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Chemical analysis of the surface of dental implants after contact with surgical gloves

Análise química da superfície de implantes dentários após contato com luvas cirúrgicas

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ABSTRACT
Objective: To perform a chemical analysis of the surface of dental implants after contact with surgical gloves by scanning electron microscopy and energy dispersive spectroscopy (EDS) and to correlate the findings with literature data. Materials and methods: Five commercially pure titanium implants submitted to surface treatment by acid attack and one pair of sterile latex gloves were selected. First, the implant and gloves were analyzed separately by EDS in order to observe the chemical elements on their surfaces. Next, the implant surface was put in contact with the surgical glove and analyzed by EDS to identify possible contaminants left by the latex glove. Results: Calcium, Zn, Mg, S, Si, and Nb were detected on the surface of the surgical glove, with Ca and Zn being the predominant elements in all samples. Titanium was detected on all implants; two implants only exhibited this element as described by the manufacturer and three contained small amounts of Ca, but Ti was the prevalent element. After contact of the implants with the surgical glove, elements that had not been detected on the gloves and implants

during separate analysis, such as S, Zn, Si and Ca, were found. Conclusion: The present results and literature data permit us to conclude that the surface of titanium implants was contaminated after contact with surgical gloves and that inorganic chemical elements can modify the titanium oxide layer, resulting in possible interferences with the process of osseointegration.

Keywords: Dental Implant, Topography, Contamination.

RESUMO
Objetivo: Realizar uma análise química da superfície dos implantes dentários após o contato com luvas cirúrgicas por microscopia eletrônica de varredura e espectroscopia de energia dispersiva (EDS) e correlacionar os achados com os dados da literatura. Materiais e métodos: Cinco implantes de titânio comercialmente puros submetidos a tratamento de superfície por ataque ácido e um par de luvas de látex estéreis foram selecionados. Primeiro, o implante e as luvas foram analisados separadamente pela EDS para observar os elementos químicos em suas superfícies. Em seguida, a superfície do implante foi colocada em contato com a luva cirúrgica e analisada pela EDS para identificar possíveis contaminantes deixados pela luva de látex. Resultados: Cálcio, Zn, Mg, S, Si e Nb foram detectados na superfície da luva cirúrgica, sendo Ca e Zn os elementos predominantes em todas as amostras. O titânio foi detectado em todos os implantes; dois implantes exibiram apenas esse elemento como descrito pelo fabricante e três continham pequenas quantidades de Ca, mas o Ti era o elemento predominante. Após o contato dos implantes com a luva cirúrgica, foram encontrados elementos que não haviam sido detectados nas luvas e implantes durante análises separadas, como S, Zn, Si e Ca. Conclusão: Os presentes resultados e dados da literatura permitem concluir que a superfície dos implantes de titânio foi contaminada após o contato com luvas cirúrgicas e que elementos químicos inorgânicos podem modificar a camada de óxido de titânio, resultando em possíveis interferências no processo de osseointegração.

Palavras-chave: Implante Dentário, Topografia, Contaminação

1 INTRODUCTION

Osseointegration depends on the surface qualities of the implants and the biocompatibility and affinity of bone tissue to oxides (TiO_2) formed on the surface of dental implants made from pure titanium have been demonstrated [1,2].

The properties of dental implants such as topographic morphology and chemical composition of the implant surface will affect the nature of the layer of proteins that adsorb to the surface and mediate osteogenesis. Similarly, these properties will influence osseointegration and control the biological response to the biomaterial. It is therefore possible to regulate this response, rendering implants more biocompatible [1-6].

According to Machnee et al.[5], the composition of the oxide formed on commercially pure titanium by reacting with air provides stability and fracture and corrosion resistance. Titanium dioxide (TiO_2) is the primary oxide, followed by titanium monoxide (TiO) and titanium trioxide (Ti_2O_3). Thus, the host tissue only comes in contact with the oxide layer and not with titanium. The oxide layer has been described by some authors to be very thin (2 to 6 nm) and to contain a contaminating layer of nitrogen (N), calcium (Ca), phosphorus (P), chlorides (Cl^-) and sodium (Na), among others[7-9].

When the implant has a high surface energy, at least in theory, the proliferation of cells, adsorption of proteins, lipoproteins and peptides, and deposition of molecules such as calcium and phosphate are increased. Interactions occur between positively charged amino acids and the negative surface of the titanium dioxide layer through electrostatic forces, or between groups of amino acids with Ca^{2+} bridges that were previously adsorbed by the negative surface of the titanium dioxide layer. As a consequence, osseointegration is stronger for implants with a high surface energy compared to implants with a low surface energy[10-12].

