groups



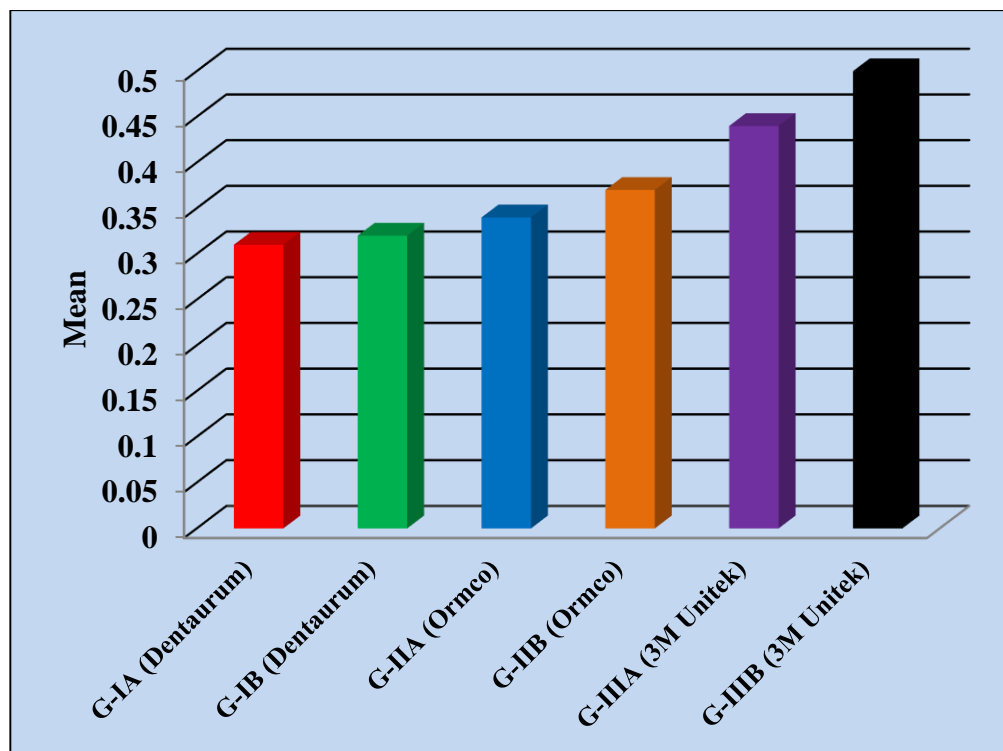
Graph-21: Comparison of mean Ni release values between the groups



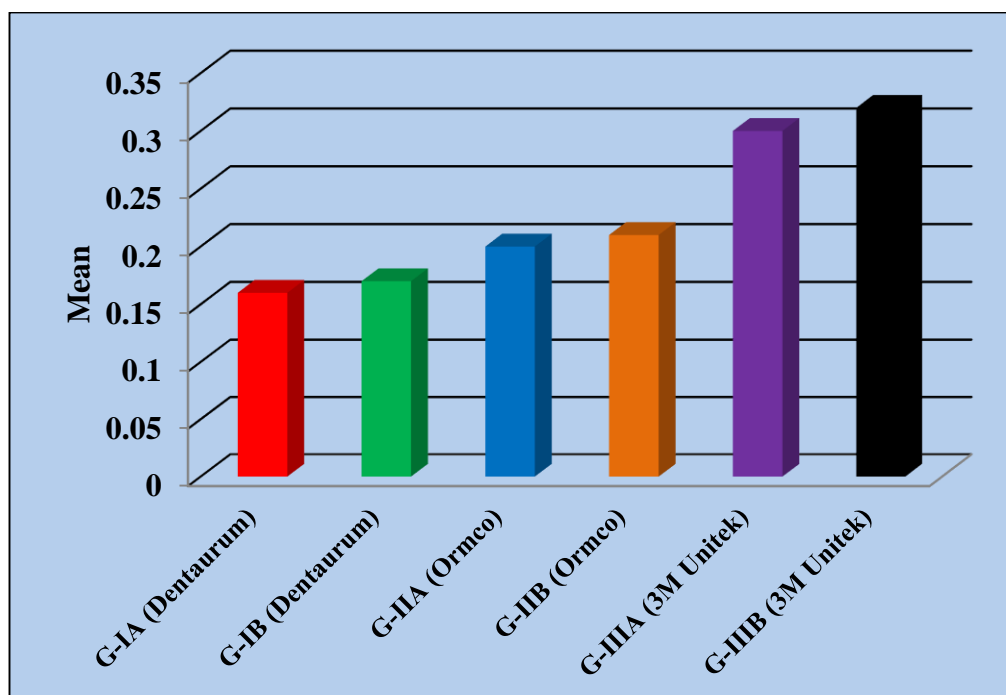
Graph-22: Comparison of mean Ti release values between the groups



