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# Surface Analysis and Metal Release of Titanium Alloy Orthodontic Mini-Screw Implants: A Clinical Prospective Study

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## ABSTRACT

**Objective:** To investigate surface lustrous, morphological, and compositional changes of retrieved orthodontic mini-screw implants (MSIs) and to asses potential metal ion release.

**Materials and methods:** Ten orthodontic mini-screw implants (Tomas-Pin SD, Dentaurum, Ispringen, Germany) placed in 5 female orthodontic patients (16.11 ± 1.68 years) to serve as anchorage for maxillary canine retraction. The MSIs were retrieved after successful service for 4 months. Digital microscope, scanning electron microscope and X-ray fluorescence spectrometer were used for analysis of surface gloss, morphology, and composition of MSIs, respectively before and after usage. At regular scheduled visits the following were recorded: unstimualated salivary pH; levels of titanium, aluminum and vanadium ions in unstimulated saliva.

**Results:** Surface elemental analysis showed significant calcium mass % increase in the head portion and significant ferrous mass % increase in the thread portion of MSIs. Scanning electron microscope examination of MSIs displayed surface changes in the form of dullness, dents of threads and tips, and to less extent corrosion, craters.

**Conclusion:** The head portion of all MSIs showed degradation after clinical use characterized by loss of the original color and gloss. The surface analysis of retrieved MSIs differed from that of as-received MSIs, with additional elements; the most noticeable were calcium and phosphorus (contact with saliva and oral fluid) in the head portion and iron in the thread portion (contact with blood and tissue fluid) of MSI ($P<0.05$). Salivary contents of Ti, Al, V did not increase after insertion of MSIs.

**Keywords:** Canine retraction, Mini-screw implants, Metal release, Surface analysis

## INTRODUCTION

Clinical studies have suggested that mini-screw implants (MSIs) may provide stable anchorage during the orthodontic treatment without requiring patient cooperation. These studies proved many successful applications in orthodontics involving; retraction of anterior teeth, correction of open bites, distalization, mesialization and intrusion of teeth [1-5].

Various materials were used in implant systems. The implant material must be nontoxic, biocompatible, mechanically sufficient, and having high tension and corrosion resistance. Commercially pure titanium (cp Ti) was the most used material in prosthetic implants because of its; high biocompatibility, high corrosion resistance in body fluids, not allergic, high specific strength and low elastic modulus when compared with other metallic biomaterials [1,4-7].

On the other hand, orthodontic MSIs are smaller than conventional prosthetic implants and should resist high orthodontic loads. These factors contribute to the possible fracture of cp Ti MSIs during placement, use and removal. To avoid such fracture, Ti alloy implants were composed through the addition of aluminum (6Al) and vanadium (4V), (Ti-6Al-4V), for adding strength and fatigue resistance than cp Ti [4,7,8]. Unfortunately, this alloy has a low corrosion resistance and can result in corrosion of the orthodontic MSIs in body fluids [9,10].

Saliva, oral fluids, chemicals introduced into the oral cavity through food and drink, the enzymatic behavior, microbial activity, pH fluctuation, temperature fluctuation and host - mini-screw implant interaction could present a brutal

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condition for the titanium alloy giving rise to potential surface corrosion and metal ion release [9,10]. So, the study was conducted to investigate the effects of clinical usage on surface lustrous, morphological, and compositional characteristics of retrieved orthodontic MSIs. Also, to investigate metal ions (titanium, aluminium and vanadium) released in saliva after insertion of MSIs.

## SUBJECTS AND METHODS

The sample was consisted of 10 orthodontic mini-screw implants (MSIs) placed in 5 female orthodontic patients between 14 and 18 years (16.11 ± 1.68 years). Mini-screw implants were installed to serve as anchorage for maxillary canine retraction after maxillary first premolar extraction. Patients consented to participate in the study which ran for 4 months. Ethical approval was obtained from the regional committee for medical and health research ethics, faculty of Medicine (boys), Al-Azhar University, Cairo, Egypt. The study was registered in clinical trials.gov identified by the following number (NCT03460132).

Sample size calculation was based on power of statistics analysis, using the standard deviation in previous studies [11-13]. The estimated sample size for a one-sample proportion test (score z-test) was calculated to be 6 orthodontic mini-screw implants.

The inclusion criteria: 1) patients indicated for bilateral extraction of maxillary first premolar, 2) Good oral and general health (Free of any disease that could affect oral conditions), and 3) All teeth should be free from caries or any metallic restorations.