During the manufacturing process, grit blasting with alumina particles used to modify the implant surface may lead to the adsorption of impurities such as aluminum which are difficult to remove[6,13]. During sterilization, fluorine, chlorine and iron can contaminate the implants[14]. According to Kasemo[15], autoclave sterilization is responsible for an additional increase in the superficial oxide layer, in addition to the possible incorporation of hydroxyl radical (OH^-). Furthermore, impurities and contamination can influence the nature of the protein layer adsorbed onto the implant surface, which will subsequently mediate bone formation. The presence of these elements impairs the adhesion of biomolecules and of cells essential for osseointegration, such as epithelial cells, fibroblasts and osteoblasts, and interferes with the healing process, causing the decalcification of bone tissue[15,16]. However, many commercially available implants already contain impurities such as carbon and nitrogen in their original packaging[17].

Intercurrences can occur during the installation of dental implants; for example, release of the implant from the titanium tweezers or the need for reinsertion of the implant to increase the entry hole in bone[14]. These intercurrences may require the implantodontist to hold the implant with gloves to facilitate its handling. This contact, in turn, can lead to the adsorption of inorganic contaminants on the implant surface which may interfere with osseointegration[17].

The aim of the present study was to perform a chemical analysis of the surface of dental implants after contact with surgical latex gloves by scanning electron microscopy (SEM) and energy dispersive spectroscopy (EDS), and to correlate the results with literature data on osseointegration.

2 MATERIALS AND METHODS

Selection of the implants and surgical gloves

Five commercially pure titanium implants (Conexão Sistemas de Prótese®) submitted to surface treatment by acid attack were used in this study. The implants were provided by the manufacturer in specific sealed and sterile bags. Table 1 shows the specifications of the manufacturer.

Brand	Implant	Surface treatment	Batch	Validity	Sterilization method
Conexão	Master Porous®	Acid attack	12872	May 2016	Gamma radiation

Table 1: Manufacturer specifications of the implant.

One pair of sterile surgical latex gloves (Madeitex® Indústria e Comércio de Artefatos de Látex Ltda.) were used. The gloves were stored in a specific sealed and sterile package and were within the period of validity. Table 2 shows the manufacturer specifications.

Brand	Type	Batch	Validity	Sterilization method
MadeitexGama	Latex gloves	R0228	February 2014	Cobalt 60 radiation

Table 2: Manufacturer specifications of the surgical gloves.

Scanning electron microscopy-energy dispersive spectroscopy analysis

EDS coupled with SEM was used for analysis. All samples were analyzed using a Tescan Vega 3 LMU scanning electron microscope. For EDS, the X-act system from Oxford Instruments (model 51-ADD0007) of the Federal Institute of Bahia (IFBA) was used. First, separate chemical analysis of the implant and gloves was performed by EDS. Next, the implant surface was put in contact with the surgical glove and analyzed by EDS to identify possible contaminants left by the glove.

Chemical analysis of the gloves

The box containing the pair of sterile latex gloves was opened carefully through its tabs without any contact with the gloves in order to avoid their contamination. One glove was removed from the package with sterile tweezers and cut into five pieces with sterile scissors. These samples were taken to a metal sputterer and then to a spectrometer for chemical analysis. An area of 900 µm in the center of the sample was analyzed. The other glove of the pair was reserved for contact with the implant.

Chemical analysis of the implant

The implants were removed from the packaging, placed directly on the sample holder, and manipulated with sterile titanium tweezers to avoid contamination of their surface. After this procedure, the implants were transferred to the spectrometer for chemical analysis. Three areas (4^{th}, 7^{th} and 10^{th} thread) per implant were analyzed over a spectrum of 60 µm. After analysis, the implant was removed from the apparatus and its surface was put in contact with the surgical glove (Figure 1).

Chemical analysis of the implant surface + glove

After separate analysis of the implant and glove, contact between the two was established as follows: the researcher put on one glove from the same manufacturer and batch and held the implant at its titanium base for one minute with light circular friction movements, simulating the possible contact a professional would have when accidentally touching the implant with gloves. After contact, the implant was placed on the sample holder and submitted to chemical analysis in the spectrometer. Three areas (corresponding to the 4^{th}, 7^{th} and 10^{th} thread) were analyzed per implant over a spectrum of 60 µm (Figure 2).