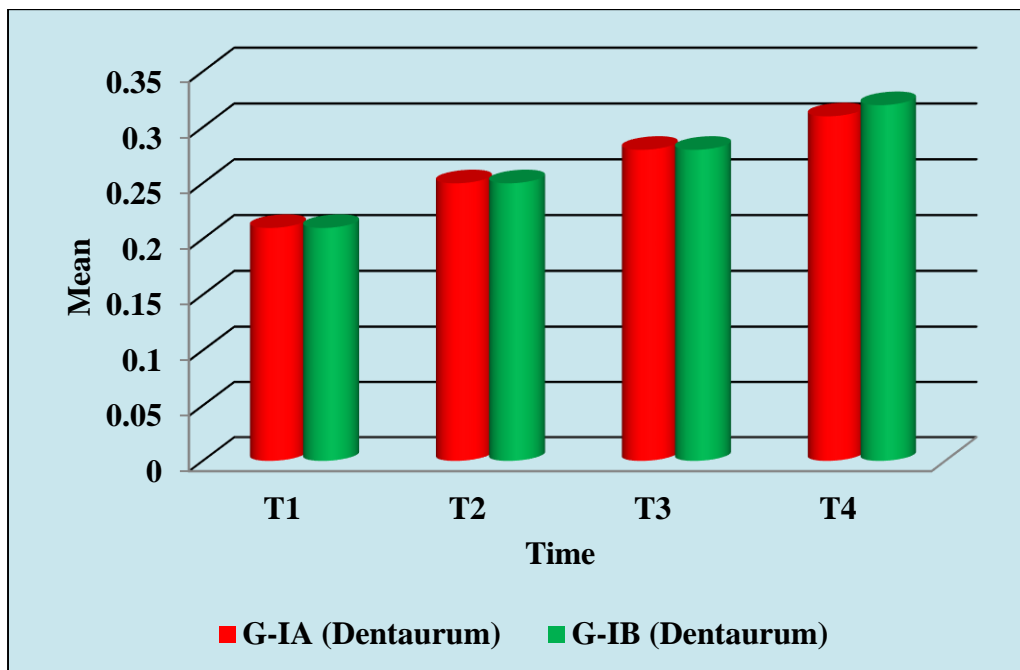
Graph-23: Comparison of mean Ni release values between the groups



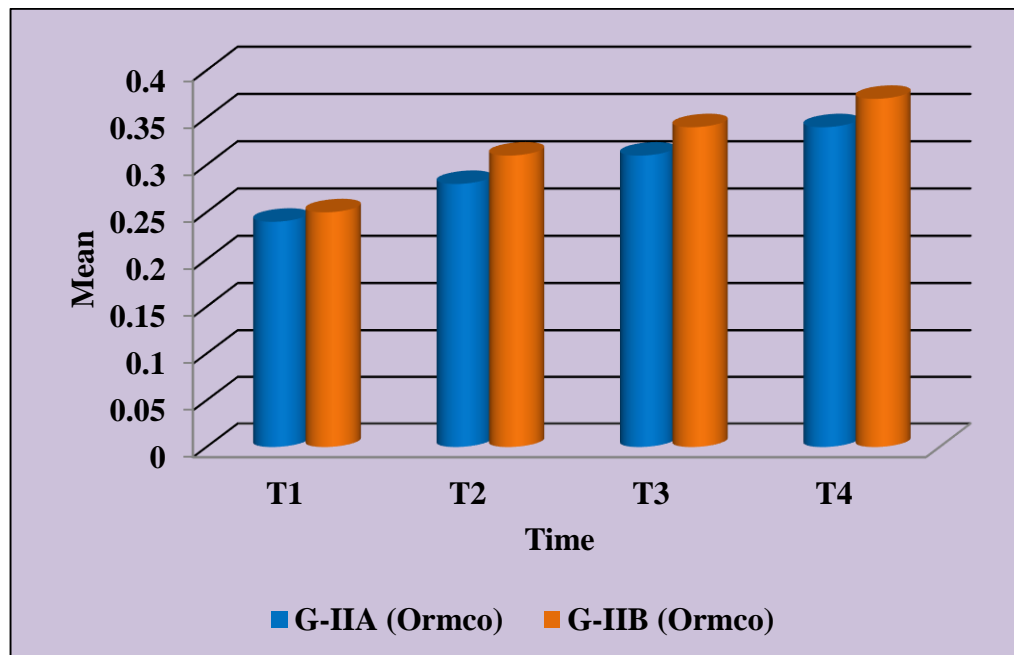
Graph-24: Comparison of mean Ti release values between the groups



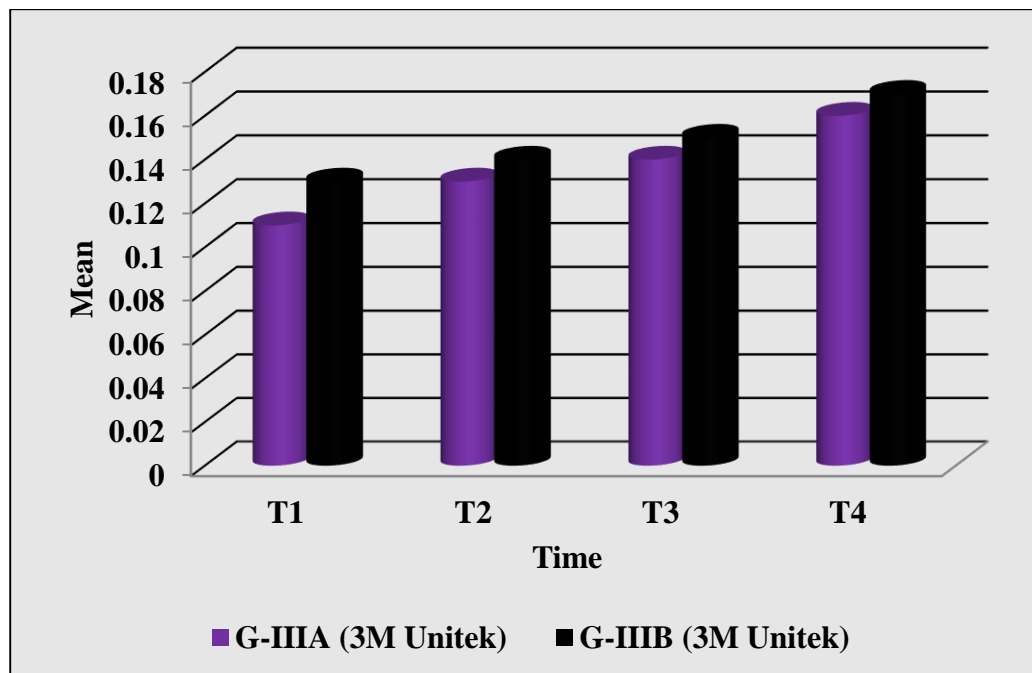
Group-25: Comparison of mean Ni release values between the groups at same time period of Group-IA and IB



