

**Determination of metal ion release from fixed orthodontic appliances – an in vivo study**Akhil shetty<sup>1</sup>, Jubin Baby Abraham<sup>2</sup>, Azhar mohammed<sup>3</sup>, E.sneha<sup>4</sup>

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**Correspondence Author:** Dr. E.Sneha, Department of orthodontics and dentofacial orthopaedics,5th floor, A B Shetty Memorial Institute of Dental Sciences, Deralakatte, Nitte University, Mangalore, Karnataka , 575018,India**Conflicts of Interest:** Nil**Abstract****Aim:** To determine the amount of Ni, Cr, Fe and Co in the saliva of patients being treated with fixed orthodontic appliances.**Materials and Methods:** A total number of 50 patients aged between 14-30 years of age were included in the study. Salivary samples were collected at four stages from the patient undergoing orthodontic treatment at the following times: T(0) – before insertion of the appliance, T(1) – 1 hour after insertion of the appliance, T(2) – 3 months after insertion of the appliance, T (3) - months after insertion of the appliance. Metal ion concentrations of saliva were evaluated using atomic absorption spectrophotometer (AAS).**Statistical analysis:** The results of the salivary ion concentrations were statistically evaluated using paired t test. A significance level of  $p>0.05$  was considered as statistically insignificant.**Results:** It was noted that there was a significant ( $p<0.001$ ) increase in concentration of metal ion in the saliva.**Conclusion:** The findings indicate that measurable amounts of nickel, chromium, iron and cobalt can be found in the saliva of the patients underwent fixed orthodontic treatment over a period of six

months but the increase in values do not reach toxic levels. Further studies should be carried out to evaluate the effects of these changes over the course of the treatment.

**Keywords:** Metal Ions, Saliva.**Introduction**In the oral environment, orthodontic appliances are exposed to harmful physical and chemical agents which may cause metallic corrosion. Abrasion by foods, liquids and tooth brushes leads to the release of ions which in turn causes corrosion.<sup>[1]</sup>Since different metal alloy combinations are used for various periods in orthodontic patients, special consideration with regard to their biocompatibility should be given. The oral cavity contains many factors which promote the biodegradation of orthodontic appliances thereby facilitating corrosion. The alteration of pH and temperature, the enzymatic and microbial activity, and the various chemicals that come in contact with oral cavity through food and drink are all corrosion conductors. Saliva acts as an electrolyte which further facilitates metal ion conduction. Each metal alloy has capacity of inherent heterogeneity and its utilization with other alloys, irregular micro surface, and the amount of forces that acts on the appliances and the friction between wires and brackets also add to the corrosion process<sup>[2]</sup>

The purpose of our study is to determine the concentrations of these corrosion products nickel (Ni), chromium (Cr) and iron (Fe) in saliva with fixed orthodontic appliances and to evaluate the toxic levels.

#### **Materials and Methods:**

This study was carried out in the Department of Orthodontics and Dentofacial Orthopaedics A.B Shetty Memorial Institute of Dental Sciences, Mangalore. Salivary samples were processed at the Nitte University Centre for Science Education and Research (NUCSER), K.S. Hegde Medical Academy (KSHEMA), Mangalore. The Research protocol was approved by the Ethical committee of Nitte University, Mangalore. Parent's /patient informed consent was taken for the collection of samples. Spectrometric analysis was done for metal ion concentration in saliva was done at the University Science Instrumentation Centre (USIC), Mangalore University, Mangalore.

Saliva was used from new patients starting orthodontic treatment. Samples from a total of 50 patients between the age group of 14-30 years were collected. The brackets used were standard stainless steel MBT brackets and archwire materials used were nickel titanium and stainless steel. The patients were non –smokers, had no previous history of orthodontic treatment and did not use any medicine or supplements. Subjects were thoroughly examined for the absence oral disease, systemic disease, oral restorations or prosthesis. Patients with no known allergy to jewellery, watches or any other sources of nickel, chromium, cobalt and iron. Salivary samples were collected in four stages from each orthodontic patient at the following times: T (0) – before insertion of the appliance, T (1) – 1 hour after insertion of the appliance, T (2) – 3 months after insertion of the appliance, T (3) - 6 months after insertion of the appliance.

