The effect of released galvanic corrosion products caused by wire bracket contact on bacterial flora: an in-vitro study

Wael Mohamed Mobarak Refai

Background **The galvanic corrosion products released from contact between dissimilar orthodontic alloys and their effect on bacterial flora have not been given the importance they deserve.**

Aim **This study was conducted to assess the effect of galvanic corrosion products of different wire bracket couples on bacterial flora.**

Methods **The study included 30 individuals from whom bacteria were collected from the gingival sulcus of the upper and lower posterior teeth. From nine types of bacteria isolated, only four types (two aerobes and two anaerobes) causing periodontitis were selected: *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Aggregatibacter actinomycetemcomitans*, and *Klebsiella* spp. These bacterial species were proliferated and counted using a colony counter. Simultaneously, three types of wires (StSt, NiTi, and CuNiTi) were coupled with stainless-steel brackets. Both round and rectangular shapes were used. The samples were immersed in an electrolytic solution and incubated at 37°C for 1, 2, and 3 weeks. The fluid containing the galvanic corrosion products was then added to the cultures.**

Results **The decrease in bacterial count was proportional to the duration of immersion. The greatest decrease was found using CuNiTi wires, followed by stainless-steel wires and finally NiTi ones.**

Conclusion **Rectangular wires showed greater decrease in bacterial counts than did round wires. However, the decrease was insignificant. *Egypt J Oral Maxillofac Surg* 3:36–44 © 2012 The Egyptian Association of Oral & Maxillofacial Surgeons.**

Egyptian Journal of Oral & Maxillofacial Surgery 2012, **3**:36–44

Keywords: corrosion products, galvanic corrosion, ion release, subgingival plaque, wire bracket couple

Department of Orthodontics, Minia University, Minia, Egypt

Correspondence to Wael Mohamed Mobarak Refai, Department of Orthodontics, Minia University, 52 Demashk St, Mohandesine, 4114 Cairo, Egypt
Tel: +2 02 37618847; e-mail: w_refai_67@hotmail.com

Received 5 October 2011 **accepted** 13 January 2012

Downloaded from https://journals.lww.com/eaoms by BhDMf5ePHKav1zEoum1tQfN4a+kJLhEZgbsIHo4XMi0hCywCX1AWnYQp/IlQrHD3i3D0OdRyi7TvSFl4Cf3VC1y0abggQZXdgGj2MwlZLeI= on 04/10/2018

Introduction

The effects of orthodontic treatment on periodontal health has been investigated in several studies [1,2], and many researchers have observed an inflammation of gingival tissues during fixed orthodontic therapy.

Fixed orthodontic metallic appliances such as brackets, archwires, and molar bands are manufactured from base metal alloys such as stainless steel (FeCrNi), cobalt–chromium–nickel (CoCrNi), nickel–titanium (NiTi), β-titanium (β-Ti), and pure titanium.

These orthodontic alloys except for β-Ti and pure titanium contain nickel, the release of which produces mild allergic reactions during orthodontic treatment, which are of clinical concern.

Whether ion release, especially nickel, has adverse effects has been debated in various studies. It was found that increased Ni ions are released initially after the orthodontic devices have been fitted, but they decay quickly. It is unlikely that orthodontic NiTi wires are a relevant additional Ni load for the patient [3].

House *et al.* [4] reported that the combination of these materials in close proximity and in hostile conditions can result in corrosion. However, Staffolani *et al.* [5] have concluded that the quantities of metal ions released in our experimental conditions should not be a cause for concern in utilizing appliances.

Moreover, it was believed that, although nickel is a strong sensitizer in contact allergy development [6], adverse reactions related to orthodontic appliances are rare [7]. This could be explained by a possible tolerance development as a result of early intraoral exposure to nickel, which has been hypothesized and supported by experimental data [8,9].

In contrast, Fors and Persson [10] concluded that nickel release occurs into the dental plaque and components of saliva of orthodontic patients, a situation that may reflect time dependence of its release from orthodontic appliances into the oral cavity and an aggregation of nickel at plaque sites.

Widu *et al.* [11] indicated that because of the increasing number of orthodontic treatments using devices containing nickel and the growing prevalence of nickel allergy in the average population, biocompatibility studies of these devices has become a topic of major interest.

In a clinical situation, two dissimilar alloys (brackets and archwires) having different corrosion potentials are placed in contact in the oral cavity during treatment (1–2 years). This can cause galvanic corrosion leading to release of metal ions. The major corrosion products are Fe, Cr, and Ni for stainless steel and Ti and Ni for NiTi alloys [12].

Nickel and chromium have received the most attention because of their reported potential for producing allergic, toxic, or carcinogenic effects [13,14]. Josef [15] indicated that galvanic corrosion can cause serious conditions such as lichen planus and leukoplakia and even oral cancer.

2090-097X © 2012 The Egyptian Association of Oral & Maxillofacial Surgeons DOI: 10.1097/01.OMX.0000412905.80544.89

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

The corrosion behavior of dental alloys is of considerable interest as it is an indication of their biocompatibility. Therefore, understanding its mechanism and characteristics is important. Corrosion of metals and alloys in aqueous solutions occurs on the surface in the form of an electrochemical reaction. These reactions are continuous, resulting in dissolution of metal and loss of ions [16].

In electrochemical corrosion, a galvanic cell is created when two different metals are coupled. The driving force for corrosion is a potential difference between the different materials. Moreover, the galvanic corrosion potential indicates the point at which the oxide film of the alloy is broken down and dissolution of the alloy begins. A lower corrosion potential indicates higher susceptibility to corrosion [17].

Many studies had focused their attention on the effects of orthodontic appliances on the specific microbial composition of the subgingival plaque. However, the effect of galvanic corrosion products on subgingival bacterial flora was overlooked. Accordingly, this study was conducted to achieve this objective.

Materials and methods

This research comprised four parts. However, all the steps except the last one were carried out simultaneously:

(1) Preparation of the fluid containing the corrosion products in the chemical engineering lab.
(2) Patient preparation and sample collection.
(3) Bacterial isolation and count in the microbiological lab.
(4) Addition of the fluid containing the corrosion products to the samples.