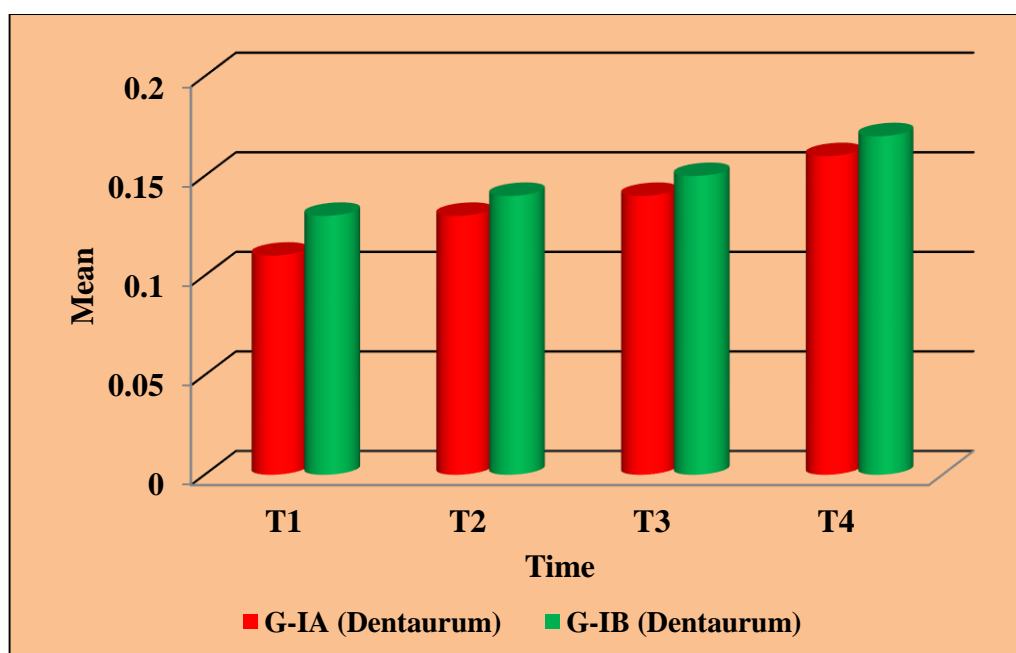
Graph-26: Comparison of mean Ni release values between the groups at same time period of Group-IIA and IIB



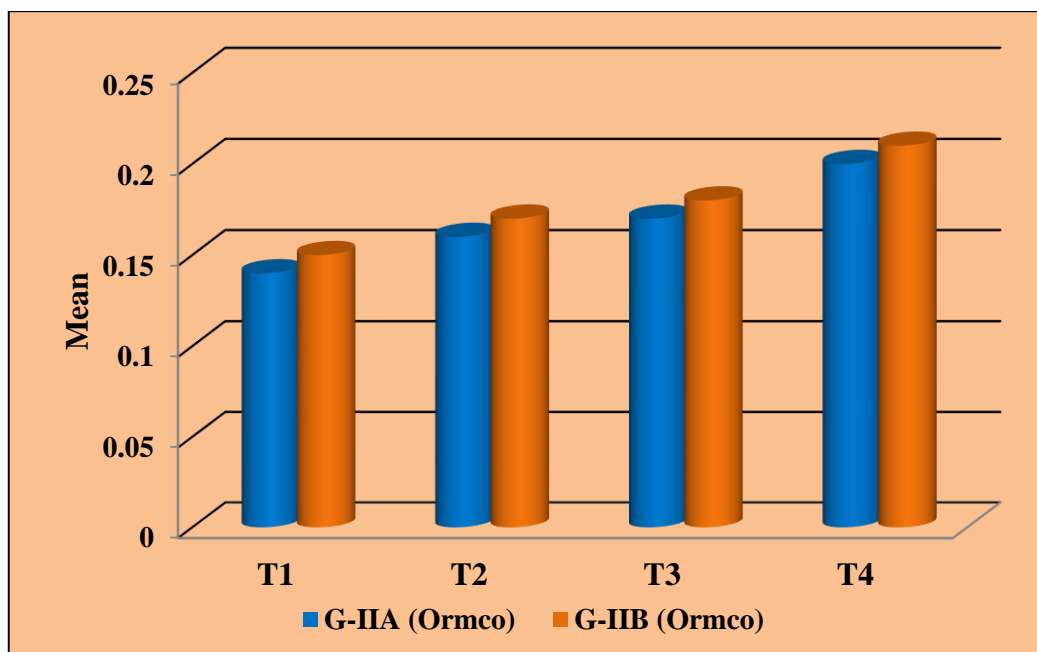
Graph-27: Comparison of mean Ni release values between the groups at same time period of Group-IIIa and IIIb



