

HISTOLOGICAL AND MICROSCOPIC COMPARISON OF THE RATE OF OSSEOINTEGRATION OF DRY AND WETTED TITANIUM IMPLANTS IN AN EXPERIMENT.

PhD. Kaplun D.V.

Abstract:

The surface of a dental implant plays a decisive role in determining its performance. Numerous methods are used or being developed to modify implant surfaces. The wettability of biomaterial surfaces determines the rate of the biological cascade of events at the biomaterial–bone interface. There is growing interest in understanding the mechanisms of implant surface wettability and its role in the biological response at the implant–bone or implant–soft tissue interface. Focusing on titanium dental implants, this study reviews current knowledge on the wettability of biomaterial surfaces, encompassing both fundamental and applied aspects, with a particular emphasis on clinical data.

Keywords:

Osseointegration, morphological aspects of the implant–bone interface, implant, moistened implant, wettability, titanium dental implant

Objective:

To determine the qualitative and kinetic parameters of osseointegration by comparing titanium samples treated with the SLA technique in dry and moistened forms. To investigate the role of oxidized titanium surfaces, formed through interaction with oxygen, in biointegration processes. To examine the microscopic and histological features of the biological interaction between bone structures and the non-oxidized surface of dental implants.

Tasks:

To determine the role of the air-oxidized titanium surface of the dental implant in the rate of biological processes.

To study the rate of osseointegration of dry and moistened dental implants.

To determine the histological and microscopic differences in the proliferation and maturation of bone structures on the surface of the studied samples.

Introduction

Implants are widely used in dental practice. The process leading to satisfactory osseointegration of the implant with the surrounding tissues is complex. This process begins with the initiation of the coagulation cascade, platelet aggregation, and the formation of a blood clot around the implant, which results in the creation of a matrix or temporary fibrin network surrounding the implant. This temporary network performs two important functions: it provides the initial stability of the implant and ensures the gradual release of platelet growth factors and cellular markers. Among other processes, cellular markers serve this purpose by stimulating cell migration to the wound area, their adhesion, differentiation, and

proliferation, as well as the secretion of extracellular matrix with its subsequent mineralization, which culminates in the formation of a characteristic bone matrix around the implant. The surface of the implant plays a decisive role in determining its performance. Many methods are used or being developed for implant surface modification. Recently, implants with a wetted surface have gained popularity; however, the specific clinical advantages of such implants have not been sufficiently investigated. The wettability of biomaterial surfaces determines the rate of the biological cascade of events at the biomaterial–bone interface. The design of modern implant surfaces is focused mainly on

specific micro- and nanotopographic characteristics and is still far from predicting the overall response of the organism to wettability. There is a growing interest in understanding the mechanisms of implant surface wettability and the role of wettability in the biological response at the implant–bone or implant–soft tissue interface. Fundamental knowledge related to the hydrophilicity or wettability of titanium and titanium alloy

surfaces, as well as various aspects associated with wettability modes, can improve our understanding of the role of hydrophilic rough implant surfaces in biological outcomes. Focusing on titanium dental implants, this study reviews current knowledge on the wettability of biomaterial surfaces, covering both fundamental and applied aspects, with particular emphasis on clinical data.

Materials and Methods

For the experiment, 16 fragments of dental implants made of Grade 4 titanium (Ti-6Al-4V) were used. The surface of the samples was represented by micro- and nanotopography obtained through sandblasting with aluminum oxide (resulting in pores of 20–40 microns) followed by a double acid-etching process at different temperatures (leading to the formation of micropores ranging from 1 to 5 microns) at the manufacturer's facility. Eight samples, after surface treatment and prior to sterilization, were dried, while the other 8 samples were immersed in physiological saline solution (0.9% NaCl) and remained stored in it permanently.

For the experiment, 8 sexually mature laboratory rabbits of the California breed were selected. Under aseptic conditions, each rabbit received implantation of 2 implant samples

(dry and moistened) into the tibial bone of both legs. The study was conducted in accordance with the required regulatory acts (the Helsinki Declaration of 2000 on humane treatment of animals and the "Rules for conducting work with experimental animals"). Telazol (100 mg diluted in 10 ml of 0.9% NaCl solution) was used as anesthesia, along with local anesthesia. The results of osseointegration were evaluated using morphological studies. Macrospecimens were examined after sectioning the bone into blocks on the 30th and 90th days following implantation and after appropriate fixation in 12% neutral formalin with four months of decalcification in a 10% solution of Trilon B. Sections were prepared and stained with hematoxylin and eosin, as well as with picrofuchsin according to the Van Gieson method.