Preparation of the fluid containing the corrosion products in the chemical engineering lab

Upper central incisor brackets were used (American Orthodontics, Sheboygan, Wisconsin, USA) to benefit from the large surface area of the slot of that bracket. With regard to wires, FeCrNi, NiTi, and copper–nickel–titanium (CuNiTi) wires were used, which were both round (0.018 inch) and rectangular (0.016×0.022 inch) (Ormco, Glendora, California, USA).

Simulating the oral environment, the mesh area was isolated using a composite (Green glue, Glendora, California, USA). Wires were inserted in the bracket slots and tied using elastomeric O-ties. The specimen was inserted in a test tube containing normal saline (NaCl), which was used as an electrolyte (Fig. 1).

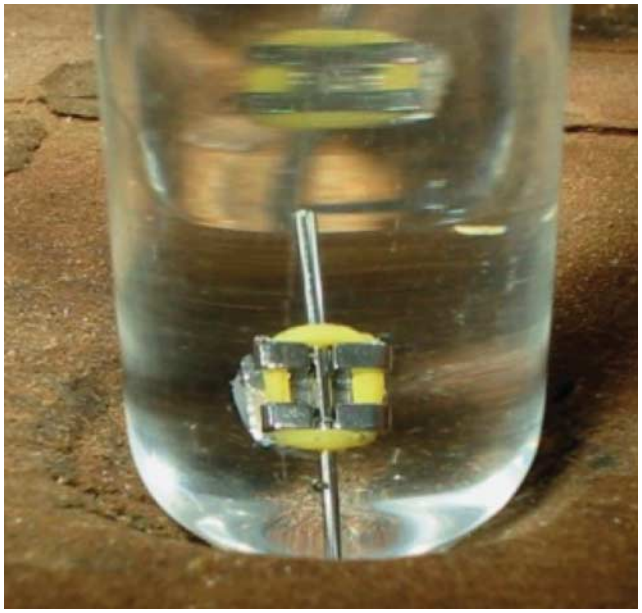
The tubes containing the wire bracket couple were divided into three groups. They were incubated at 37°C. The first group was incubated for 1 week, the second for 2 weeks, and finally the third for 3 weeks (Fig. 2).

The specimens were given the following codes:

S: stainless steel; NT: NiTi alloy; CNT: CuNiTi; R: round cross-section; S (before the number): rectangular cross-section; and finally the number representing the week of incubation.

For example, NTR1 stands for rounded NiTi wire after 1 week of incubation.

Fig. 1



The specimen inside the electrolyte.

Fig. 2

The three groups in the incubator.

Patient preparation and sample collection

This study included 30 patients (11 male, 19 female) who intended to undergo orthodontic treatment. Their ages ranged between 17 and 24 years to avoid the hormonal effect of puberty on bacterial count or species. There was no administration of antibiotics for 4 weeks before sample collection to avoid the effect of drugs on bacterial types or growth [18]. All female patients were unmarried to avoid the effect of hormonal changes on bacterial results [19].

All patients were subjected to thorough scaling and teeth polishing 1 week before participating in this study to ensure that they all had good oral hygiene and were free of periodontal diseases.

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

Informed consent was signed by every participant admitting that he is a volunteer in the study and that he will strictly follow the rules pertaining to oral hygiene. All patients were instructed to use the same type of toothpaste (Signal 2 toothpaste, Unilever Mashreq, Cairo, Egypt) to avoid the effects of different tooth pastes on bacterial types or count.

Patients were asked to brush their teeth three times a day (after breakfast, lunch, and dinner, or before going to bed). This type of prophylaxis was carried out because the manual brushing proved to be less superior than Sonic brushing in improving periodontal health in adolescent orthodontic patients [20].

Fixed orthodontic treatment was carried out and molars were banded. Subgingival crevicular fluid samples were collected from the proximal sites of the index teeth 1 month after starting the treatment. Moreover, antibiotic intake for at least 4 weeks before the time of sample collection had to be avoided.

The index sites were the mesial proximal site of the gingival sulcus of the upper and lower permanent canines, upper and lower first permanent molars, besides being the distal proximal site of the gingival sulcus of both the last permanent upper and lower molars.

A sterile paper point (Meta, Korea) was used to collect the samples by inserting it in the index site for 15 s. The sterile paper point was then pooled in a sterile sealed plastic container containing tryptone soy broth (LAB M, Detroit, MI, USA) for aerobic culture and thioglycolate broth (Difco, Internatinal Diagnostic Group, Lancashire, UK) for anaerobic culture. Samples were delivered within 2 h to the microbiological lab.

Bacterial isolation and count in the microbiological lab

Culture and biochemical reaction

With regard to culture formation, samples were cultured in different types of media for bacterial growth in the microbiological laboratory. For anaerobic bacterial growth, dishes containing medium were kept in an anaerobic GasPack system (Fig. 3). Samples were incubated for 24 h at 37°C in the incubator.

Aerobic cultures were examined by methyl red reaction (Difco, UK), Voges–Proskauer's reaction (Difco, UK), and indole reaction (LAB M, USA) to identify different types of bacterial species. The anaerobic profile index (Bio Merieux, Becton, Dickinson, France) was used to identify different types of anaerobic bacterial species (Fig. 4). The cultivation media were nutrient agar (Difco, USA) (Figs 5 and 6) and MacConkey's agar (LAB M, UK) (Fig. 7).

All these media and tests were autoclaved for 15 min at 121°C to ensure complete sterilization before use to avoid other bacterial incorporation in the culture [21].

Nine types of bacterial species were revealed by microbial examination: *Staphylococcus epidermidis*, *Staphylococcus aureus*, *Streptococcus mutans*, *Streptococcus mitis*, *Lactobacilli* spp., *Prevotella* spp., *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, and *Klebsiella* spp.

Fig. 3

The anaerobic GasPack system.

Fig. 4

Anaerobic profile index.

