

Comparative Study

## OSTEOBLAST ADHESION ON ACID-ETCHED AND MACHINED SURFACES

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### ABSTRACT

Implants with surface chemical or biomechanical qualities that should encourage bone formation have been developed over the last 30 years. Few systematic investigations have been conducted on surface topography despite the widespread recognition that it is a significant factor influencing bone-implant contact. Four white New Zealand mature rabbits were used in the present investigation. Each rabbit received 2 implants, specially made with 2 surfaces on it (one sandblasted and acid etched and one machined) into each tibia. A total of 16 implants were inserted. All animals were killed 21 days after implant placement. Osteoblast in contact with the implant surface was statistically evaluated. The sandblasted and acid-etched surfaces showed a greater osteoblast contact than the machined, which was statistically significant. The sandblasted and acid-etched surface improves osteoblast adhesion on the implant surface.

**KEYWORDS:** *bone contact, machined implants, acid etched, osteoblast, bone healing, dental implants*

### INTRODUCTION

Dental prostheses have been successfully fixed using osseointegrated dental implants (1). Surface modification research aims to raise the percentage of bone-implant contact (BIC). Implants with surface chemical or biomechanical qualities that should encourage bone formation have been developed over the last 30 years. Few systematic investigations have been conducted on surface topography despite the widespread recognition that it is a significant factor influencing bone-implant contact (2). For this reason, a rough, textured, porous surface is incorporated into the design of many dental implants. This is because studies have demonstrated that roughened implant surfaces improve osseointegration. Such designs provide a larger surface area, which increases the possibility of cell attachment.

Tissue ingrowth into the implant is anticipated to stabilize the device mechanically by carefully gliding the rounded, fire-polished end of a glass rod across the tissue culture polystyrene. Fibroblasts avoided these uneven surfaces and gathered on the tissue culture dish's smooth areas. In contrast, macrophages preferred the rough surfaces to the smooth ones, a behavior described as "rugophilia" (3, 4). A comparable pattern of behavior has been reported for implants *in vivo*; it has been reported that macrophages preferred abraded Teflon implants(5). Interesting questions are raised by the

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roughened surfaces' propensity to draw monocytic series cells. For instance, since it is known that cells coming from the monocyte lineage generate osteoclasts, it is advantageous for implants in contact with bone to have surfaces that attract cells of the monocytic series (6).

Cell function and structure can be directly impacted by the surfaces on which cells can cling. In smooth substrata, cells produced on grooved substrata are more spherical (7, 8). Cell shape influences several cellular features, such as growth (9), secretion of proteinases (10), and gene expression (7). An implant's surface texture may selectively target particular cell populations and modify their activities. Detailed research is still needed to determine how implant surfaces impact the cells they come into touch with. This study used a split implant design to compare the osteoblast in contact with machined and those that were sandblasted acid etched.

## MATERIALS AND METHODS

Threaded machined and acid-etched pure titanium screw-shaped implants have been used in this study. The fixtures were placed into the tibia of 4 white New Zealand mature male rabbits according to a previously described technique (11). Each rabbit received 2 implants 4x8 mm, specially made with 2 surfaces on it (one sandblasted and acid etched and one machined) (Isomed System, Due Carrare, Padova, Italy) for each tibia. A total of 16 implants were inserted. The animals were anesthetized with intramuscular injection of fluanisone (0.7 mg/kg) and diazepam (1.5 mg/kg), and local anesthesia was given using 1 ml of 2% lidocaine/adrenalin solution. A skin incision with a periosteal flap was used to expose the tibial plate.

The preparation of the surgical sites was done with a series of burs under copious saline irrigation. Implant placement was performed according to a previously described protocol (11). The periosteum and fascia were sutured with absorbable sutures, whereas the skin was sutured with non-resorbable sutures. No postoperative complications or deaths occurred. The animals were killed 21 days after implant placement. An overdose of anesthesia was given, an incision was made on the tibia, and a block section was taken. All 16 implants were retrieved. The specimens were immediately stored in 10% buffered formalin and processed to obtain thin ground sections with a cutting-grinding machine. The specimens were dehydrated in an ascending series of alcohol rinses and embedded in a glycolmethacrylate resin. After polymerization, the specimens were sectioned longitudinally along the central axis of the implant with a high-precision diamond disc at about 150 microns and ground down to about 70 microns. One slide was obtained for each implant and stained with basic fuchsin and toluidine blue. Osteoblast in contact with the implant was done under a light microscope using a dedicated computer.