3 RESULTS

For all samples, the chemical composition of the titanium implant surface and of the surgical gloves, as well as the chemical composition of the implant surface after contact with the surgical glove, was analyzed and the results are shown in Tables 3, 4 and 5.

Calcium, Zn, S, Si and Nb were found on the surface of the surgical gloves, with Ca and Zn being the predominant elements in all samples. Nb was detected in only two samples, but in significant amounts compared to the other elements present in the same sample. Sulfur was detected in four samples, while Si was found in only one sample and in small amounts compared to the other elements (Table 3).

Table 3: Chemical elements detected on the surface of the glove samples analyzed and their weight in %.

Gloves	Weight of the chemical elements (%)
Sample 1	Ca (67.43%); Zn(32.57)
Sample 2	Ca (49.37%); Zn (22.95%); S (27.68%)
Sample 3	Ca (35.04%); Zn (14.31%); S (44.36%); Si (6.29%)
Sample 4	Ca (41.98%); Zn (10.35%); S (9.94%); Nb (37.56%)
Sample 5	Ca (25.44%); Zn (14.73%); S (14.89%); Nb (44.93%)

Ti was detected on the surface of all implants and was the only element present in two implants (except for the 10th thread) as described by the manufacturer. Ca was found in the other samples, but in small amounts, and Ti was prevalent on the implant surface (Table 4).

Implants	4th thread	7th thread	10th thread
Implant 1	Ti (100%)	Ti (100%)	Ti (98.30%); Ca (1.70%)
Implant 2	Ti (100%)	Ti (100%)	Ti (99.22%); Ca (0.78%)
Implant 3	Ti (100%)	Ti (97.82%); Ca (2.18%)	Ti (98.57%); Ca (1.43%)
Implant 4	Ti (100%)	Ti (97.50%); Ca (2.50%)	Ti (98.16%); Ca (1.84%)
Implant 5	Ti (100%)	Ti (99.05%); Ca (0.95%)	Ti (99.11%); Ca (0.89%)

Table 4: Chemical elements detected on the implant surface and their weight in %.

Analysis of the implants after contact with the surgical glove identified Ti and Ca as the main elements, as well as S, Zn and Si. The areas corresponding to the 4th, 7th and 10th thread were analyzed and Ti was the only element detected on the 4th thread. The greatest variation of chemical elements was observed for the 10th thread (Table 5).

Implants + gloves	4th thread	7th thread	10th thread
Sample 1	Ti (100%)	Ti (98.98%); Ca (1.02%)	Ti (98.80%); Ca (1.20%)
Sample 2	Ti (100%)	Ti (98.42%); Ca (0.91%); S (0.68%)	Ti (96.57%); Ca (0.93%); Zn (1.41%); S (1.08%)
Sample 3	Ti (100%)	Ti (99.62%); Ca (0.38%)	Ti (93.48%); Ca (3.92%); Zn (1.51 %); S (1.09%)
Sample 4	Ti (100%)	Ti (94.82%); Ca (2.32%); S (1.37%); Zn (1.48%)	Ti (96.07%); Ca (1.98%); S (1.40%); Si (0.54%)
Sample 5	Ti (100%)	Ti (97.88%); Ca (1.56%); S (0.55%)	Ti (94.06%); Ca (1.97%); Zn (2.03%); S (1.94%)

Table 5: Chemical elements detected on the implant surface after contact with the surgical glove and their weight in %.

4 DISCUSSION

Studies analyzing the surface properties of dental implants by SEM and EDS, in addition to other techniques, have been reported in the literature[18,19]. These techniques permit to examine the chemical composition, the presence of impurities introduced during fabrication or handling, the thickness of the oxide layer, and the surface roughness of dental implants[6,7,9,12].

In the present study, EDS was used as an analysis method whose purpose is to identify the surface chemical composition of implants. This analysis method is similar to those employed in other studies[18,20] for the identification of chemical elements present on the surface of samples. According to Pedrosa[21], surface analysis by this method permits to identify any chemical element with an atom number higher than 2. Furthermore, this technique permits analysis at a depth of 1-2 µm and to vary the area analyzed in the samples by up to 1 µm. However, according to some authors, different techniques are necessary to obtain more accurate results since each method provides results of different magnitudes[18,20].