Patients with history of occupational exposure to metals, seriously fractured appliance, deteriorated oral hygiene condition, and/or broken appointment that affect the progress of treatment or study protocol were excluded.

For each patient, non-metallic fixed orthodontic appliance consisted of: 1) direct bond, pre-adjusted Roth ceramic brackets (0.022" × 0.028"), 2) teflon-coated nickel titanium round arch wires (0.012", 0.014", 0.016", 0.018") for the leveling and alignment stage and teflon-coated stainless steel arch wires (0.018" or 0.016" × 0.022") for the canine retraction, 3) the arch wires were ligated by clear latex-free elastic O-ties, 4) Self-drilling (SD) orthodontic MSI (Tomas-Pin SD, Dentaurum, Ispringen, Germany) (1.6 mm in diameter, 8 mm in thread length, gingival collar (2.8 mm maximum diameter, 2 mm in height) and a head with cross slot 2.25 mm in height) was used as an anchorage for maxillary canine retraction [4].

An insertion guide was used to standardize the receptor site between maxillary first molar and second premolar. The wire was inserted into the first molar bracket with a coil at the other end of the wire to locate accurately the vertical (6-8 mm from the bracket slot apically) and anterio-posterior (center of inter-dental bone) position of the MSI. The MSI was directed perpendicular to the buccal alveolar bone surface.

Canine retraction was started simultaneously in both sides, immediately after MSIs installation, by extending elastomeric chain (clear, short) between the head of the MSIs and the maxillary canine hooks [14]. Activation was done every three weeks (the elastic chain replaced by a new one) to have standardized force all over the experimental period. The initial force should be in the range of 200-250 g to obtain the standardized canine retraction force (approximately 150 g) after the first day sharp decay of force [15].

All MSIs were removed either after four months of the experimental period and/or complete retraction of one of the canines into the extraction space by the application of counter clockwise torque load with the screwdriver supplied from the manufacturer [14]. Each MSI was placed in sterile glass rubber-seal top test tube containing 1 ml of saline solution for cleaning using vortex vibrator. After removal of MSIs, the orthodontic treatment was continued according to the proposed line of treatment for each patient.

**Weight changes** were measured using an electronic sensitive balance by weighting each MSI before placement and after removal. Each measurement was recorded twice and the average weight to the nearest ± 0.01 mg was used as the result [16].

**Saliva samples** were collected immediately before MSIs insertion, one week, one month, three months and four months after MSIs insertion to measure salivary potential of hydrogen (pH) and to analyze the amount of metal ions (titanium, aluminium and vanadium) released from the MSIs.

The sample was collected in the morning after usual breakfast and tooth brushing. The patients rinsed her mouth thoroughly with 100 ml distilled water for 1 min, expectorate and then wait for a while until saliva accumulated in her mouth. Approximately 4 ml of non-stimulated saliva was collected in a sterile, glass rubber seal-top test tube, then coded and stored at -20°C in a freezer [16,17].

**Microcomputer based pH/temperature bench meter** (Model 6173/6173R, JENCO Electronics, Shanghi, China) was used for analysis of salivary pH. Each measurement was recorded twice and the average pH value to the nearest ± 0.01 was used as the result [16-18].

**Inductively coupled plasma optical emission spectroscopy** (ICP-OES) (Prodigy High Dispersion ICP, Leeman, USA) was used to determine the initial salivary concentration of titanium, aluminium and vanadium and the concentration of these metal ions drained into the salivary solution after MSIs insertion [6,7,17-19].

**Surface color changes** were evaluated by imaging MSIs before and after placement using digital microscope (DM-UM012A-1.3m USB Digital Microscope with 8 LED Lights, Guangdong, China). The images **(Figure 1)** were imported into image processing software (Image J, National Institute of Health NIH, USA) which detect the luminance or lightness component, ranging from (0 to 100). At least 10 points in each part of MSI (head, thread and tip) were analyzed and the average was taken as the result [11,20].

**Scanning electron microscope** (JEOL-JSM 5600 LV) was used for scanning MSI surfaces before and after insertion for the detection and imaging (magnification=50x) of surface roughness, craters and integument adhered to any part of MSIs **(Figure 2)**. Operating parameters for ICP-OES: (1.2 kW) RF power, (19 L/min) coolant Flow, (34 psi Concentric) nebulizer Pressure, (0.7 l/min Argon) purge gas flow, (0.8L/m) auxiliary flow [11,21].