The results of scanning electron microscopy were obtained using a Tescan Vega 3 SB device equipped with an Oxford Instruments X-Act EDS analysis system.

Results of the Study

It is known that titanium, as a metal, is capable of oxidizing both through interaction

with oxygen and in water. To compare the surface characteristics of the studied implants, electron microscopy of the samples was performed (Figs. 1, 2) prior to implantation.

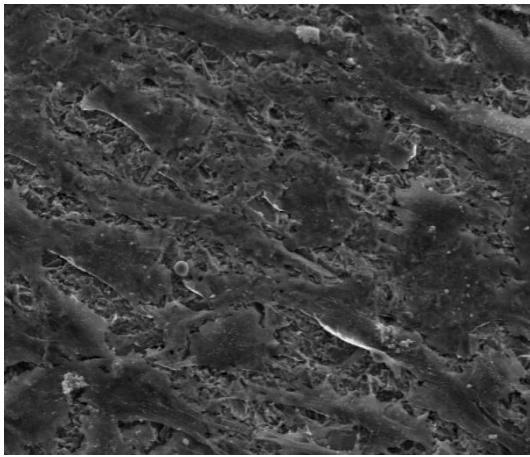


Fig. 1. Surface area of titanium samples from the first clinical group (SLA-treated, dry) before implantation.

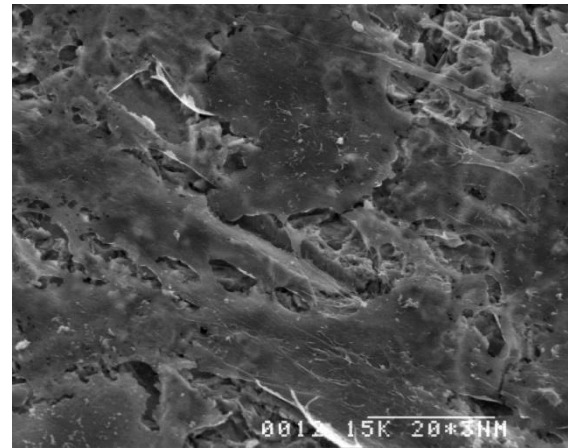


Fig. 2. Surface area of titanium samples from the second clinical group (SLA-treated, wetted) before implantation.

Microscopic examination at magnification up to 3 nm does not provide clear data on the advantages of either sample. The surface of both samples appears similar and shows no characteristic differences. Histological studies performed one month after implantation (Figs. 3, 4) demonstrate the formation of a fibrous capsule around the implant, which is combined with the formation of new bone trabeculae. The dense fibrous capsule is lined with a thin epithelial

layer. The connective tissue layer between the implant and the bone is of medium width, with a moderate number of cellular elements, mainly fibroblasts with well-developed fibrillar structures. In all samples with a moistened surface, the collagen fibers forming bundles with areas of fibrillation have significantly more contact points with the implant surface. A thin fibrous capsule separates the implant from the newly formed spongy bone.

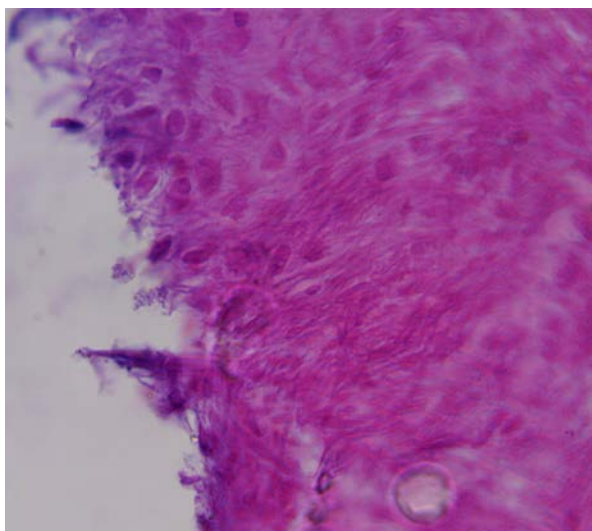


Fig. 3. Tibial bone of an experimental rabbit on the 30th day after implantation. Sample with a moistened surface. Stained with hematoxylin and eosin. Magnification ×250.