However, only four types (two aerobes and two anaerobes) were selected: *S. epidermidis*, *S. aureus*, *A. actinomycetemcomitans*, and *Klebsiella* spp.

Bacterial count

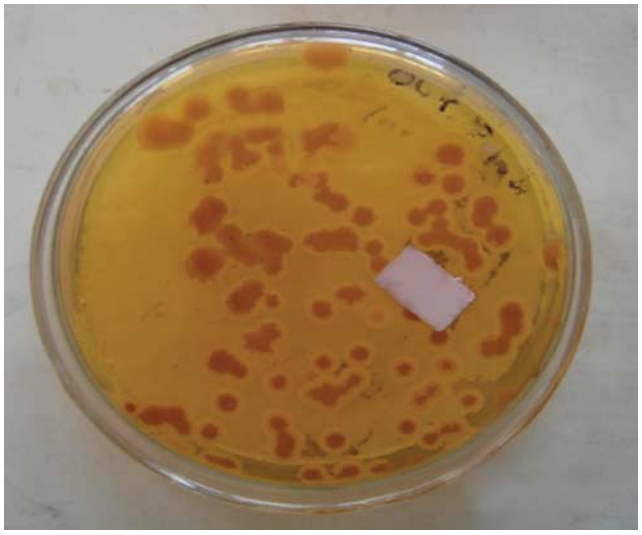
Because of their very small size, counting the number of bacteria in a sample can be difficult. Usually, bacterial samples must be diluted considerably to obtain reasonable counts. This was done by carrying out serial dilution.

Bacterial cell numbers were reduced by repeatedly diluting the amount of bacteria in the sample. A 0.1 ml of bacterial sample was mixed with 9.9 ml of diluent solution (0.9% NaCl), and then successive dilutions were made.

A small amount of each of the diluted bacterial samples was then spread on an agar plate. The number of bacterial colonies that grew on each plate was counted. By multiplying the number of colonies with dilution factor

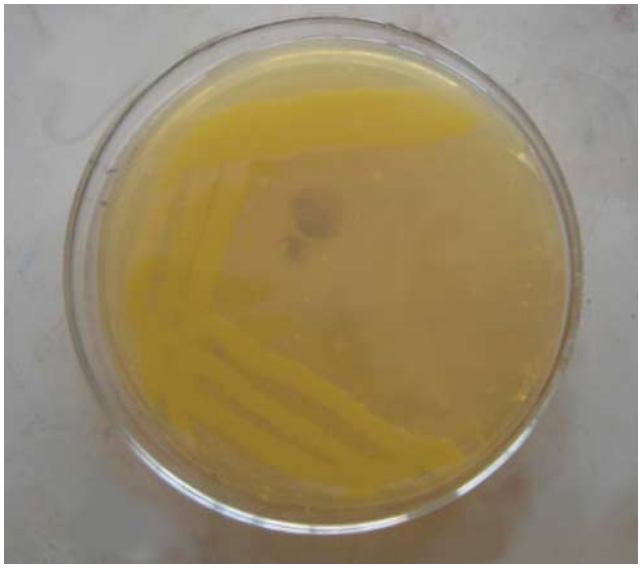
Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

Fig. 5



Staphylococcus epidermidis over nutrient agar.

Fig. 6



Staphylococcus aureus over nutrient agar.

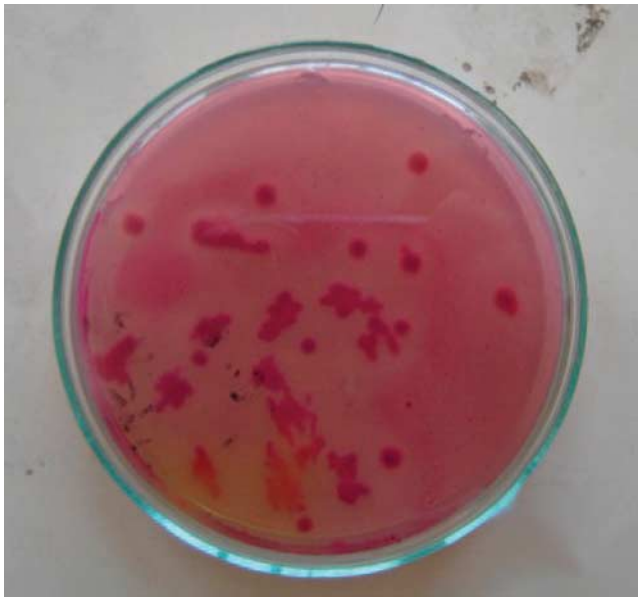
(the number of times that you have diluted the bacterial sample with the diluent solution), it was possible to determine the number of bacteria in the original sample [21].

The procedure was carried out using a colony counter. It consisted of lighted surfaces on which the plate was placed, with the colonies marked off with a felt-tipped pen on the outer surface of the plate while the operator recorded the count manually (Fig. 8).

Addition of fluid containing the corrosion products to the bacterial samples

This was the final step. Thereafter, the bacterial count was repeated.

Fig. 7



Klebsiella colony on MacConkey's agar.

Fig. 8

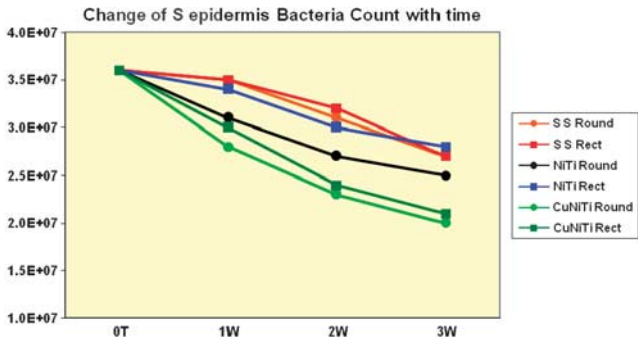
Colony counter.

Results

Data were checked, coded, entered, and analyzed using the Statistical Package for Social Sciences (SPSS, IBM, Armonk, NY, USA) version 1.0 software. Descriptive analysis was carried out to estimate the difference in bacterial counts after 1, 2, and 3 weeks (T0 represents the initial bacterial count before adding the fluid containing the corrosion products).