The surface's qualitative **elemental microanalysis (Figure 3)** of each MSI was determined before and after usage with X-ray fluorescence spectrometer (XRF) (JEOL, JSX-3222 analyzer) [11,22].

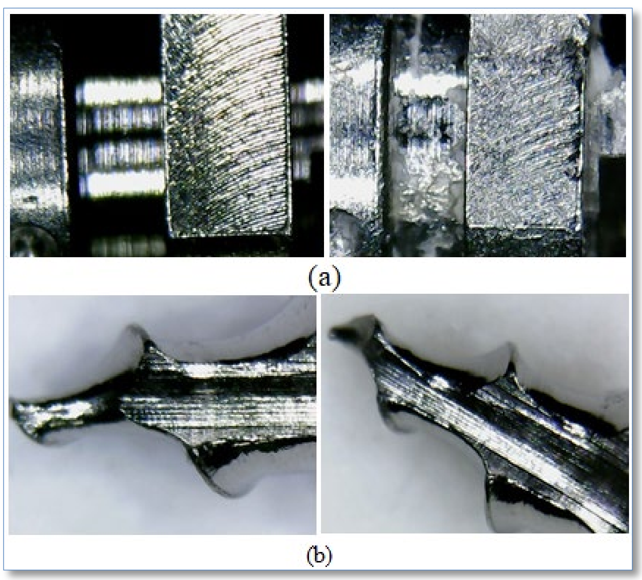


**Figure 1.** Digital microscopic image showing orthodontic mini-screw implant before insertion (left side) and after removal (right side): a) Head, and b) Tip.

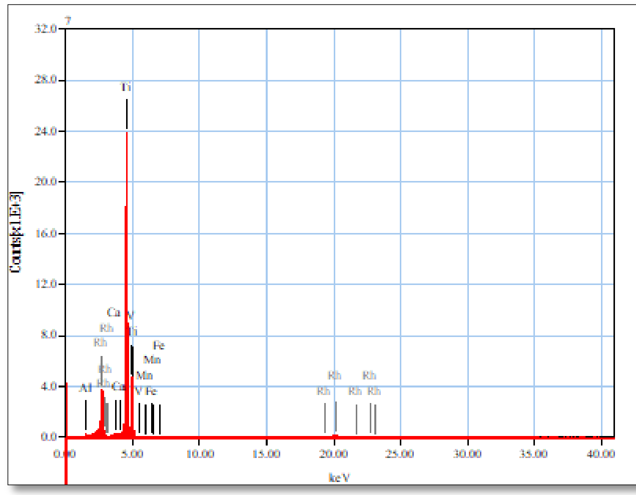


**Figure 2.** Graphical representation of surface composition of orthodontic mini-screw implants by X-ray fluorescence.

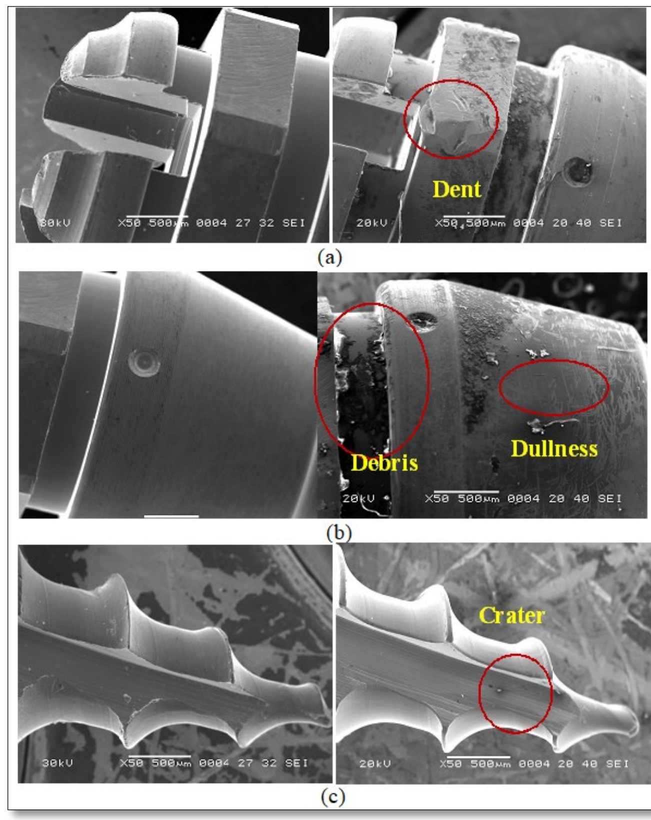


**Figure 3.** Scanning electron microscopic image showing the morphology and surface changes of MSI before insertion (left side) and after removal (right side): a) Head, b) Collar, and c) Tip.

