Effect of Air-Powder System on Titanium Surface on Fibroblast Adhesion and Morphology

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A direct contact between the dental implant surface and surrounding bone is preferred for the long-term success of dental implants. Nevertheless, in spite of a satisfactory osseointegration, this clinical success of dental implants can be guaranteed only if the integrity of peri-implant mucosa is maintained by an attachment by hemidesmosomal connection of soft-tissue to the transmucosal implant surfaces.¹ ²

On the other hand, it must be emphasized that clinical studies of the interface between gingival tissues and dental implants in humans are hampered by many difficulties, including ethical considerations. Many uncontrollable factors in the oral environment, as well as technical problems on sample preparations, impairexperimental studies on soft-tissue behavior. In vitro experiments appear to circumvent most of these difficulties and thus can provide useful information on this subject.³

Common clinical procedures such as professional maintenance performed with stainless steel and plastic curettes or abrasive pumice or air-powder abrasive system could to lead to alterations on the surface of titanium abutment, impairing, for example, adhesion of fibroblasts to this sur-

Face. Comparative experiments on the attachment and growth of human gingival fibroblasts and epithelial cells on titanium with different surface textures were carried out.⁴ ⁵ These studies showed that epithelial cells present more extensive migration on rough surfaces. However, gingival fibroblasts showed a more marked and oriented development on porous surfaces, which was also observed by other authors.⁶ ⁷

Even though rough surfaces could enhance fibroblast responses, they can also be considered rather disadvantageous because of the possibility of promoting growth and organization of bacterial biofilms, thus facilitating perimplant tissue infections such as mucositis and periimplantitis.⁸

The purpose of this in vitro study was to evaluate the effect of using an air-powder abrasive system on titanium abutments on adhesion and morphology of fibroblasts.

**MATERIALS AND METHODS**

**Cell Lineage**

A continuous cell lineage of fibroblastic morphology (McCoy) from the Adolfo Lutz Institute, Sao Paulo, Brazil, was used. These cells were cultured in 25 cm² flasks with minimum essential media supplemented with 7.5% of fetal bovine serum and 40 μg/mL of gentamicin. The cells were maintained in an incubator at 37°C and 98% humidity atmosphere. Cell suspensions were prepared at the exponential growth phase always from the same passage throughout the experiment. The same batch of supplemented culture media was used.
throughout the experiment to minimize possible variations on cell growth.

Treatment of Specimens

Twenty-six new commercially pure, titanium healing abutment surfaces (4 mm × 8 mm) (ImplaMed; Attleboro, MA) were used in this study. These abutments were removed from the original packing, cleaned on an ultrasonic device for 10 minutes, and then sterilized by steam heat (autoclave). Care was taken not to contact the abutment cylinder surface with any foreign object other than the test instruments and materials.

Two titanium abutments were designed as negative control (no treatment with air-abrasive system and no cells) and two positive controls (air-powder system treatment and no cells). The remaining 22 specimens were assigned to two experimental groups: control group (no air-powder treatment) and test group (air-powder system) (Prophy-Ceramic II; Dabi Atlante, Ribeirão Preto, SP, Brazil) for 30 seconds on a 45° incidence. The air-powder system was performed with sodium bicarbonate. A single operator used the Prophy-Jet device loaded with sodium bicarbonate on all titanium abutments of the test group. Immediately after treatment, the specimens were coded and individually placed in 24-well plates. To each well, 2 mL of supplemented cell culture medium was removed by aspirating with an ultrasonic device for 10 minutes, and then sterilized by steam heat (autoclave). Care was taken not to contact the abutment cylinder surface with any foreign object other than the test instruments and materials.

The disruption of this seal by inflammatory periimplant disease can permit increased accessibility of biofilm-derived substances into the connective tissues. Several studies have tested various measures for cleaning smooth implant surfaces.8,11-13 Surface cleaning with an air-powder abrasive system has been suggested.14-16

The results of this in vitro study showed that proliferation and migration of fibroblasts are possible on titanium surfaces, in accordance with Gould et al.17, whose in vitro results indicated a hemidesmosomal connection between epithelial cells and titanium surfaces. It was also shown that even the orientation of fibroblasts could be influenced by titanium surface characteristics.4,18

Results

The distribution of morphology scores (Fig. 1) according to experimental group Mann-Whitney tests did not indicate significant differences between groups (P > 0.05), suggesting that cell morphology was not affected by treating the healing abutments with the air-abrasive system.

Fig. 1. Frequency distribution of the percentage of scores for cell morphology according to the experimental groups.

Discussion

Acquisition and maintenance of an effective attachment around the cervical portion of a dental implant is essential to establish a favorable prognosis. The periimplant seal provides a biologic barrier between the oral environment and periimplant bone tissue. The disruption of this seal by inflammatory periimplant disease can permit increased accessibility of biofilm-derived substances into the connective tissues. Several studies have tested various measures for cleaning smooth implant surfaces.8,11-13 Surface cleaning with an air-powder abrasive system has been suggested.14-16

The results of this in vitro study showed that proliferation and migration of fibroblasts are possible on titanium surfaces, in accordance with Gould et al.17, whose in vitro results indicated a hemidesmosomal connection between epithelial cells and titanium surfaces. It was also shown that even the orientation of fibroblasts could be influenced by titanium surface characteristics.4,18
Using an air-powder abrasive system with sodium bicarbonate for 30 seconds on a 45° device on commercially pure titanium did not alter the morphology of fibroblasts. In agreement with the literature,19,20 morphology of these cells was predominantly elongated or flattened (bipolar or multipolar), which was considered a sign of adhesion to the substrate. This indicates that surface roughness and the presence of particles of bicarbonate were not able to alter this phenotypic expression of fibroblasts. However, what consequences this type of surface instrumentation can have on the attachment of periimplant soft tissues in the long-term remains unclear.