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

Fig. 9



Change of *Staphylococcus epidermidis* bacteria count with time.

S. epidermidis showed an ascending decrease with time (Fig. 9) when the round FeCrNi wire was used. The same results were also revealed with the rectangular FeCrNi wire. The comparative difference in results was non-significant.

Both round and rectangular NiTi wires showed a decrease in *S. epidermidis* bacterial count with time. The greatest decrease was observed in the third week. The rectangular wire resulted in a greater decrease than the round one. However, the difference in bacterial count between both shapes was statistically nonsignificant.

With regard to CuNiTi wires, *S. epidermidis* count decreased with time. The greatest decrease was observed in the third week. Similarly, the decrease was greater with rectangular wires.

Comparing the three shapes, the greatest decrease was observed with CuNiTi wires followed by FeCrNi wires and finally NiTi wires in case of round cross-sections. The same was detected with the rectangular wire during the first and second week. In the third week the greatest decrease was observed among CuNiTi wires followed by NiTi wires and finally FeCrNi wires (Table 1).

The *S. aureus* count changed with time (Fig. 10). A decrease was revealed with the three types of round wires. The greatest decrease was seen in the third week. CuNiTi wires yielded the greatest decrease, followed by FeCrNi wires. The NiTi wire yielded the least decrease. With regard to rectangular wires, the three types showed the same results as the round ones. The decrease in bacterial count was higher with rectangular wires than with round ones. The greatest difference was found between round and rectangular NiTi wires. However, the bacterial count after the first week showed greater decrease than when CuNiTi wires were used (Table 2).

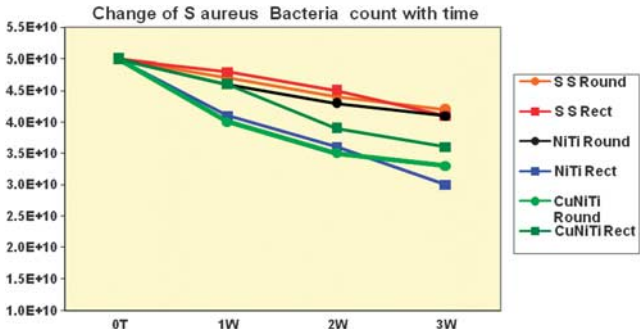
No statistically significant difference was detected between round and rectangular wires when examining the change in the bacterial count of *A. actinomycetemcomitans* (Fig. 11). The decrease was ascending, signifying that the greatest decrease was observed after 3 weeks. With regard to round wires, both NiTi and CuNiTi wires exhibited the same bacterial count in the second and third weeks. Round FeCrNi wires led to a lesser decrease in all the three periods compared with the other two types.

Table 1 Changes in *Staphylococcus epidermidis* bacterial counts between round and rectangular wires after addition of the ion-containing fluid

	Round			Rectangular		
	Stainless steel	NiTi	CuNiTi	Stainless steel	NiTi	CuNiTi
T0	3.6E+07	3.6E+07	3.6E+07	3.6E+07	3.6E+07	3.6E+07
W1	3.5E+07	3.1E+07	2.8E+07	3.5E+07	3.4E+07	3.0E+07
W2	3.1E+07	2.7E+07	2.3E+07	3.2E+07	3.0E+07	2.4E+07
W3	2.7E+07	2.5E+07	2.0E+07	2.7E+07	2.8E+07	2.1E+07

NiTi, nickel–titanium; CuNiTi, copper–nickel–titanium.

Fig. 10



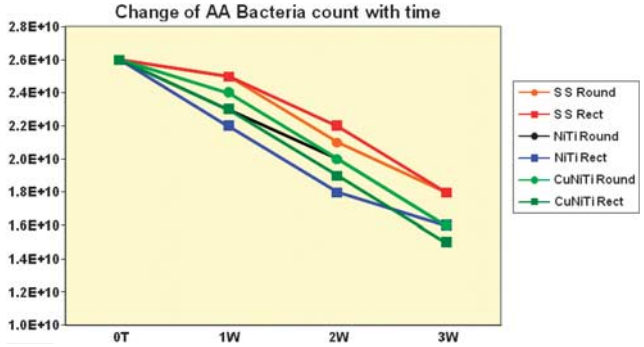
Change of *Staphylococcus aureus* bacteria count with time.

Table 2 Changes in *Staphylococcus aureus* bacterial counts between round and rectangular wires after addition of the ion-containing fluid

	Round			Rectangular		
	Stainless steel	NiTi	CuNiTi	Stainless steel	NiTi	CuNiTi
T0	5.0E+10	5.0E+10	5.0E+10	5.0E+10	5.0E+10	5.0E+10
W1	4.7E+10	4.6E+10	4.0E+09	4.8E+10	4.1E+10	4.6E+10
W2	4.4E+10	4.3E+10	3.5E+10	4.5E+10	3.6E+10	3.9E+10
W3	4.2E+10	4.1E+10	3.3E+10	4.1E+10	3.0E+10	3.6E+10

NiTi, nickel–titanium; CuNiTi, copper–nickel–titanium.

Fig. 11



Change of *Aggregatibacter actinomycetemcomitans* bacteria count with time.

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

With regard to rectangular wires, the least decrease was observed with CuNiTi wires. However, no statistically significant difference was detected between the three types. Moreover, the difference between the change in bacterial count of *A. actinomycetemcomitans* in both round and rectangular wires was nonsignificant. The least decrease in bacterial counts was detected in this type of bacteria after 3 weeks with CuNiTi wires (Table 3).

The round wire revealed the same results when the change in the bacterial count of *Klebsiella* spp. was inspected (Fig. 12). In the first week, the least decrease was observed with NiTi wires. In the second week, the least decrease was observed with CuNiTi wires followed by FeCrNi and finally NiTi wires. In the third week, bacterial counts decreased similarly with both NiTi and CuNiTi wires but less than with FeCrNi wires.