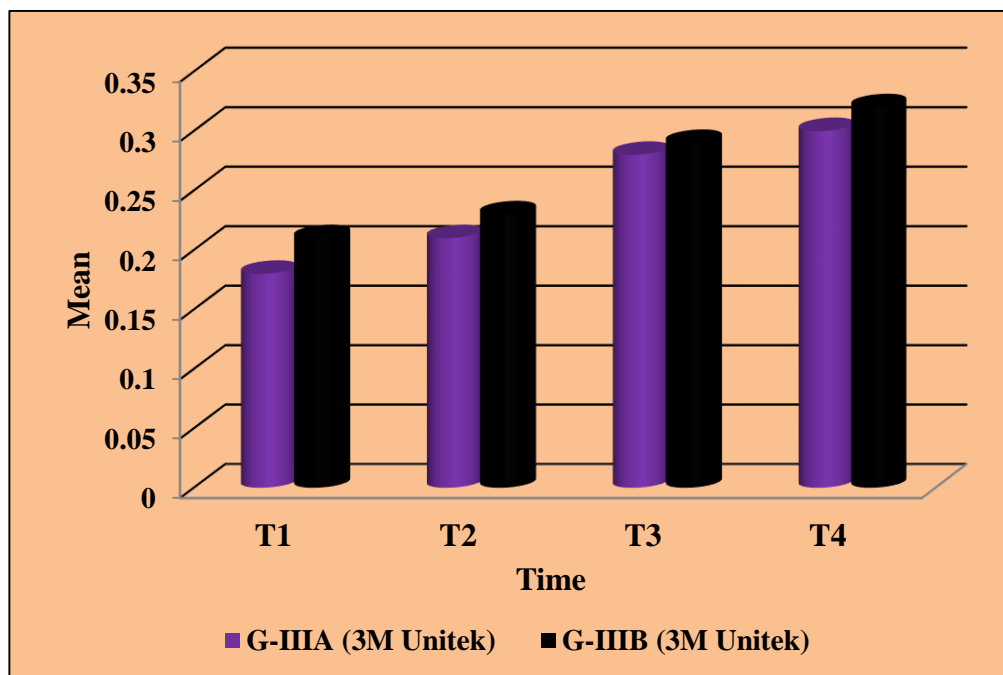
Graph-28: Comparison of mean Ti release values between the groups at same time period of Group-IA and Ib



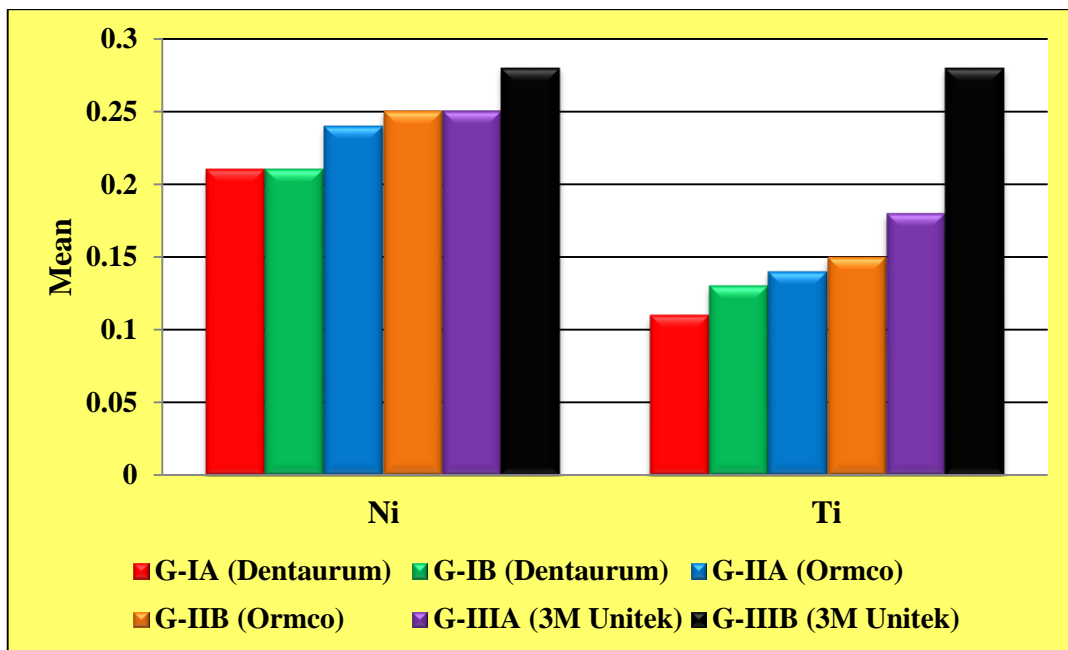
Graph-29: Comparison of mean Ti release values between the groups at same time period of Group-IIA and IIB



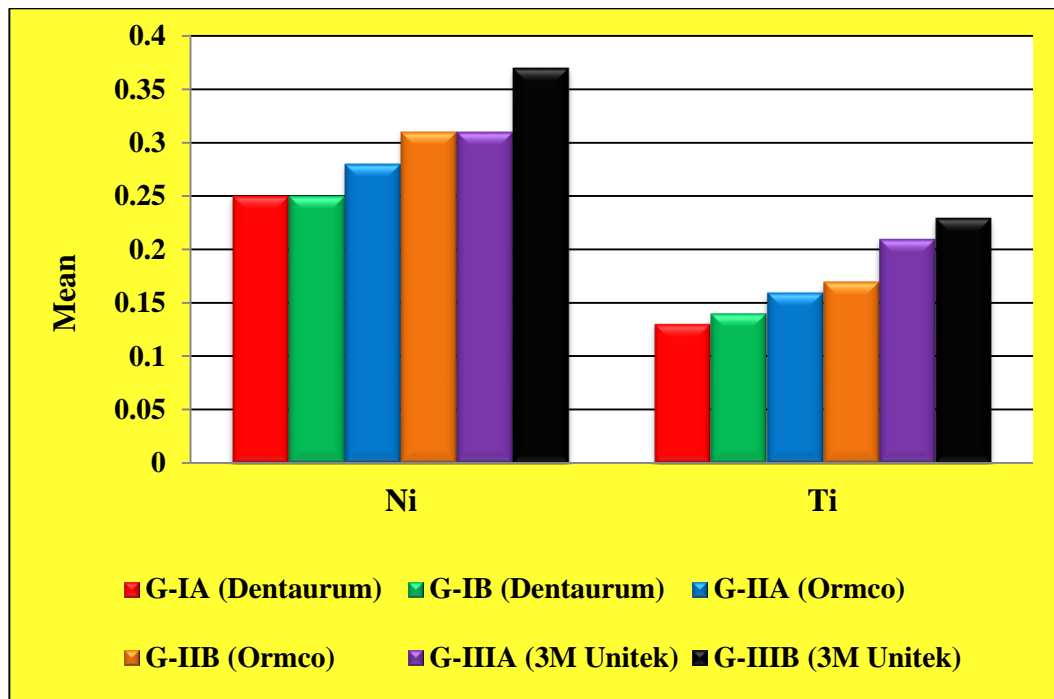
Graph-30: Comparison of mean Ti release values between the groups at same time period of Group-IIIA and IIIB