According to the manufacturer, cobalt 60 radiation was used for sterilization of the surgical gloves. Calcium was the predominant chemical element in the samples and Ca and Zn were detected in all samples. Sulfur was identified by EDS in four samples, Si in only one sample, and Nb in two samples. We found no studies in the literature on the chemical composition of surgical gloves for comparison of the data obtained here.

Analysis of the five commercially pure titanium implants before contact with the surgical gloves showed a predominance of Ti; two of these implants exhibited only Ti on their surface, except for the 10th thread, and the other three implants also exhibited Ca, without major variations in the area analyzed. Similar results have been reported by Henry et al.[16]. According to these authors, the presence of Ca in the three samples may be the result of etching used for opaque coating of the implant surface. In contrast, Vieira et al.[22] suggested the presence of calcium to be a consequence of the cleaning process with detergents, or its precipitation during the sterilization process. The implants used in the present study were sterilized by gamma radiation which is not related to contamination with calcium since it is a radiation method.

Contaminants can also be adsorbed by direct contact with non-titanium materials such as the powder on gloves used by dentists which forms a film on the implants[23,24]. Dental implant surgery procedures are performed using surgical gloves because they are made of more resistant and sterile material. Latex is the gold standard used for the fabrication of these gloves and nanoparticles are introduced into the natural rubber to improve its properties[25].

The surface of dental implants can be contaminated with organic and inorganic substances during their fabrication or manipulation during surgery. Inorganic substances contribute to the failure of osseointegration by interfering with the biocompatibility of titanium which is mediated by the titanium dioxide layer, modifying the surface energy of the implant and causing variable degrees of metal destruction[1,26-28]. Inorganic elements were detected in the implants after contact with the surgical glove, suggesting a possible interference with osseointegration since these elements can catalyze reactions with the oxygen present in the titanium dioxide layer and consequently alter the concentrations of this oxide on the surface of dental implants.

In the present study, carbon and oxygen were found in all samples submitted to EDS analysis. However, these elements were excluded since they may have originated from air at the time when the implant package was opened. This finding agrees with other studies investigating the chemical surface composition of dental implants[3,29-31]. According to ASTM F67 specifications, carbon can be present on the implant surface and not interfere with bone healing as along as its concentrations is less than 0.10%[18]. Carbon is an organic contaminant that can be air-borne or arise from failures in the cleaning, sterilization and packaging processes[19,29]. The carbon found in the samples studied here may have originated from carbon compounds such as carbon dioxide (CO_2) present in the ambient air. According to Wälivaara et al.[38], the presence of carbon as a surface contaminant of titanium implants did not influence the adsorption of proteins such as albumin and fibrinogens. However, Sardinha[3] suggested that possible carbon bonding to oxygen may generate compounds that compete with oxygen for binding to titanium.

Analysis of the implants after contact with the surgical glove revealed the presence of chemical elements not detected on the implants before contact. These elements were S, Zn, Si and Ca, which are not part of the surface composition of implants and are therefore considered to be contaminants derived from the surgical glove. Henry et al.[34] detected some of these elements, such as Ca and Si, on the surface of commercially pure titanium implants as a result of etching used for opaque coating of the implant. According to these authors, the presence of these contaminants may render the implant more susceptible to corrosion, compromising osseointegration. Vieira et al.[22] also identified the presence of S and Si, and Wälivaara et al.[30] detected contaminants such as Zn, S, Si and Ca on the surface of their samples, in addition to other elements.

These inorganic contaminants present on dental implants can detach from the superficial layer and increase the intensity and duration of the inflammatory response, modifying bone healing and causing decomposition of the implant[7,28]. Elements such as C, Ca, S and Si have been identified by EDS in the case of a failed implant in the anterior region of the mandible, exhibiting mobility, pain and exudation[9]. However, Piattelli et al. [14], evaluating osseointegration based on the percentage of bone-implant contact, observed no significant difference between implants with inorganic contaminants (test group) and decontaminated implants (control). There was also no significant difference in the number of multinucleated cells or osteoclasts in contact with the implant surface.

Si was detected in the present study on gloves and on the implants after contact with the glove. Klauber et al.[17] also identified Si on the implant surface as a contaminant of the upper portion of the thin natural oxide layer. According to these authors, after installation of the implant, the formation of silicates can be an initial trigger of implant failure, compromising the bone-implant interface at the molecular level.