With the use of rectangular wires, the bacterial count of *Klebsiella* spp. decreased. The greatest decrease was observed with CuNiTi wires followed by NiTi and then FeCrNi wires. In the second week, the bacterial count was the same with NiTi and CuNiTi wires but less than that with FeCrNi wires. In the third week, the least decrease was observed with NiTi wires followed by CuNiTi wires and finally FeCrNi wires.

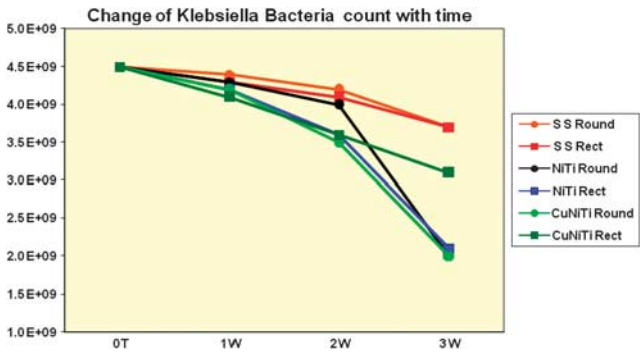
A comparison between the results of round and rectangular wires revealed no statistically significant difference, except the decrease in the bacterial count at the third week with NiTi wires (Table 4).

Table 3 Changes in *Aggregatibacter actinomycetemcomitans* bacterial counts between round and rectangular wires after addition of the ion-containing fluid

	Round			Rectangular		
	Stainless steel	NiTi	CuNiTi	Stainless S	NiTi	CuNiTi
T0	2.6E+10	2.6E+10	2.6E+10	2.6E+10	2.6E+10	2.6E+10
W1	2.5E+10	2.3E+10	2.4E+10	2.5E+10	2.2E+10	2.3E+10
W2	2.1E+10	2.0E+10	2.0E+10	2.2E+10	1.8E+10	1.9E+10
W3	1.8E+10	1.6E+10	1.6E+10	1.8E+10	1.6E+10	1.5E+10

NiTi, nickel–titanium; CuNiTi, copper–nickel–titanium.

Fig. 12



Change of *Klebsiella* bacteria count with time.

Table 4 Changes in Klebsiella bacterial counts between round and rectangular wires after addition of the ion-containing fluid

	Round			Rectangular		
	Stainless steel	NiTi	CuNiTi	Stainless steel	NiTi	CuNiTi
T0	4.5E+09	4.5E+09	4.5E+09	4.5E+09	4.5E+09	4.5E+09
W1	4.4E+09	4.3E+09	4.2E+09	4.3E+09	4.2E+09	4.1E+09
W2	4.2E+09	4.0E+09	3.5E+09	4.1E+09	3.6E+09	3.6E+09
W3	3.7E+09	2.0E+09	2.0E+09	3.7E+09	2.1E+09	3.1E+09

NiTi, nickel–titanium; CuNiTi, copper–nickel–titanium.

Discussion

In-vitro release of nickel from orthodontic appliances has been noted using microscopic analysis of corrosion and chemical analyses of orthodontic components when exposed to an artificial oral environment [22–24]. Release of nickel is reported to vary with the composition and manufacturing details of the appliance components [25] and between archwire alloys and mechanical straining [26].

The surface area ratio of two dissimilar alloys is a very important factor as it affects the galvanic corrosion behavior. An unfavorable area ratio, which consists of a large cathode and a small anode, might lead to a greater corrosion rate from the anodic alloy. When this is applied to the orthodontic field, it is difficult to determine the real surface area ratio between brackets and archwire in clinical use. Accordingly, an in-vitro study was carried out.

The effect of corrosion products on bacteria in the oral cavity was not given the importance it deserved. This study was therefore conducted to accomplish this objective.

First, the fluid containing corrosion products was prepared. In this study the electrochemical measurement method was used to produce galvanic corrosion products of various orthodontic wire-bracket couples.

Normal saline solution (0.9% NaCl) was used because it is a corrosive agent. As corrosion testing of dental material should be carried out at a standard temperature of 37°C [27], the solution temperature was controlled at this temperature with a thermostat to simulate the oral condition. This temperature (37°C) represents the normal temperature of the oral cavity [28]. Static immersion tests were carried out at different periods (7, 14, and 21 days), and at the end of each period the specimens were removed.

Three different wire alloys, namely, CuNiTi, NiTi, and FeCrNi, and one type of the most commonly used bracket made of iron–chromium–nickel (FeCrNi) were tested in a reference saline solution.

Thirty adult patients were selected to participate in this study. The ages of all patients ranged from 17 to 25 years. Banded and bonded attachments were inserted. After 1 month, samples were taken from the proximal surfaces of all index teeth to standardize the site of sample collection. This site was selected because the incidence of hyperplasia was greater in the posterior areas of the mouth than in the anterior and was greater interproximally than at the center of the crown [29]. Moreover,

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

Tzannetou *et al.* [30] had concluded that phosphatase activities in gingival crevicular fluid might be a useful means for monitoring tissue responses to orthodontic treatment.

Paper points of size 30 were used as they are rigid enough and can be easily inserted into the bottom of the gingival sulcus. The paper point was kept in the gingival sulcus for 15 s to allow sufficient time for absorption of the gingival crevicular fluid [31]. Thereafter, the samples were transported to the microbiological laboratory within 2 h.

Thioglycolate broth and tryptone soy broth were used as transporting and nourishing media for anaerobic and aerobic bacteria, respectively. Nutrient agar was used for all bacterial cultures. MacConkey's agar was used for culture of *Klebsiella* spp. [31].

With regard to the difference between the two strains of staphylococci bacteria, *S. epidermidis* was coagulase negative, whereas *S. aureus* was coagulase positive. Klebsiella bacteria tested negatively in both indole and methyl red tests, whereas it tested positively in Voges–Proskauer's test. Identification of different types of anaerobic bacteria was carried out using anaerobic profile index.

The following Gram-negative and Gram-positive bacteria were investigated: *S. epidermidis* and *S. aureus* (Gram-positive aerobic bacteria), *A. actinomycetemcomitans* (Gram-negative anaerobic bacteria), and finally *Klebsiella* spp. (Gram-negative aerobic bacteria).