The patients initially rinsed their mouth thoroughly with a mouthful of distilled, deionised water for 30 seconds. Unstimulated whole saliva of 5ml approximately was collected and the samples were stored at - 20°C before they were processed. The concentrations of metal ion in saliva are stable for 6 months when stored at -20°C. Atomic absorption spectrophotometer was used to analyze metal ions in biological samples without any separation of the metal ion from its biological matrix. In spectrophotometric method, there is no necessity for extraction procedures to analyze the elements, only dilution of the samples was enough to eliminate the interference and effects of the biological matrix. Before the analysis, samples were centrifuged at 3000 rpm for 10minutes to settle the particulate matter. 1ml of saliva samples of each patient was treated with nitric acid (2 mL, 0.5%), heated in sand bath to get a clear solution and then diluted to 10 ml with deionised distilled water. To measure the amount of Nickel, Chromium, Cobalt and Iron release, atomic absorption spectrophotometry was used. Standard solutions of Nickel, Chromium, Cobalt and Iron were prepared of concentrations between 0.1 ng/l and 0.8 ng/l. Each test was analyzed 3 times and the average was used as the result.

Before each test, 1 distilled water sample was processed in order to prevent possible contamination. The insoluble precipitate was excluded in the analysis due to variation in the results because of particles. The samples were analyzed and the nickel, chromium, cobalt and iron concentrations were calculated as parts per billion (ppb).



Figure 1: Atomic Absorption Spectrophotometer

**Stastical Analysis:** The results of the salivary ion concentrations were statistically evaluated using paired t test. A significance level of  $p > 0.05$  was considered as statistically insignificant.

**Results:**

**NICKEL**

A variation in the concentration of nickel in saliva was observed. On examining the concentration of nickel in

saliva released over a period of six months, it was seen that the concentrations increased from T0 ( $30 \pm 2.3$  ppb) to T3 ( $80.1 \pm 2.5$  ppb). The results showed a statistically highly significant p value ( $p < 0.001$ ) when salivary concentrations were compared at different time intervals with each other. (Table -1)

	Group					Paired differences		t	df	P-value
		Mean	N	Std.deviation	Mean	Std.Deviation				
Pair 1	T0	30.002	50	2.36828	-15.262	3.57251	-30.208	49	<0.001	
	T1	45.264	50	2.67999						
Pair 2	T0	30.002	50	2.36828	-30.686	2.63028	-82.494	49	<0.001	
	T2	60.688	50	2.67228						
Pair 3	T0	30.002	50	2.36828	-50.164	2.74105	-129.408	49	<0.001	
	T3	80.166	50	2.54587						
Pair 4	T1	45.264	50	2.67999	-15.424	4.11283	-26.518	49	<0.001	
	T2	60.688	50	2.67228						
Pair 5	T1	45.264	50	2.67999	-34.902	4.01134	-61.524	49	<0.001	

	T3	80.166	50	2.54587					
Pair 6	T2	60.688	50	2.67228	-19.478	3.14333	-43.817	49	<0.001
	T3	80.166	50	2.54587					

Table 1: Comparison of the mean salivary nickel ion concentration using paired t test

**2. CHROMIUM**

On examining the concentration of chromium in saliva released over a period of six months, it was seen that the

concentrations increased from T0 (3.4±0.67ppb) to T3 (20.7±1.5ppb).The results showed a statistically highly significant p value (p<0.001) when salivary concentrations were compared at different time intervals with each other. (Table-2)

	Group	Mean	N	Std.deviation	Paired differences				
					Mean	Std.Deviation	t	df	P-value
Pair 1	T0	3.472	50	0.67098	-4.676	1.03816	-31.849	49	<0.001
	T1	8.148	50	0.76618					
Pair 2	T0	3.472	50	0.67098	-11.44	1.11429	-72.596	49	<0.001
	T2	14.912	50	1.11733					
Pair 3	T0	3.472	50	0.67098	-17.232	1.60782	-75.785	49	<0.001
	T3	20.704	50	1.5036					
Pair 4	T1	8.148	50	0.76618	-6.764	0.99668	-47.988	49	<0.001
	T2	14.912	50	1.11733					
Pair 5	T1	8.148	50	0.76618	-12.556	1.81897	-48.81	49	<0.001
	T3	20.704	50	1.5036					
Pair 6	T2	14.912	50	1.11733	-5.792	2.13693	-19.166	49	<0.001
	T3	20.704	50	1.5036					