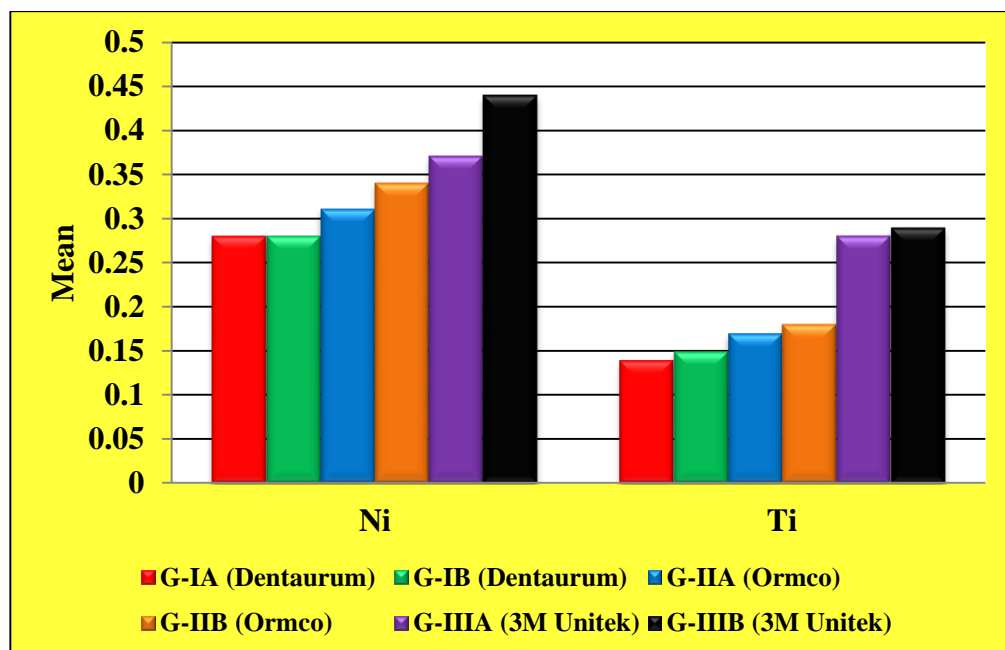
Graph-31: Comparison of mean Ti and Ni release values between the groups at T1 time period



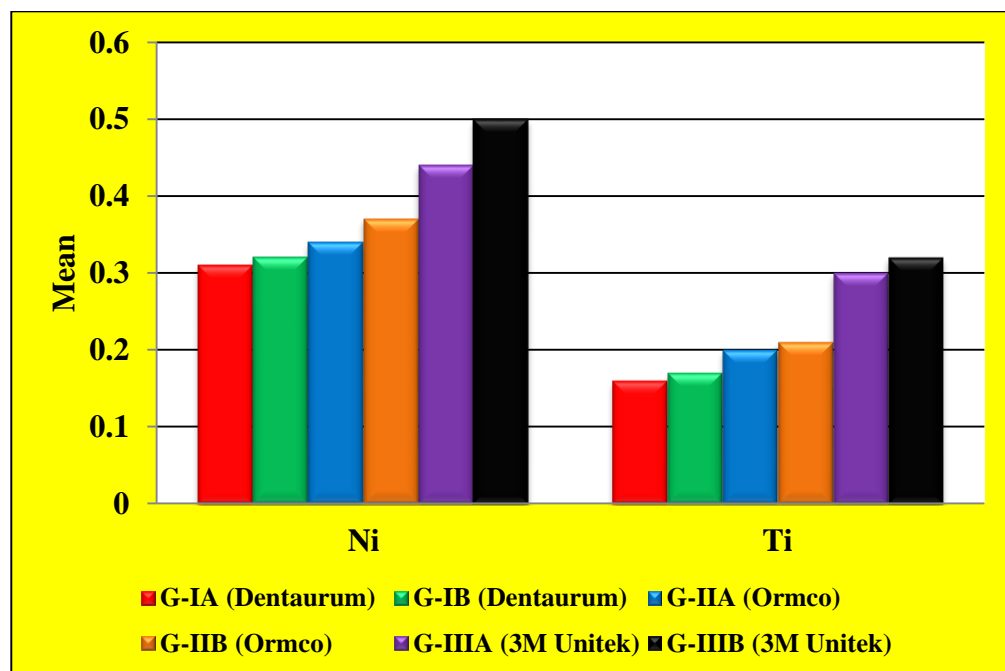
Graph-32: Comparison of mean Ti and Ni release values between the groups at T2 time period



Graph-33: Comparison of mean Ti and Ni release values between the groups at T3 time period



Graph-34: Comparison of mean Ni release values between the groups at T4 time period



DISCUSSION

DISCUSSION

The purpose of this study was to estimate the amount of nickel and titanium ions leached from NiTi wires of three different manufactures by immersion of the wires in artificial saliva. Our main goal is to find out a best wire for clinical use which has a main feature of least nickel and titanium ions leaching.