## STATISTICAL ANALYSIS

The collected data was tabulated and statistically analyzed using S.P.S.S. (SPSS 18(2009); SPSS Inc., Chicago, IL, USA). The data was analyzed using t-test and ANOVA test followed by Tukey's post hoc test when ANOVA revealed a significant difference ($p \leq 0.05$).

**RESULTS**

The weight of MSI (mg) was increased after removal **(Table 1)**. The mean difference was (0.185 mg). Paired t-test revealed that the difference was statistically insignificant (p=0.80).

**Table 1.** Paired t test showing the weight change of MSI (mg) before insertion and after removal.

| n=10 | Mean (mg) | SD | SE Mean | Mean Diff | SD Diff | SE Diff | t | P-value |
|---|---|---|---|---|---|---|---|---|
| Before insertion | 106.38 | 1.60 | 0.51 | 0.185 | 0.29 | 0.71 | 0.26 | **0.80 ns** |
| After removal | 106.56 | 1.56 | 0.49 | | | | | |

*n: number of MSI; SD: Standard Deviation; SE: Standard Error; Diff: difference; Significance level $p \leq 0.05$, ns: non-significant*

ANOVA test revealed that the difference in pH level **(Table 2)** was statistically significant (p<0.05*). The mean salivary pH before insertion of MSIs was 7.46 which become more acidic after one week 6.98, one month 6.90 and four months 6.81.

**Table 2.** Salivary pH and trace elements (titanium, aluminum and vanadium) in different observation times (ANOVA test).

| (n=5) | Salivary pH | | | Titanium | | | Aluminum | | | Vanadium | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Mean | SD | P-value | Mean (PPM) | SD | P-value | Mean (PPM) | SD | P-value | Mean (PPM) | SD | P-value |
| Before insertion | 7.46$^{a}$ | 0.24 | | 0.07$^{a}$ | 0.02 | | 9.17$^{a}$ | 3.05 | | 0.01 | 0.00 | |
| One week | 6.98$^{b,c}$ | 0.20 | | 0.07$^{a}$ | 0.02 | | 12.31$^{a}$ | 3.95 | | 0.009 | 0.007 | |
| One month | 6.90$^{b,c}$ | 0.22 | <0.05* | 0.09$^{a}$ | 0.03 | <0.05* | 5.27$^{b}$ | 1.79 | <0.05* | 0.005 | 0.003 | 0.13$^{ns}$ |
| Three months | 7.16$^{a,b}$ | 0.16 | | 0.01$^{b}$ | 0.00 | | 0.16$^{c}$ | 0.04 | | ND | | |
| Four months | 6.81$^{c}$ | 0.45 | | 0.01$^{b}$ | 0.00 | | 0.28$^{c}$ | 0.08 | | ND | | |

*SD: Standard Deviation; Significance level $p \leq 0.05$; ns: non-significant; *: significant*
*PPM: Part Per Million; ND: Not Detected*

Tukey's post hoc test: means sharing the same superscript letter were not significantly different

Salivary titanium concentration increased at 1 month and then significantly decreased at 3 months and after removal. ANOVA test revealed that the difference in salivary (Ti) concentration was statistically significant (P<0.05*). Tukey's post hoc test revealed statistically insignificant increase in salivary titanium concentration at one month and statistically significant decrease after three, four months of MSIs insertion.

Salivary aluminium concentration **(Table 2)** increased at 1 week, then significantly decreased at 1 month, then further decreased at 3 months and after removal. ANOVA test revealed that the difference in salivary (Al) concentration was statistically significant (P<0.05*). Tukey's post hoc test revealed statistically insignificant increase in salivary aluminium concentration at one week and statistically significant decrease after one, three and four months of MSIs insertion.

Salivary vanadium concentrations **(Table 3)** were gradually decreased at 1 week, 1 month and were not detectable thereafter. ANOVA test revealed that the difference in salivary (V) concentration was statistically insignificant (p=0.13).