Table 2: Comparison of the mean salivary chromium ion concentration using paired t test

**3) IRON**

On examining the concentration of chromium in saliva released over a period of six months, it was seen that the concentrations increased from T0 (51.9±6.8ppb) to T3

(100±4.3ppb).The results showed a statistically highly significant p value (p<0.001) when salivary concentrations

were compared at different time intervals with each other. (Table -3)

	Group				Paired differences				
		Mean	N	Std.deviation	Mean	Std.Deviation	t	df	P-value
Pair 1	T0	51.96	51	6.86405	-20.428	7.08875	-20.58	50	<0.001
	T1	72.388	51	5.58117					
Pair 2	T0	51.96	51	6.86405	-37.8	6.92682	-38.971	50	<0.001
	T2	89.76	51	3.64675					
Pair 3	T0	51.96	51	6.86405	-48.074	7.91034	-43.401	50	<0.001
	T3	100.034	51	4.39225					
Pair 4	T1	72.388	51	5.58117	-17.372	5.9645	-20.8	50	<0.001
	T2	89.76	51	3.64675					
Pair 5	T1	72.388	51	5.58117	-27.646	6.92067	-28.528	50	<0.001
	T3	100.034	51	4.39225					
Pair 6	T2	89.76	51	3.64675	-10.274	3.47228	-21.131	50	<0.001
	T3	100.034	51	4.39225					

Table 3: Comparison of the mean salivary iron ion concentration using paired t test

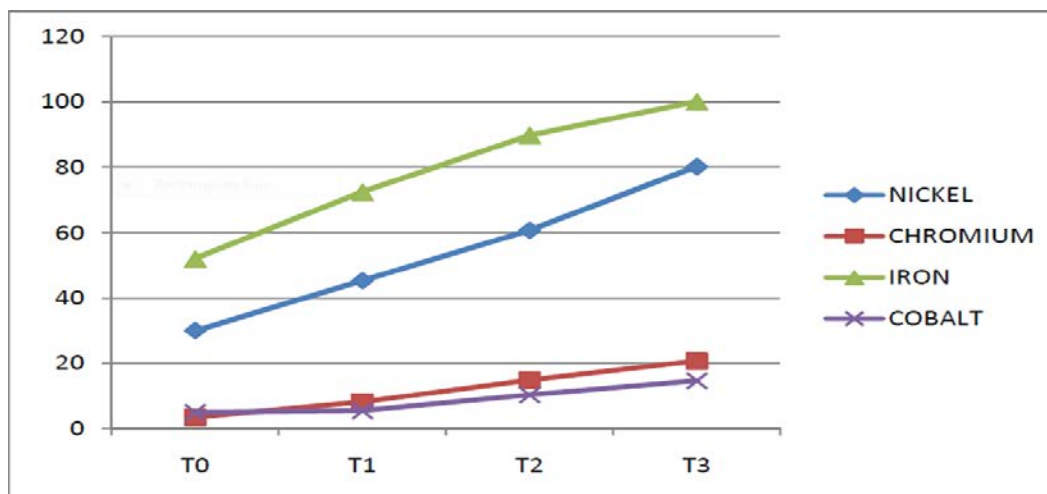
concentrations increased from T0 (4.9±1.1ppb) to T3 (14.6±1.1ppb).The results showed a statistically highly significant p value (p<0.001) when salivary concentrations were compared at different time intervals with each other. (Table-4)

**4. COBALT**

On examining the concentration of chromium in saliva released over a period of six months, it was seen that the

	Group				Paired differences		t	df	P-value
		Mean	N	Std.deviation	Mean	Std.Deviation			
Pair 1	T0	4.968	50	1.17133	-0.492	0.48058	-7.239	50	<0.001
	T1	5.46	50	1.16479					
Pair 2	T0	4.968	50	1.17133	-5.326	1.61281	-23.351	50	<0.001
	T2	10.294	50	1.18415					
Pair 3	T0	4.968	50	1.17133	-9.688	1.81554	-37.732	50	<0.001
	T3	14.656	50	1.16936					
Pair 4	T1	5.46	50	1.16479	-4.834	1.67717	-20.38	50	<0.001
	T2	10.294	50	1.18415					
Pair 5	T1	5.46	50	1.16479	-9.196	1.87823	-34.621	50	<0.001
	T3	14.656	50	1.16936					
Pair 6	T2	10.294	50	1.18415	-4.362	1.6886	-18.266	50	<0.001

Table 4: Comparison of the mean salivary cobalt ion concentration using paired t test.