In the present study three main groups and six subgroups and two subtypes were studied for the assessment ion release from superelastic NiTi wires.

NiTi wires are the inevitable archwires in orthodontics, for its good mechanical and clinical properties¹. The major property of nickel titanium wires are their springback, which enables a wide deflection and activation range².

When using nickel titanium (NiTi) arch wire for dental orthodontic treatment, the possible danger associated with arch wire corrosion derives from the biologically harmful effects due to the released Ni ion⁵.

Nickel-titanium (NiTi) archwires contain 47%–50% Ni, which are the common source of Ni in the intraoral environment of an orthodontic patient. It has been found that several cytotoxic, allergenic and mutagenic actions to Ni in various forms and compounds are also present⁸. Studies also shown that Ni is attributed with different carcinogenic problems. There is a trend for state laws to create awareness and emphasize the necessity for patient awareness about the harmful effects of NiTi⁹.

The present study was carried out with one round NiTi wire, 0.016 superelastic NiTi wire and one rectangular, 19x25 superelastic NiTi wires of three different manufactures. These two Round and Rectangular NiTi wires are chosen

since these are the most commonly using archwires in the initial and final stages of orthodontic treatment and it was found in previous studies that the nickel ion leaching from NiTi wires are the common problem associated with fixed orthodontic therapy and corrosion of nickel and titanium ions may differ with different manufactures.

Traditionally, Round Nickel titanium (NiTi) alloy wires are used in orthodontics in the initial stages of treatment for 2-3 weeks in the oral environment because of their excellent shape memory and superelasticity, and final stage of orthodontic treatment can be done using rectangular wires^{31,32}.

The corrosive property of orthodontic wires is a significant factor determining their biocompatibility²⁶.

The corrosion of orthodontic appliances and their subsequent metal ion release in the oral environment is controlled by two main factors. Manufacturing process is the primary factor, which includes the type of alloy and the characteristics of the metals used. Second is environmental factors, such as mechanical stress, diet, time of the day, salivary flow rate, and health and psychosomatic condition of the individual^{54,69}.

According to Bernhard Schwaninger(1982) the failure of nitinol orthodontic wire is due to surface irregularities generated during manufacturing and not to the effects of corrosion³².

According to Faccioni (2003) another problem is Nickel induced DNA damage⁶⁴. In a study T Eliades (2004) also found that Ni can activate monocytes and endothelial cells and affect the expression of intercellular adhesion molecule by

endothelial cells, nontoxic concentrations of Ni may inflict direct DNA base damage and site specific DNA strand. Ni ions at nontoxic concentrations may promote microsatellite mutations and inhibit the repair of nucleotide excisions, there by contributing to genetic instability^{56,62,64}.

According to Noble J(2008)the response by the immune system to nickel include a Type IV cell mediated delayed hypersensitivity also called an allergic contact dermatitis. It is mediated by T-cells and monocytes/macrophages. The major sensitisation routes are nickel-containing jewellery and foods^{7,49,65}.

The poor corrosion resistance of a wire might not only affect the treatment effectiveness but may also result in toxic and allergic reactions due to nickel release. The wire itself has some form of protective layer,when it comes in contact with the aqueous environment⁶³. Titanium oxide (TiO₂) is present on the titanium surface, on NiTi the ultrathin film has the presence of little amounts of nickel oxide or metallic Ni,which make it more prone to chemical attack^{55,56,61}.

Surface modified NiTi arch wires showed significant improvement in corrosion resistance compared with conventional NiTi. Similarly, surface roughness values also underwent considerable modification with coating^{12,23,24}.

This study focused mainly on the leaching from NiTi archwires at neutral PH, here, could achieved by quantification of nickel and titanium ions using inductively coupled plasma mass spectrometry, the study shows three brands have three different corrosion resistance.

On comparison of Ni and Ti release of three round wires at T1, Group Ia compared with IIa and IIIa shows there is no significant difference compared with

other groups. P value of Group IIa and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three rectangular wires at T1, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.03 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three round wires at T2, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.04 and 0.03 respectively for Ni and Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three rectangular wires at T2, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.03 and 0.02 respectively for Ni and 0.04 and 0.03 for Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three round wires at T3, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.04 and 0.03 respectively for Ni and 0.04 and 0.02 for Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three rectangular wires at T3, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three round wires at T4, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.04 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

On comparison of Ni and Ti release of three round wires at T4, Group Ia compared with IIa and IIIa shows there is no significant difference compared with other groups. P value of Group IIa and IIIa is 0.03 and 0.02 respectively for Ni and Ti, shows there is significant difference compared with other groups.

From the findings of present study Group I is better than other two Groups ie, Round and Rectangular wire of Group I shows least metal ion release than Group II and Group III. Each archwire has its distinctive biologic and surface properties. This may differ according to the manufacturers choice. Different manufacturers wires comes in variant cross sections and mechanical properties. So selection of archwires is very important for clinical use^{7,15}.

Release of metal ions from orthodontic appliances depend on the pH of saliva, components of arch wire, and duration of immersion and depended on the composition instead of quantity of metal in the wire^{19,28}.