**Table 3.** Paired t test showing the difference in surface luminance of orthodontic MSIs (head, thread and tip) between before insertion and after removal.

| n=10 | Mean (0-100) | SD | SE Mean | Mean Diff | SD Diff | SE Diff | t | P-value |
|---|---|---|---|---|---|---|---|---|
| Before insertion (Head) | 58.84 | 10.78 | 3.41 | 20.99 | 18.78 | 5.94 | 3.53 | **<0.05*** |
| After removal (Head) | 37.85 | 15.06 | 4.76 | | | | | |
| Before insertion (Thread) | 56.81 | 9.69 | 3.06 | -4.18 | 16.55 | 5.23 | -0.80 | **0.45 ns** |
| After removal (Thread) | 60.99 | 14.03 | 4.44 | | | | | |
| Before insertion (Tip) | 71.30 | 15.76 | 4.98 | 5.17 | 11.58 | 3.66 | 1.41 | **0.19 ns** |
| After removal (Tip) | 66.13 | 7.36 | 2.33 | | | | | |

*n: number of MSI; SD: Standard Deviation; SE: Standard error*
*Diff: difference; Significance level $p \leq 0.05$; ns: non-significant; *: Significant*

The difference in surface luminance or lightening (indicator of surface finish and gloss) between before insertion of MSIs and after removal in three different parts of MSIs (head, threads and tip) had been shown in **Table 4**. The table showing statistically significant difference in the luminance of the head of MSIs (P<0.05*) and non-significant difference in the thread (p=0.45) and tip (p=0.19) of MSIs.

**Table 4.** Frequency of surface changes on orthodontic MSI assessed by scanning electron microscope (percentage %).

| n=10 | Head | Collar | Body | Tip |
|---|---|---|---|---|
| Bone like materials | NA | NA | 70% | 80% |
| Debris | 100% | 90% | NA | NA |
| Dullness | 100% | 100% | 50% | 50% |
| Cracks | 0% | 0% | 0% | 0% |
| Craters | 0% | 10% | 10% | 20% |
| Fracture | 0% | 0% | 0% | 0% |
| Dent | 60% | 0% | 30% | 30% |

*n: number of MSI; NA: Not Applicable*

Surface elemental composition analysis of MSIs (X-ray fluorescence spectrometer) revealed insignificant increase (paired t-test) in calcium ms % in head portion and phosphorus ms % in thread portion of MSIs **(Table 5)**. Paired t test also revealed statistically significant increase in ferrous ms % on the thread portion of MSIs.

**Table 5.** Paired t test showing the difference in X-ray fluorescence surface elemental analysis (ms %) before insertion and after removal in head and threads.

| | **(n=10)** | **Mean (ms %)** | **SD** | **Mean Diff** | **P-value** | | **(n=10)** | **Mean (ms %)** | **SD** | **Mean Diff** | **P-value** |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Titanium** | Before insertion (Head) | 91.70 | 91.70 | 0.79 | $0.27^{ns}$ | Aluminum | Before insertion (Head) | 3.48 | 0.21 | 0.05 | $0.56^{ns}$ |
| | After removal (Head) | 90.91 | 90.91 | | | | After removal (Head) | 3.53 | 0.16 | | |
| | Before insertion (Threads) | 91.70 | 91.70 | 0.68 | $0.38^{ns}$ | | Before insertion (Threads) | 3.48 | 0.21 | 0.24 | $0.16^{ns}$ |
| | After removal (Threads) | 91.02 | 91.02 | | | | After removal (Threads) | 3.25 | 0.45 | | |
| **Vanadium** | Before insertion (Head) | 4.65 | 0.45 | 0.16 | $0.29^{ns}$ | Calcium | Before insertion (Head) | 0.07 | 0.09 | 0.52 | $0.12^{ns}$ |
| | After removal (Head) | 4.81 | 0.10 | | | | After removal (Head) | 0.59 | 0.96 | | |
| | Before insertion (Threads) | 4.65 | 0.45 | 0.05 | $0.75^{ns}$ | | Before insertion (Threads) | 0.07 | 0.09 | 0.07 | <0.05* |
| | After removal (Threads) | 4.70 | 0.18 | | | | After removal (Threads) | 0.00 | 0.00 | | |
| **Phosphorus** | Before insertion (Head) | 0.00 | 0.00 | 0.00 | $1.00^{ns}$ | Ferrous | Before insertion (Head) | 0.20 | 0.08 | 0.02 | $0.77^{ns}$ |
| | After removal (Head) | 0.00 | 0.00 | | | | After removal (Head) | 0.22 | 0.24 | | |
| | Before insertion (Threads) | 0.00 | 0.00 | 0.15 | $0.16^{ns}$ | | Before insertion (Threads) | 0.20 | 0.08 | 0.22 | <0.05* |
| | After removal (Threads) | 0.15 | 0.30 | | | | After removal (Threads) | 0.42 | 0.14 | | |