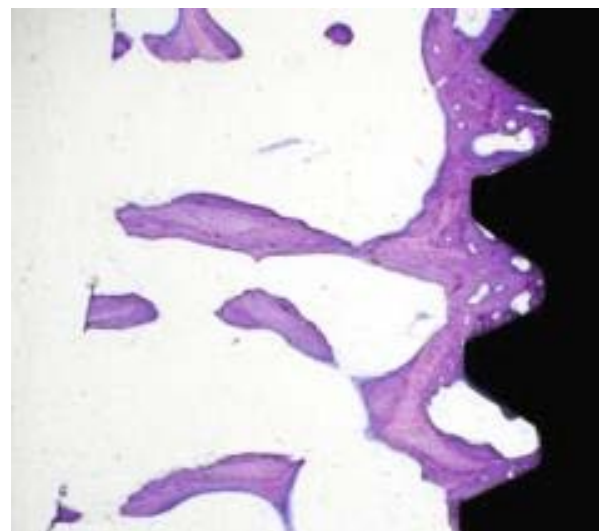


Fig. 4. Tibial bone of an experimental rabbit on the 30th day after implantation. Sample with a dry surface. Stained with hematoxylin and eosin. Magnification ×150.

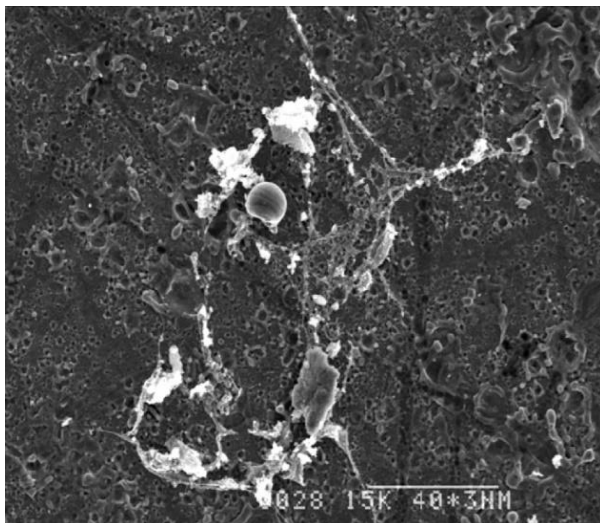


Fig. 5. Surface area of titanium samples from the first clinical group (SLA-treated, dry) four weeks after implantation.

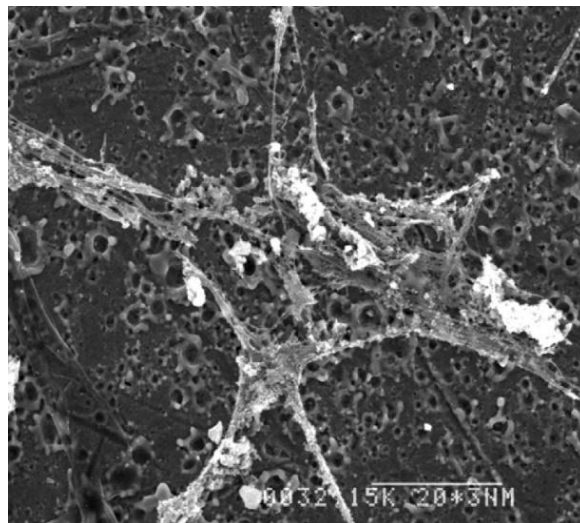


Fig. 6. Surface area of titanium samples from the second clinical group (SLA-treated, wetted) four weeks after implantation.