Due to high corrosion resistance and favorable biocompatibility Titanium and titanium alloys are widely used in dentistry, NiTi wires have shown surface corrosion, degradation, and fracture in the oral cavity. Nickel can produce toxic and allergic responses, whereas Ti is not cytotoxic a protective passive film exists on the NiTi alloy, Ni or Ti ions may still be released from the metal surface in the acidic oral environment through the corrosion processes. The potential danger associated with corrosion in the use of NiTi wire comes from the biologically negative effects

of the Ni ion. NiTi wires produced by various manufacturers may have different corrosion resistances^{28,31}.

Comparison of mean Ni release between the groups at T1, P value of Group Ia is 0.14 and P value 0.13 of Group Ib shows there is no significant difference compared Group Ia and Group Ib with other groups. P value of 0.04, 0.03, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group III b respectively shows there is no significant difference between each other.

Comparison of mean Ti release of T1, on comparison with P value of 0.04 shows there is statistically significant difference when compared Group Ia with all other groups; P value of 0.04 shows there is statistically significant difference when compared Group Ib with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIa with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIb with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIa with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIb with all other groups;

On comparison of Ni release at T2, P value of Group Ia and Group Ib is 0.14 and 0.17 respectively, which is not statistically significant with other groups; P value of 0.04,0.03,0.03,0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T2, P value of Group Ia and Group Ib is 0.21 and 0.19 respectively, which is not statistically significant with other groups; P value

of 0.04, 0.04, 0.03, 0.03 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ni release at T3, P value of Group Ia and Group Ib is 0.14 and 0.13 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T3, P value of Group Ia and Group Ib is 0.16 and 0.19 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ni release at T4, P value of Group Ia and Group Ib is 0.11 and 0.12 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T4, P value of Group Ia and Group Ib is 0.16 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

These results clearly revealed that these three groups manufacturing process is different and these may or may not affect their wire properties. So Selection of the orthodontic wires on the basis of the alloy and manufacturing process may be fundamental for biocompatibility²¹.

According to Kuhta M (2009) NiTi orthodontic wire products from different manufacturers would have different corrosion resistance. Factors which influencing the leaching of ions include the manufacturer, pH value, and immersion period^{16,26}.

When comparing Ni release of Group Ia and Group Ib, P value at T1 is 0.34, T2 is 0.48, T3 is 0.12, T4 is 0.45 respectively shows there is not statistically significant difference between groups. When comparing Ni release of Group IIa and Group IIb, P value at T1 is 0.17, shows there is not statistically significant difference between groups. T2 is 0.04, T3 is 0.04, T4 is 0.04 respectively shows there is statistically significant difference between groups. When comparing Ni release of Group IIIa and Group IIIb, P value at T1 is 0.4, T2 is 0.3, T3 is 0.3, T4 is 0.3 respectively shows there is statistically significant difference between groups.

When comparing Ti release of Group Ia and Group Ib, P value at T1 is 0.14, T2 is 0.12, T3 is 0.26, T4 is 0.31 respectively shows there is no statistically significant difference between groups. When comparing Ti release of Group IIa and Group IIb, P value at T1 is 0.34, T2 is 0.28, T3 is 0.17, T4 is 0.16 respectively shows there is no statistically significant difference between groups. When comparing Ti release of Group IIIa and Group IIIb, P value at T1 is 0.12, T2 is 0.16, T3 is 0.18, T4 is 0.32 respectively shows there is no statistically significant difference between groups.

As in the findings by Azizi et al The current study showed that, round wire is better than rectangular wire as the amount of metal ions leached from rectangular wires to the saliva is obviously greater than that of round, it is due to the fact that round and rectangular bar shapes with the same surface areas do not have the same volume and also be due to the different edges of the cross sections⁷.

When comparing mean Ni and Ti release at different immersion time at T1, on comparison of P value 0.12 of Group Ia and P value 0.13 of Group Ib shows there is no significant difference compared Group Ia and Group Ib with other groups. P value of 0.04, 0.03, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group III b respectively shows there is no significant difference between each other

Comparison of mean Ti release of T1, on comparison with P value of 0.04 shows there is statistically significant difference when compared Group Ia with all other groups; P value of 0.04 shows there is statistically significant difference when compared Group Ib with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIa with all other groups; P value of 0.03 shows there is statistically significant difference when compared Group IIb with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIa with all other groups; P value of 0.02 shows there is statistically significant difference when compared Group IIIb with all other groups.

On comparison of Ni release at T2, P value of Group Ia and Group Ib is 0.14 and 0.17 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.03, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T2, P value of Group Ia and Group Ib is 0.17 and 0.19 respectively, which is not statistically significant with other groups; P value of 0.04, 0.04, 0.03, 0.03 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ni release at T3, P value of Group Ia and Group Ib is 0.11 and 0.13 respectively, which is not statistically significant with other groups; P

value of 0.04,0.04,0.03,0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T3, P value of Group Ia and Group Ib is 0.13 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04,0.04,0.02,0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ni release at T4, P value of Group Ia and Group Ib is 0.11 and 0.12 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

On comparison of Ti release at T4, P value of Group Ia and Group Ib is 0.13 and 0.14 respectively, which is not statistically significant with other groups; P value of 0.04, 0.03, 0.02, 0.02 of Group IIa, Group IIb, Group IIIa, Group IIIb, respectively shows there is statistically significant difference between each other.

These results clearly indicate that group I is better than all three groups then Group II and Group III respectively. The round wire of all groups is better than rectangular wire; when comparing three manufactures the least Ni and Ti ion release is seen in Group Ia at all immersion periods and highest Ni and Ti ion release is shown by Group IIIb at all immersion periods and T1 shows least Ni and Ti ion release than other time periods.

As in the findings of Chung-Ju Hwang et al, in my study also there is a decrease in metal released as immersion time increased and there was no change in the amount of metal released after one month³³.