*n: number of MSI; ms %: mass percentage; SD: Standard Deviation; Diff: difference; Significance level $p \leq 0.05$; ns: non-significant*

## DISCUSSION

The weight gain of orthodontic MSI after four months of clinical use could be attributed to the accumulation of food debris on the head of MSIs and foreign body adherence on the threads of MSIs. To our knowledge after searching the scientific literature, no data were available for evidence about the weight changes of orthodontic mini-screw implants after usage.

Saliva has a dynamic structure that could be affected by many physiologic variables. Temperature, quantity and quality of saliva, salivary pH, plaque and amount of protein in saliva, physical and chemical properties of food and liquids and general and oral health conditions were factors that could influence orthodontic MSI corrosion in the oral cavity [19].

In an effort to report these concerns, in the current study, the saliva was collected from the same patients who did not have any health problems or any caries in the mouth and good oral hygiene. All the samples were taken in the morning before breakfast and the patients rinsed their mouths thoroughly with distilled water before the collection. Salivary pH was measured for each patient before insertion of MSIs, one week, one month, three months and four months of MSIs insertion in order to analyze a reported factor that could increase corrosion of titanium alloy MSIs. Although the changes in salivary pH were statistically significant, it was considered within the neutral pH condition [3].

Metal ions released were analyzed using inductively coupled plasma optical emission spectrophotometry (ICP-OES) which was a common, documented and very sensitive method used for trace element analysis in the literature. [19,23].

The clinical usage of orthodontic mini-screw implants appeared to do not significantly affect the salivary concentrations of titanium, aluminium and vanadium. These were assured by the insignificant increase in the salivary concentrations of titanium and aluminium after one week of MSI insertion. The decrease in salivary concentrations of titanium, aluminium and vanadium could be attributed to the precipitation of calcium/phosphorus minerals from the body fluid into the titanium surface, which decreases the ion release [24].

Retrieved MSIs presented loss of gloss and surface finish, resulting in a dull surface in head portion of all 4 examined zones. It was assumed that insertion and removal of MSIs causes the surface to wear out to some extent. Blunting of threads and tip (less sharp thread and tip compared with as-received screws) was also linked to wear during the process of insertion and removal [11,25,26].

Titanium alloys used to manufacture MSIs were less resistant to corrosion because the alloys represent discontinuities in the protective oxide film [25]. The surface milling and polishing defects during the manufacturing process were seen in the form of stripes and scratches.

These tiny defects can be a starting point for electrochemical attack when mini-screws were inserted in the body [27]. Although titanium alloys have been considered highly corrosion resistant because of the stable passive titanium oxide layer on the surface, they were not inert to corrosive attack. When the stable surface oxide layer broken down or removed and cannot reform on parts of the surface, titanium can be corrosive, as many other base metals [28].

The current study revealed statistically insignificant difference in the main component of orthodontic mini-screw implants (titanium, aluminium and vanadium), statistically insignificant increase in the amount of calcium in the thread portion of MSI and statistically significant increase ferrous ms % in the head of MSI. These results indicate substantial changes in the surface profile of mini-screw implants including adsorption of a calcified integument from the contact of the material with saliva, food, blood, biological fluids and tissue fluids. The presence of iron and calcium derives from the contact of implant surface with blood and agrees with research on biomedical materials including cardiac valves and orthopedic prostheses [11,25].

## CONCLUSION

1) Despite the statistically significant decrease in salivary pH, it was considered within the neutral pH.
2) Salivary contents of Ti, Al, V did not increased after insertion of MSIs.
3) The head portion of all MSIs showed degradation after clinical use characterized by loss of the original color and gloss.
4) Scanning electron microscope examination of MSIs displayed surface changes in the form of dullness, dents of threads and tips and to less extent corrosion, craters.
5) The surface elemental composition of retrieved MSIs differed from that of as-received MSIs, with additional elements; the most noticeable were calcium (contact with saliva and oral fluid) in the head region, iron and phosphorus in the thread region (contact with blood and tissue fluid) of MSI ($P<0.05$).

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