In the study of Shibli et al.[28] in which the surface composition and presence of contaminants on failed titanium implants were analyzed by SEM and EDS, the surface of the samples consisted of a layer of TiO_2 and variable amounts of contaminants, including C, Zn, O, N, Na, Ca, Al, P, and S. The degree of contamination of the titanium surface determines the mechanical stability of implants and the quality of osseointegration. According to these authors, the presence of Zn in solid or liquid form can cause titanium fragility. However, it remains unknown whether the Ca^{2+} and Si^{4+} found are able to dissolve titanium and thus cause corrosion on the implant surface. In the present study, the detection of Zn, S and Si after contact of the implant surface with the surgical glove suggests contamination of the implant, in agreement with Shibli et al.[28] and Aparicio and Olivé[20].

The fact that titanium implants are subjected to different processes of surface treatment, as well as the handling during the surgical procedure, makes the introduction of impurities or implant contamination inevitable. Several studies have therefore evaluated different types of surfaces and the presence of contaminants in order to obtain the ideal surface. Some authors argue that the proliferation and differentiation of osteoblasts differ as a function of the surface properties of implants[8,30].

Disinfection of implants prior to the surgical procedure is currently discussed as a possibility of detaching ion contaminants from the surface. If persisting, these contaminants can cause prolonged and aggressive periods of inflammation, a condition known as peri-implantitis[7].

We do not know if the amounts of contaminants observed in this study can lead to implant failure or have some clinical implication. The results of this study suggest that the adsorption of chemical elements from surgical gloves onto titanium implants is possible if the implant comes in contact with the glove during its installation. Further studies are needed to determine whether the chemical elements adsorbed to the implant surface interfere with osseointegration.

5 CONCLUSION

According to the method used and the results obtained, it can be concluded that Ti and Ca were the predominant chemical elements on the surface of titanium implants. The elements detected on the surface of surgical latex gloves were Ca, Zn, S, Si, and Nb. The chemical elements found on the surface of titanium implants after contact with the surgical glove were Ti, Ca, S, Zn and Si, suggesting contamination of the implants which could interfere with osseointegration.

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REFERENCES

1. Manera RS, Rodrigues AR, Matsumoto H, Gallego J. Avaliação da integridade superficial de implantes dentarios de titanio. In: anais do V Congresso Brasileiro de Engenharia de Fabricação, 2009; Belo Horizonte MG, 2009.

2. Bonachela WC. Avaliação de implante dental comprometido empregando microscopia eletrônica de varredura e espectrometria dispersiva de raio X. Innov. Implant. J.Biomater. Esthet. 2010 set./dez;5(3):75-8.

3. Brandão ML, Esposti TBD, Bisognin ED, Harari ND, Jr Vidigal GM, Conz MB. Superficie dos implantes osseointegrados x resposta biológica. Rev Implant News. 2010;7(1):95-101.
4. Diniz MG, Pinheiro MAS, Andrade Junior ACC, Fischer RG. Characterization of titanium surfaces for dental implants with inorganic contaminant. Braz. oral res. 2005;19(2):106-11.

5. Machnee CH, Wagner WC, Jaarda MJ, Lang BR. Identification of oxide layers of commercially pure titanium in response to cleaning procedures. Int. j. oral maxillofac. implants. Lombard. 1993;8(5):529-33.

6. Saberlich S, Klee D, Richter E-J,Höcker H, Spiekermann H. Cell culture tests for assessing the tolerance of soft tissue to variously modified titanium surfaces. Clin. oral Implants res.1999;10:379-93.

7. Jr Tavares CARF, Sendyk WR, Matos AB, Sansiviero A. Contaminação química superficial de implantes osseointegrados: estagio atual. Rev. Inst. Invest. Cienc. Salud. 2005;23(2):139-43.

8. Buser D, Broggini N, Wieland M, Schenk RK, Denzer AJ, Cochran DL, et al. Enhanced bone apposition to a chemically modified SLA titanium surface. J. dent. res. 2004;83(7):529-33.

9. Hallgren C, Reimers H, Chakarov D, Gold J, Wennerberg A. An in vivo study of bone response to implants topographically modified by laser micromachining. Biomaterials. 2003;24:701-10.

10. Albrektsson T, Wennerberg A. Oral implant surfaces: Review focusing on topographic and chemical properties of different surfaces and in vivo responses to them. Int. j. prosthodont. 2004;17:536-64.

11. Keller JC, DraughnRA, Wightman JP, Dougherty WJ, Meletiou SD. Characterization of sterilized CP titanium implant surfaces. Int. j. oral maxillofac. implants.1990;5:360-67.