Another significant finding of this study is that the amount of metal ion leached was below daily dietary intake, and the threshold value necessary to induce hypersensitivity overall quantity of ions leached is 700 times lower than the concentrations necessary to elicit cytotoxic reactions in human.^{20,35}

Daily body intake of nickel via food is approximately 300-500 mg/dl. Release of titanium from fixed orthodontic appliances is less than nickel. However, release of these ions is below the daily dietary intake and does not bring about biological concerns^{61,69,70}.

Prevalence of nickel allergy is higher in females than males (28% in females, 0% in males). Nickel-containing orthodontic appliances had little or no effect on the gingival and oral health of the patient. Nickel hypersensitivity may occur in patients with a prior history of hypersensitivity to these metals^{16,18,47}.

According to Kolokitha OE(2008) and Janson GR(1998) Orthodontic patients with no cutaneous piercing or with skin pierced have no statistically significant differences of nickel hypersensitivity after treatment compared with the general population also found that orthodontic treatment is not associated with an increase in the prevalence of nickel hypersensitivity unless subjects have a history of cutaneous piercing. Orthodontic therapy with nickel containing appliances does not initiate or aggravate a nickel hypersensitivity reaction^{24,46}.

The range of salivary metal levels found did not exceed those of daily intake through food and air^{5,19}.

This present study has shown that at neutral P^H nickel and titanium release is much below than the toxic limits. Further test via any other P^H level of saliva or any other dimension of wires or any other manufactures could be carried out in future.

CONCLUSION

CONCLUSION

The release of metal ions from NiTi wires cannot be fully avoided. Based on the statistical analysis, the following conclusions were drawn:

- When comparing three manufactures, Group I(0.016” and 19*25”Dentauram NiTi) shows least Ni and Ti ion leaching among other two groups.
- When comparing round and rectangular wires; round wires shows less ion release than rectangular wires.
- The least Ni and Ti ion release is shown by Group Ia(0.016”Dentauram NiTi) at all time periods. The highest Ni and Ti ion release is shown by Group IIIb (19*25 3M NiTi) at all time periods.
- At each immersion time,T1 shows least Ni and Ti ion release than other time periods, which gradually increases with immersion period
- The average amount of ions leached per day from round and rectangular of three manufactures was well below the tolerable daily dietary intake level.

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ANNEXURE

ANNEXURE - I

SREE MOOKAMBIKA INSTITUTE OF DENTAL SCIENCES
KULASEKHARAM, KANYAKUMARI DIST., TAMIL NADU, INDIA.



INSTITUTIONAL RESEARCH COMMITTEE

Certificate

This is to certify that the research project protocol, *Ref no. 19/07/2016* titled, *"A comparative evaluation of release of metal ions from round and rectangular orthodontic NiTi wires of three different manufacturers – an in vitro study"* submitted by *Dr. Chandana L R, II Year MDS, Department of Orthodontics and DentofacialOrthopaedics* has been approved by the Institutional Research Committee at its meeting held on *19th August 2016*.

Convener
Dr. T. Sreelal

Secretary
Dr. Pradeesh Sathyan

ANNEXURE - II



INSTITUTIONAL HUMAN ETHICS COMMITTEE

SREE MOOKAMBIKA INSTITUTE OF MEDICAL SCIENCES,
KULASEKHARAM, TAMILNADU

Communication of Decision of the Institutional Human Ethics Committee(IHEC)

SMIMS/IHEC No: 2 /Protocol no: 3 / 2017

Protocol title: A COMPARATIVE EVALUATION OF THE RELEASE OF METAL IONS FROM ROUND AND RECTANGULAR NITI WIRES OF THREE DIFFERENT MANUFACTURERS – AN IN VITRO STUDY.					
Principal Investigator: Dr.Chandana. L.R					
Name& Address of Institution: Department of Orthodontics Sree Mookambika Institute of Dental Sciences					
<input checked="" type="checkbox"/>	New review	<input type="checkbox"/>	Revised review	<input type="checkbox"/>	Expedited review
Date of review (D/M/Y): 04-12-2017					
Date of previous review , if revised application:					
Decision of the IHEC:					
<input checked="" type="checkbox"/>	Recommended	<input type="checkbox"/>	Recommended with suggestions		
<input type="checkbox"/>	Revision	<input type="checkbox"/>	Rejected		
Suggestions/ Reasons/ Remarks:					
Recommended for a period of : Two month					

Please note*

- Inform IHEC immediately in case of any Adverse events and Serious adverse events.
- Inform IHEC in case of any change of study procedure, site and investigator
- This permission is only for period mentioned above. Annual report to be submitted to IHEC.
- Members of IHEC have right to monitor the trial with prior intimation.



Renigalanganadhar
Signature of Member Secretary (IHEC)

ANNEXURE - III



UNIVERSITY OF KERALA

SOPHISTICATED INSTRUMENTATION AND COMPUTATION CENTRE (SICC)

Kariavattom Campus, Thiruvananthapuram, Kerala, India - 695 581

E-mail: siccuok@gmail.com

Website: www.keralauniversity.ac.in

Client: Chandana L R
Chandanam, Keezhammakom, Vlathankara PO
Neyyatinkara, Trivandrum 695134

Instrument : ICP-MS
Sample : Artificial Saliva
No: of samples : 126
Test Required : Quantification of Nickel and Titanium from Artificial saliva
Test Completed : 27.04.2018
Issue Date : 30.04.2018

Detailed results are given in the attached sheets. Levels are record in ppm.

Reviewed by


Director SICC



Analysed by


Technical Officer (ICP-MS Lab)

Director
Sophisticated Instrumentation and Computation Centre