They were selected because they increase in number in cases of periodontitis [1]. It was also proven that appliance-free young individuals initially infected with *A. actinomycetemcomitans* had a higher risk of experiencing more gingival inflammation than did individuals who were not infected with the bacterium during a 3-year observation period [32,33].

A colony counter was used to count bacteria before adding the ion-containing fluid (T0). The number of each type of bacterium was fixed among all the samples to ensure accuracy of the results. Bacteria were counted 1 week, 2 weeks, and 3 weeks after addition of galvanic corrosion product-containing solutions.

Bearing in mind the fact that this is an in-vitro study and salivary flow is absent, the results denote the direct relationship between corrosion products and bacterial counts. It was revealed that the decrease in bacterial counts was proportional to the incubation period. Accordingly, it was proved that the longer the period of immersion, the more the reduction in bacterial count.

Many index sites were selected. The reason was the change in bacterial count before and during treatment in the same mouth. In-vivo studies revealed an increase in both the *S. epidermidis* and *S. aureus* counts of the gingival sulcus during the course of the orthodontic treatment. The correlation between the increase in microbial count and duration of follow-up was insignificant at the upper canine and lower second molar. However, this increase was significant at the upper first molar, upper second molar, lower canine, and lower first molar [34]. These results agreed with those of Türkkahraman *et al.* [35] and disagreed with those of Petti *et al.* [36] and Speer *et al.* [37].

In other studies, *A. actinomycetemcomitans* increased significantly during the course of the orthodontic treatment. The correlation between the increase in microbial count and duration of follow-up was insignificant at the upper canine and lower canine. However, it was significant at the upper first molar, upper second molar, lower first molar, and lower second molar [35]. These results agreed with those of Ho [20], Paolantonio *et al.* [38], Sallum *et al.* [39], Türkkahraman *et al.* [35], Naranjo *et al.* [31], Leung *et al.* [33], Petti *et al.* [36,] and Thornberg *et al.* [18]. However, the results disagreed with those of Sinclair *et al.* [40] and Speer *et al.* [37].

In-vivo studies showed an absence of *Klebsiella* spp. before and 1 week after the fixed orthodontic treatment; it increased significantly along the course of the orthodontic treatment. The correlation between the increase in microbial count and duration of follow-up was significant at the upper canine, upper first molar, upper second molar, lower canine, and lower first and second molars. These results were in agreement with those of many other studies [35,38] and disagreed with those of others [37].

From all the previously mentioned results, specific bacteria found in periodontitis cases had to be collected from different index sites.

In this study no surface polishing of orthodontic wire or bracket was carried out by any common polishing methods, the reason being that wire-bracket couples are practically fixed in the oral cavity without surface polishing, which leads to real corrosion.

It was proved that galvanic corrosion potential varied with time of immersion in saline solution. In NiTi wire alloys and FeCrNi (StSt) wire alloys coupled with stainless-steel brackets, the corrosion potential moved in a more desirable direction with a longer immersion period; in contrast, the corrosion potential of the CuNiTi wire alloy coupled with FeCrNi moved in a less desirable direction after starting immersion [41]. In addition, no significant potential difference could be found between the FeCrNi wire and NiTi wire-coupled alloys [42].

The previously mentioned findings support the results of the current study. The longer the immersion time, the more the number of corrosion products released, and hence the more the bacterial count decreased. This explains why the bacterial count was intended to be equal when comparing the couple alloys. Moreover, our results are supported by the studies by Darabara *et al.* [12] and Schiff *et al.* [43].

From the present study, it was found that the orthodontic NiTi wire coupled with a stainless-steel bracket has the best corrosion resistance when compared with other specimens, whereas the orthodontic CuNiTi wire coupled with a stainless-steel bracket had the least corrosion resistance. These findings are in agreement with those of other studies [44–47]. This, in turn, reexplains the greater decrease in bacterial count after 3 weeks of immersion.

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

With regard to the CuNiTi wire, $TiCu_2$ is a phase present in the alloy. Copper constitutes the precipitated phase and plays an important role in the corrosion rate [48].

The orthodontic FeCrNi wire-coupled alloy showed the second highest decrease in bacterial count. This can be explained by the formation of passive film. This passive film is composed of Cr_2O_2, which precipitates on the surface of the FeCrNi wire and prevents further oxygen diffusion, resulting in increased corrosion resistance [49–51].

The NiTi wire-coupled alloy also formed a passive film that mainly consisted of several oxides of TiO_2, TiO, and Ti_2O_5, which proved to have good biocompatibility with the NiTi alloy. No significant difference was observed between the results of the present study and those reported in references [52–55].

The decrease in bacterial count was higher with rectangular wires than with round ones. This can be attributed to the larger surface area of the rectangular wire. A significant difference was found between the electrochemical parameters of both rectangular and round wires. The results revealed that both NiTi and FeCrNi orthodontic wires had a similar trend for corrosion resistance, but the CuNiTi wire showed less desirability [41].

Conclusion

(1) The longer the immersion period of the wire bracket couple, the greater the decrease in bacterial count.
(2) CuNiTi wires exhibited the highest decrease in bacterial counts, followed by FeCrNi wires and finally NiTi wires.
(3) Rectangular wires showed a greater decrease in bacterial counts compared with round wires.

Recommendations

Many factors are encountered when considering the oral environment. Among these are the different pH values and various temperatures caused by cold or hot intakes, besides normal ones. Accordingly, when an in-vivo study testing galvanic corrosion behavior is carried out, none of the factors must be overlooked.

Acknowledgements

The author would like to express his deep gratitude to those who assisted him in this work: Professor Dr Ibrahim Hamed Mahmoud, Professor of material engineering, Department of chemical engineering, Faculty of Engineering, Minia University; and Dr Mohammed Sayed Mohammed, Lecturer of microbiology and immunology, Faculty of medicine, Minia University.

Conflicts of interest

There are no conflicts of interest.