In the samples with a dry surface, electron microscopy at this stage (Fig. 5) shows individual osteogenic stromal precursor cells on the metal surface with ectoplasmic projections. At the same time, in the samples that were wetted, cellular structures “spread out” on the metal surface are also observed, with ectoplasmic projections of bone marrow osteogenic stromal precursor cells fixed within existing irregularities, but appearing somewhat more mature (Fig. 6), which indicates shorter adhesion times of blood clot cells to these samples. Three months after implant placement, an uneven inner line with serrations corresponding to the screw threads

of the implant is observed in the connective tissue capsule around the implant. There is no connective tissue layer between the implant and the bone. The capsule is closely attached to the mature compact bone tissue. A distinct “bonding” line is detected in the samples with a moistened surface. Thinning of the capsule is associated with the continuation of the osseointegration process and the accumulation of bone mass within the bone. The number of immature bone trabeculae is significantly reduced. Numerous bonding lines indicate the gradual layering of newly formed bone tissue. The bone tissue is mature and compact (Figs. 7, 8).

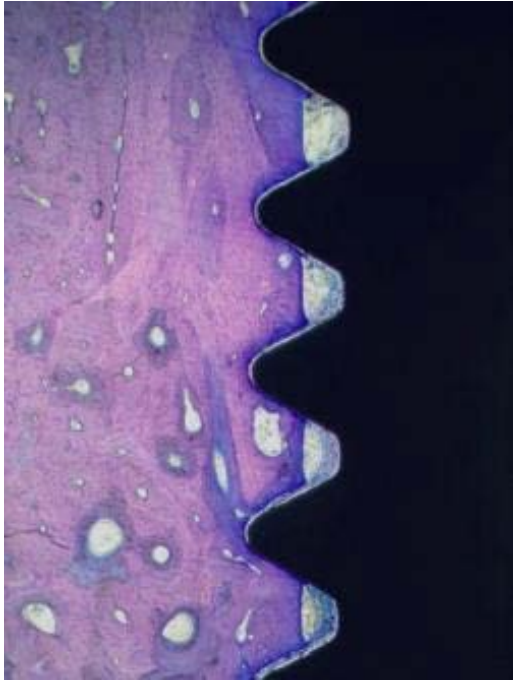


Fig. 7. Tibial bone of an experimental rabbit on the 90th day after implantation. Implant with a moistened surface. Stained with hematoxylin and eosin. Magnification $\times 250$.

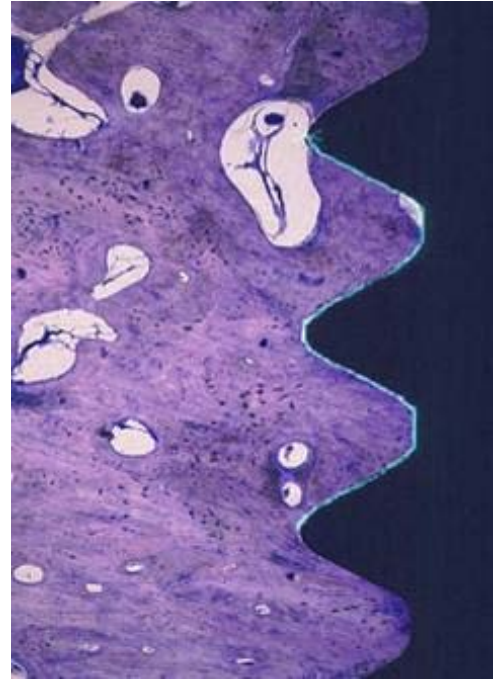


Fig. 8. Tibial bone of an experimental rabbit on the 90th day after implantation. Implant with a moistened surface. Stained with hematoxylin and eosin. Magnification $\times 250$.

Conclusions

1. It has been established that air oxidation of the titanium surface of a dental implant slows down the processes of stromal cell fixation on the titanium surface, which in turn delays the rate of the biological cascade of events at the biomaterial–bone interface.
2. It has been proven that the connective tissue layer between the implant and the bone differs in thickness at the same observation periods. The dry samples demonstrated a thicker layer of poorly mineralized bone tissue compared to the moistened samples, where the bone tissue was more mature.

Summary

The design of modern implant surfaces involves various processing methods that stimulate cell migration to the wound area, their adhesion, differentiation, and proliferation. Special attention should be given to the wettability of biomaterial surfaces, which determines the rate of the biological cascade of events at the biomaterial–bone interface. It is evident, and has been demonstrated by us, that titanium interacts with oxygen, and the resulting nano-film of titanium oxide on its surface somewhat slows down the process of osseointegration. When comparing identical implant fragments, osseointegration occurs significantly faster on the moistened surface compared to the dry one, as demonstrated microscopically and morphologically.

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