12. Kasemo B. Biocompatibility of titanium implants: surface science aspects. J. prosthet. dent. 1993;49:832-7.

13. Piattelli A, Degidi M, Paolantonio M, Mangano C, Scarano A. Residual aluminum oxide on the surface of titanium implants has no effect on osseointegration. Biomaterials. 2003;24:4081- 89.

14. Piattelli A, Scarano A, Piattelli M, Podda G. Implant periapical lesions: Clinical, histologic, and histochemical aspects. A case report. Int. J. Periodont. Rest. Dent. 1998;18(2):181-7.

15. Mouhyi J, Sennerby L, Pireaux JJ, Dourov N, Nammour S, Van Reck J. An XPS and SEM evaluation of six chemical and physical techniques for cleaning of contaminated titanium implants. Clin. Oral Implants res. 1998;9:185-94.

16. Placko, HE; Mishra, S; Weimer, JJ; Lucas, LC. Surface characterization of titanium-based implant materials. Int. j. oral maxilofac. implants. 2000;15(3):355-63.

17. Davies JED. Mechanisms of endosseous integration. Int J Prosthodont 1998;11:391–401.

18. Gehrke AS, Santos M, Dedavid BA, Martins R, Carvalho NTA, Carbonari MJ. Potencial de osseointegração versus características da superfície microtexturiza da Vellox. Implant News. 2011;8(3b-PBA):129-37.

19. Vieira, RM; Namen, FM; Galan Junior, J. **Characterization of the surfaces of dental implants commercial in scanning electron microscopy / energy dispersive spectroscopy.**RGO. 2013 jan./mar.;61(1):27-39.

20. Henry PJ. Comparative surface analysis of two osseointegrated implant systems. Int. j. oral maxillofac. implants. Lombard. 1987;2(1):23-7.

21. Mendonça G, Mendonça DBS, Simões LGP, Araújo AL, Golin AL, Duarte WR, Cooper LF, Aragão FJL. Efeito de Superfícies de Implantes Nano-Estruturadas na Expressão de Genes de Osteoblastos e no Contato Osso-Implante In Vivo. Rev. bras. odontol.2010;19(50).

22. Pedrosa AR. Analise quimica e topografica em superficies tratadas de implantes osseointegráveis de titânio. Belo Horizonte; 2010. [Dissertação de Mestrado - Centro de Desenvolvimentoda Tecnologia Nuclear].

23. Fadanelli AB, Stemmer AC, Beltrão GC. Falha prematura em implantes orais. Rev. odontociênc. 2005 abr./jun.;20(48).

24. Collantes HDC. Fabricação de luvas cirúrgicas com látex de borracha natural vulcanizado com raios gama. São Paulo; 1995. [Dissertação de Mestrado - Universidade de São Paulo].

25. Shibli JA, Marcantonio E, d'Avila S, Guastaldi AC, Marcantonio E Analysis of failed commercially pure titanium dental implants: a scanning electron microscopy and energy-dispersive spectrometer x-ray study. J. periodontol. 2005;76(7):1092-9.

26. Brentel AS, Vasconcellos LMR, Oliveira MV, Graça MLA, Vasconcellos LGO, Cairo CAA, Carvalho YR. Histomorphometric analysis of pure titanium Implants with porous surface versus rough surface. J. appl. oralsci.2006;14(3):213-8.

27. Klauber C, Lenz LJ, Henry PJ. Oxide thickness and surface contamination of six endosseous dental implants determined by electron spectroscopy for chemical analysis: a preliminary report. Quintessenz j. 1990 Mar;5:264-271.

28. Mustafa K, Wennerberg A, Wroblewski J, Hultenby K, Silva Lopez B, Arvidson K. Determining optimal surface roughness of TiO2 blasted titanium implant material for attachment, proliferation and differentiation of cells derived from human mandibular alveolar bone. Clin. oralimplants res.2001;12:515–25.

29. Sardinha, SCS. Análise Química e Topográfica da Superfície de Implantes de Titânio Comercialmente Puro Através de Espectroscopia de Fotoelétrons Excitada por Raios – X (XPS) e Microscopia Eletrônica de Varredura (MEV). Piracicaba; 2003. [Dissertação de Doutorado - Universidade Estadual de Campinas].

30. Walivaara B, Aronsson BO, Rodahl M, Lausmaa J, Tengvall P. Titanium with different oxides: in V&U studies of protein adsorption and contact activation. Biomaterials. 1994;15(10).