References

1 Kounoupis V, Tsalikis L, Dangalis P. Clinical and microbiological changes during orthodontic treatment and after appliance removal. *Hel Orthod Rev* 2006; **9**:103–121.
2 Glans R, Larsson E, Øgaard B. Longitudinal changes in gingival condition in crowded and noncrowded dentitions subjected to fixed orthodontic treatment. *Am J Orthod Dentofacial Orthop* 2003; **124**:679–682.
3 Petoumeno E, Kislyuk M, Hoederath H, Keilig L, Bourauel C, Jäger A. Corrosion susceptibility and nickel release of nickel titanium wires during clinical application. *J Orofac Orthop* 2008; **69**:411–423.
4 House K, Sernetz F, Dymock D, Sandy JR, Ireland AJ. Corrosion of orthodontic appliances – should we care? *Am J Orthod Dentofacial Orthop* 2008; **133**:584–592.
5 Staffolani N, Damiani F, Lilli C, Guerra M, Staffolani NJ, Beicastro S, *et al.* Ion release from orthodontic appliances. *J Dent* 1999; **27**:449–454.
6 Nielsen NH, Menné T. Nickel sensitization and ear piercing in an unselected Danish population. Glostrup Allergy Study. *Contact Dermatitis* 1993; **29**:16–21.
7 Schuster G, Reichle R, Bauer RR, Schopf PM. Allergies induced by orthodontic alloys: incidence and impact on treatment. Results of a survey in private orthodontic offices in the federal state of Hesse, Germany. *J Orofac Orthop* 2004; **65**:48–59.
8 Vreeburg KJJ, De Groot K, Von Blomberg M, Scheper RJ. Induction of immunological tolerance by oral administration of nickel and chromium. *J Dent Res* 1984; **63**:124–128.
9 Kerosuo H, Moe G, Hensten Pettersen A. Salivary nickel and chromium in subjects with different types of fixed orthodontic appliances. *Am J Orthod Dentofacial Orthop* 1997; **111**:595–598.
10 Fors R, Persson M. Nickel in dental plaque and saliva in patients with and without orthodontic appliances. *Eur J Orthod* 2006; **28**:292–297.
11 Widu F, Drescher D, Junker R, Bourauel C. Corrosion and biocompatibility of orthodontic wires. *J Mater Sci Mater Med* 1999; **10**:275–281.
12 Darabara MS, Bourithis LI, Zinelis S, Papadimitriou GD. Metallurgical characterization, galvanic corrosion and ionic release of orthodontic brackets coupled with Ni-Ti archwires. *J Biomed Mater Res B Appl Biomater* 2007; **81**:126–134.
13 McKay GC, Macnair R, MacDonald C, Grant MH. Interactions of orthopaedic metals with an immortalized rat osteoblast cell line. *Biomaterials* 1996; **17**:1339–1344.
14 Maruthamuthu S, Rajasekar A, Sathiyanarayanan S, Muthukumar N, Palaniswamy N. Electrochemical behaviour of microbes on orthodontic wires. *Curr Sci* 2005; **89**:988–996.
15 Von Fraunhofer JA. Corrosion of orthodontic devices. *Semin Orthod* 1997; **3**:198–205.
16 Al Hity RR, Kappert HF, Viennot S, Dalard F, Grosgogeat B. Corrosion resistance measurements of dental alloys, are they correlated? *Dent Mater* 2007; **23**:679–687.
17 Iijima M, Endo K, Yuasa T, Ohno H, Hayashi K, Kakizaki M, *et al.* Galvanic corrosion behavior of orthodontic archwire alloys coupled to bracket alloys. *Angle Orthod* 2006; **76**:705–711.
18 Thornberg MJ, Riolo CS, Bayirli B, Riolo ML, Van Tubergen EA, Kulbersh R. Periodontal pathogen levels in adolescents before, during and after fixed orthodontic appliance therapy. *Am J Orthod Dentofacial Orthop* 2009; **135**:95–98.
19 Kloehn JS, Pfeifer JS. The effect of orthodontic treatment on the periodontium. *Angle Orthod* 1974; **44**:127–134.
20 Ho HP, Niederman R. Effectiveness of the Sonicare® sonic toothbrush on reduction of plaque, gingivitis, probing pocket depth and subgingival bacteria in adolescent orthodontic patients. *J Clin Dent* 1997; **8**:15–19.
21 El Mishad AM. *Manual of practical microbiology*. 6th ed. Cairo: Al-Ahram press; 1998.
22 Park HY, Shearer TR. In vitro release of nickel and chromium from simulated orthodontic appliances. *Am J Orthod* 1983; **84**:156–159.
23 Kratzenstein B, Koppenburg P, Sauer KH, Geis Gerstorfer J, Weber H. Saliva analysis in orthodontic patients to prove content of dissolved metals. *Quintessenz* 1988; **39**:693–703.
24 Kerosuo H, Moe G, Kleven E. In vitro release of nickel and chromium from different types of simulated orthodontic appliances. *Angle Orthod* 1995; **65**:111–116.
25 Grimsdottir MR, Gjerdet NR, Hensten Pettersen A. Composition and in vitro corrosion of orthodontic appliances. *Am J Orthod Dentofacial Orthop* 1992; **101**:525–532.
26 Jia W, Beatty MW, Reinhardt RA, Petro TM, Cohen DM, Maze CR, *et al.* Nickel release from orthodontic arch wires and cellular immune response to various nickel concentrations. *J Biomed Mater Res* 1999; **48**:488–495.
27 Olsson S, Berglund A, Bergman M. Release of elements due to electrochemical corrosion of dental amalgam. *J Dent Res* 1994; **73**:33–43.
28 Kim H, Johnson JW. Corrosion of stainless steel, nickel–titanium, coated nickel–titanium and titanium orthodontic wires. *Angle Orthod* 1999; **69**:39–44.
29 Kedici SP, Aksut AA, Kiliçarslan MA, Bayramoglu G, Gokdemir K. Corrosion behavior of dental metals and alloys in different media. *J Oral Rehabil* 1998; **25**:800–808.
30 Tzannetou S, Efstratiadis S, Nicolay O, Grbic J, Lamster I. Interleukin-1beta and beta-glucuronidase in gingival crevicular fluid from molars during rapid palatal expansion. *Am J Orthod Dentofacial Orthop* 1999; **115**:686–696.
31 Naranjo AA, Triviño ML, Jaramillo A, Betancourth M, Botero JE. Changes in the subgingival microbiota and periodontal parameters before and 3 months after bracket placement. *Am J Orthod Dentofacial Orthop* 2006; **130**:275.e17–275.e22.