Graph 1: Representation of mean metal ion concentration in saliva at various time intervals.

## Discussion:

Orthodontic appliances are capable of releasing metal ions in the oral cavity as most of them are made of stainless steel and NiTi alloys. The two factors mainly involved in the corrosion of orthodontic appliances and metal ion release in the oral cavity are the manufacturing process and environmental factor. The manufacturing process is mainly based on the type of alloy and the characteristics of the metals used. The environmental factors, includes mechanical stress, diet, time of the day, salivary flow rate, and health and psychosomatic condition of the individual.<sup>3</sup>

Water contains hydrogen (H<sup>+</sup>) and hydroxyl (OH<sup>-</sup>) ions. When both these ions are present in equal proportions, it has the neutral pH of 7. When there are excess amount of H<sup>+</sup> ions, then the water is called to be 'acidic'. If OH<sup>-</sup> ions exceed H<sup>+</sup> ions, then the water is called to be 'alkaline'. The pH scale ranges from 0 to 14 and is logarithmic, which means that each step is 10 times the previous one. In other words, a pH of 4.5 is 10 times more acidic than 5.5, 100 times more acidic than 6.5 and 1000 times more acidic than 7.5.<sup>[3]</sup>

Chlorine, sulphur, and phosphoric acid are said to be acidic minerals and they carry negative charges as they get attracted to H<sup>+</sup> ions. These negatively charged OH<sup>-</sup> ions that gets attracted positive electrical charge H<sup>+</sup> ions are called alkaline. Alkaline minerals like calcium, potassium, magnesium, and sodium are of nutritionally important.<sup>[3]</sup>

Kuhta *et al.* (2009) reported that if the salivary pH is reduced from 6.75 to 3.5, it can increase the release of metal ions from orthodontic appliances up to 100-fold.<sup>[4]</sup>

Low pH values also reduce the resistance of dental alloys to corrosion (Huang *et al.*, 2003).<sup>[5]</sup>

The corrosion of metal and ion release such as Ni has absolute relationship with time (Eliades *et al.*, 2003).<sup>[6]</sup> Except for a few reports (Agaoglu *et al.*, 2001<sup>[7]</sup> Eliades *et al.*, 2003<sup>[6]</sup> Fors and Persson, 2006),<sup>[8]</sup> the

majority of previous publications have calculated the amount of metal ions released inside the oral cavity over a short period of time

(1–3 months).<sup>[4]</sup>

Eliades *et al.*<sup>[6]</sup> in his study reported that short duration is not sufficient to effectively analyze the salivary metal ion content of orthodontic patients. However the present study was done over 6 months and the salivary concentrations of the metal ions were much higher than the concentrations reported in other studies done over a short and were also statistically significant. Our study in general showed an increase in salivary Ni, Cr, Fe and Co concentrations in patients with fixed orthodontic appliances over a period of six months.

According to the WHO guidelines (2003) normal ranges of metal ions assessed in this study are <1mg/L nickel (<1000ppb), 0.05mg/L of chromium (50ppb), 0.3 mg/L of Iron (300ppb) and Cobalt 0.05 mg /l (50 ppb). According to the study at the end of 6months the concentrations of various metal ions studied were as follows: Ni - 80.1±2.5 ppb, Cr - 20.7±1.5ppb, Fe - 100±4.3ppb and Co - 14.6±1.1ppb. So it is seen that even though there is an increase in metal ion concentrations (Ni, Cr, Fe and Co) which was seen over the study period, these values were well within the normal limits and none of the metal ion concentrations reached toxic levels. This study showed significant changes occurring in patients treated with fixed orthodontic appliances over a 6-month period. The outcome of results included increases in cellular nickel, chromium, iron and cobalt content.

Although saliva is a non-invasive parameter, it has certain limitations as the saliva can be continuously washed out or swallowed which gives information at the moment of sampling only.

## **Conclusion**

1. The findings indicate that measurable amounts of nickel, chromium, iron and cobalt can be found in the saliva of the patients undergoing fixed orthodontic treatment over a period of six months but the increase in values do not reach toxic levels.
2. Further studies should be carried out to evaluate the effects of these changes over the course of the treatment.

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