On the other hand, on titanium surfaces treated with an air-powder abrasive system, a significant decrease in the number of fibroblasts was observed in comparison with nontreated control titanium surfaces. Another study has documented similar results after surface treatment with stainless steel curettes.21 There are some in vitro data suggesting that smooth surfaces are superior in promoting fibroblast proliferation as well as the number of cells attaching to the surface.22 These results can be attributed to the release of toxic ions from the titanium alloy23 or to the presence of powder particles on instrumented surfaces, which can disturb cellular adhesion.

Results obtained in these studies show that the nature and surface geometry of the implant surfaces may influence gingival fibroblasts attachment in vitro, in agreement with our data. However, one has to bear in mind the limitation of the methods used in this study when considering these results. In this study, a continuous lineage of fibroblastic cells was used. The advantages of this type of culture are the rapid proliferation of cells (reducing the probability of contamination), the infinite life-span of cells, allowing for many repetitions of experiments, in addition to the fact these cells are also easier to grow and maintain. In consideration of the purpose of this study, which was to perform an initial evaluation of adhesion and proliferation of fibroblasts on titanium surfaces after treatment with an air-powder system, cells from continuous lineages are considered adequate.24

Appropriate care was taken to minimize possible sources of variation on the assay. This care included preparation of cell suspensions at the exponential growth phase as well as always obtaining these cells from the same 75 cm² cell culture flask to avoid possible differences on cell behavior caused by variations in culture conditions. In this sense, the same supplemented culture medium batch was used throughout the study.

**Conclusion**

Treatment of titanium abutments with an air-powder device using sodium bicarbonate significantly reduced the number of fibroblasts attached to these surfaces. On the other hand, no morphologic alterations were observed on the cells present on treated titanium surfaces, indicating that the adhesion of fibroblasts was not significantly affected. Clinically, these findings indicate that using an air-powder abrasive system on titanium abutments to remove bacterial biofilm during treatment of periimplant mucositis or maintenance care does not reduce the biocompatibility of these surfaces.

**Disclosure**

The authors claim to have no financial interest in any company or any of the products mentioned in this article.

**References**


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ABSTRACT: Propósito: Evaluar el número y morfología de los fibroblastos que han crecido en los postes de curación de titanio pulidos a máquina tratados con un sistema de polvo de aire. Materiales y Métodos: Veintiséis postes fueron asignados a 2 grupos experimentales: control (sin tratamiento) y tratados - expuestos a Prophy-Jet durante 30 segundos. Los especímenes fueron incubados durante 24 horas con células fibroblásticas en placas con múltiples pocillos, seguidos por procesamiento de rutina en laboratorios para el análisis SEM. Los especímenes fueron fotografiados en 350X y el número de células se contaron en una área de aproximadamente 200 um². Resultados: No se encontraron diferencias significativas en morfología entre los grupos (p > 0.05), sin embargo, el grupo de control presentó una cantidad más importante de células (71,44 ± 31,93, mediana ± d.s.) en comparación con el grupo tratado (35,31 ± 28,14), según lo indica una prueba T sin pares (p = 0,001). Conclusión: El uso de un sistema de profilaxis abrasivo de aire sobre la superficie de los postes de curación de titanio redujo la proliferación de células pero no tuvo influencia en la morfología de las células.

PALABRAS CLAVES: Implantes dentales, cultivo de células, titanio, fibroblastos, mantenimiento, microscopía por escapeado de electrones.
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**SINOPSE:** OBJETIVO: avaliação do número e morfologia dos fibroblastos desenvolvidos em pivós de cicatrização de titânio usinado, tratados com um sistema de jato abrasivo. MATERIAIS E MÉTODOS: vinte e seis pivós foram distribuídos em dois grupos experimentais: controle (sem tratamento) e com tratamento – expostos ao ProphyJet por 30 segundos. As espécies foram incubadas por 24 horas com células fibrolásticas em placas de orifícios múltiplos (multilwell), seguidas de processamento laboratorial rotineiro para a análise SEM. Os espécies foram fotografados em 350X e foi realizada a contagem do número de células em uma área de aproximadamente 200 μm². RESULTADOS: não foram encontradas diferenças significativas na morfologia entre os grupos (p>0,05) entretanto, o grupo de controle apresentou uma quantidade de células significativamente maior (71,44 ± 31,93, média ± s.d.) em comparação com o grupo com tratamento (35,31 ± 28,14), conforme indicado por um teste t sem paridade (p=0,001). CONCLUSÃO: a utilização do sistema profilático de jato de areia na superfície dos pivós de cicatrização de titânio reduziram a proliferação das células mas não influenciaram a morfologia da célula.

**PALAVRAS-CHAVES:** implantes odontológicos, cultura celular, titânio, fibroblasto, manutenção, microscopia eletrônica de varredura

**AIR-PowDER SYSTEM AND FIBROBLAST ADHESION**