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.

32 Paolantonio M. Clinical significance of *Actinobacillus actinomycetemcomitans* in young individuals during orthodontic treatment: a 3-year longitudinal study. *J Clin Periodontol* 1997; **24 (9 Pt 1):**610–617.

33 Leung NM, Chen R, Rudney JD. Oral bacteria in plaque and invading buccal cells of young orthodontic patients. *Am J Orthod Dentofacial Orthop* 2006; **130**:698.e11–698.e18.

34 El Shal MG. *Changes in the subgingival microbiota before and during fixed orthodontic treatment*. MSc (Master) thesis, Faculty of Dentistry, Minia University; 2009.

35 Türkkahraman H, Sayin MÖ, Bozkurt FY, Yetkin Z, Kaya S, Önal S. Archwire ligation techniques, microbial colonization and periodontal status in orthodontically treated patients. *Angle Orthod* 2005; **75**:231–236.

36 Petti S, Barbato E, Simonetti D'Arca A. Effect of orthodontic therapy with fixed and removable appliances on oral microbiota: a six-month longitudinal study. *New Microbiol* 1997; **20**:55–62.

37 Speer C, Pelz K, Hopfenmüller W, Holtgrave EA. Investigations on the influencing of the subgingival microflora in chronic periodontitis. A study in adult patients during fixed appliance therapy. *J Orofac Orthop* 2004; **65**:34–47.

38 Paolantonio M, Festa F, di Placido G, D'Attilio M, Catamo G, Piccolomini R. Site-specific subgingival colonization by *Actinobacillus actinomycetemcomitans* in orthodontic patients. *Am J Orthod Dentofacial Orthop* 1999; **115**:423–428.

39 Sallum EJ, Nouer DF, Klein MI, Gonçalves RB, MacHion L, Wilson Sallum A, *et al*. Clinical and microbiologic changes after removal of orthodontic appliances. *Am J Orthod Dentofacial Orthop* 2004; **126**:363–366.

40 Sinclair PM, Berry CW, Bennett CL, Israelson H. Changes in gingiva and gingival flora with bonding and banding. *Angle Orthod* 1987; **57**:271–278.

41 Hamed EI. *Effect of contact between dissimilar alloys used in orthodontics on galvanic corrosion behavior in simulating oral media*. MSc (Master) thesis, Faculty of Dentistry, Minia University; 2011.

42 Pun DK, Berzins DW. Corrosion behavior of shape memory, superelastic and nonsuperelastic nickel-titanium-based orthodontic wires at various temperatures. *Dent Mater* 2008; **24**:221–227.

43 Schiff N, Boinet M, Morgon L, Lissac M, Dalard F, Grosgogeat B. Galvanic corrosion between orthodontic wires and brackets in fluoride mouthwashes. *Eur J Orthod* 2006; **28**:298–304.

44 El Medawar L, Rocher P, Hornez JC, Traisnel M, Breme J, Hildebrand HF. Electrochemical and cytocompatibility assessment of NiTiNOL memory shape alloy for orthodontic use. *Biomol Eng* 2002; **19 (2–6):** 153–160.

45 Rondelli G, Vicentini B. Localized corrosion behaviour in simulated human body fluids of commercial Ni-Ti orthodontic wires. *Biomaterials* 1999; **20**:785–792.

46 Speck KM, Fraker AC. Anodic polarization behaviour of Ti-Ni and Ti-6Al-4V in simulated physiological solutions. *J Dent Res* 1980; **59**:1590–1595.

47 Sedriks AJ, Green JAS, Novak DL. Electrochemical behavior of Ti-Ni alloys in acidic chloride solutions. *Corrosion* 1972; **28**:137–142.

48 Davis JR. *Corrosion: understanding the basics*. ASM International, Park OH. *Am Soci Metals* 2000: 33–34.

49 Huang HH, Lee TH, Huang TK, Lin SY, Chen LK, Chou MY. Corrosion resistance of different nickel-titanium archwires in acidic fluoride-containing artificial saliva. *Angle Orthod* 2010; **80**:547–553.

50 Walker MP, Ries D, Kula K, Ellis M, Fricke B. Mechanical properties and surface characterization of beta titanium and stainless steel orthodontic wire following topical fluoride treatment. *Angle Orthod* 2007; **77**: 342–348.

51 Eliades T, Athanasiou AE. In vivo aging of orthodontic alloys: implications for corrosion potential, nickel release and biocompatibility. *Angle Orthod* 2002; **72**:222–237.

52 Peitsch T, Klocke A, Kahl Nieke B, Prymak O, Epple M. The release of nickel from orthodontic NiTi wires is increased by dynamic mechanical loading but not constrained by surface nitridation. *J Biomed Mater Res A* 2007; **82**:731–739.

53 Lin MC, Lin SC, Lee TH, Huang HH. Surface analysis and corrosion resistance of different stainless steel orthodontic brackets in artificial saliva. *Angle Orthod* 2006; **76**:322–329.

54 Huang HH. Surface characterizations and corrosion resistance of nickel-titanium orthodontic archwires in artificial saliva of various degrees of acidity. *J Biomed Mater Res A* 2005; **74**:629–639.

55 Hwang CJ, Shin JS, Cha JY. Metal release from simulated fixed orthodontic appliances. *Am J Orthod Dentofacial Orthop* 2001; **120**:383–391.

Copyright © The Egyptian Association of Oral and Maxillofacial Surgeons. Unauthorized reproduction of this article is prohibited.