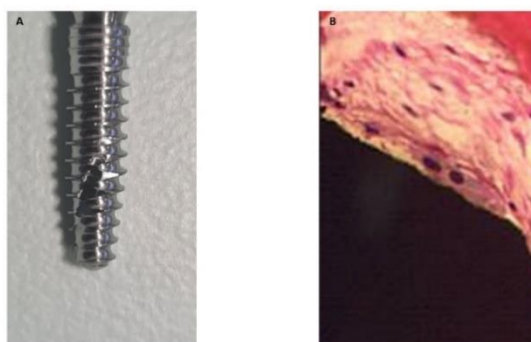
### Data analysis

The differences in the number of osteoblasts in contact between test (sandblasted and acid-etched) and control (machined) implants were evaluated. The RMANOVA analyzed the differences between the surface and the treatment groups, and the significance was assessed using the multi-comparison Tukey Test at  $p \leq 0.05$ . Three mm areas were examined for each sample.

## RESULTS

### Machined surface

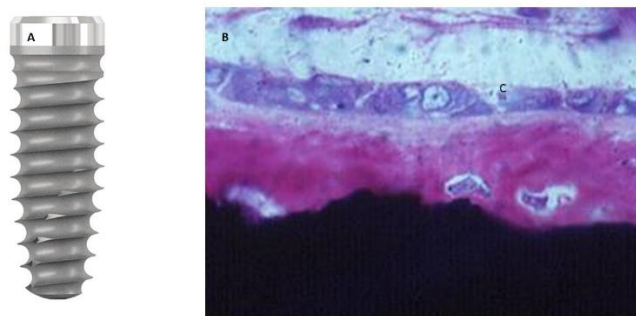
The number of osteoblasts near the implant surface was  $15 \pm 3$  (Fig. 1).



**Fig. 1. A):** Implant with machined surface; **B):** Four osteoblasts are recorded near the implant surface.

### *Sandblasted and acid-etched surface*

The number of osteoblasts in contact with the implant surface was  $22 \pm 4$  (Fig. 2).



**Fig. 2.** A): Implant with sandblasted and acid-etched surface; B): Nine osteoblasts are observed in contact with the implant surface.

### *Statistical evaluation*

The difference in mean values among the treatment groups was evaluated using RMANOVA. The test result shows a statistically significant difference ( $p = 0.001$ ).

## **DISCUSSION**

This research shows greater osteoblast adhesion on the sandblasted and acid-etched implant surface than on the machined surface. For patients who are totally or partially edentulous, using osseointegrated dental implants implanted in the jaws with direct bone-implant contact has evolved over the past 30 years into a dependable and popular treatment option (1). Several factors that affect the apposition of bone on implant surfaces have been studied in recent years to improve the bone anchoring of dental implants. The surface is the one that significantly influences the implant's integration (12). A favorable association has been demonstrated between the roughness values of five evaluated titanium surfaces and the percentage of bone-implant contact (13).

The best outcomes were obtained with 52% and 58% bone contact on the sandblasted and acid-etched surfaces. In tissue culture, the surfaces exhibit more alkaline phosphatase activity in osteoblast-like cells than smooth surfaces (14, 15). These data imply that the bone cells in touch with an etched surface may be more differentiated since alkaline phosphatase activity is a marker of bone cell maturation.

The results of Kaluđerović et al. (16) indicating that titanium disks with a microporous TPS surface demonstrated significantly faster bone-implant contact compared to those with smooth titanium surfaces could also be explained by these findings, which demonstrated a significant advantage for the etched surface during the early healing period. Torque removal forces have been employed biomechanically to disrupt the anchoring of osseointegration. Klokkevold et al. (17) examined rabbit femur resistance to removal forces. They employed machined and acid-etched surfaces. Following a two-month healing period, it was discovered that four times more force was required to remove the acid-etched implants than the machined implants. Based on the reverse torque rotation, they concluded that chemical etching of the titanium implant surface improves osseointegration.

Another author examined the removal torque values of titanium implants in the small pigs' maxillae. They contrasted implants that were machined, acid-etched, sandblasted, and sandblasted and acid-etched (SLA). Removal torque values for SLA implants were 75% higher than those of acid-etched implants following a 12-week healing period. Cordioli et al. (18) also performed a histomorphometric examination in the rabbit's tibia and determined the removal torque value. Four surface types were compared: machined, sprayed, acid-etched, and grit-blasted.

Histomorphometric analysis and removal torque values showed a considerable increase for the acid-etched implants after a 5-week healing period compared to the machined, blasted, and sprayed surfaces (4). The findings showed that compared to machined surfaces, a microrough titanium surface produced by acid etching techniques produced a larger percentage of bone-implant contact. In eight weeks, the bunnies were put to death. Around the implants, cortical bone development was more developed than cancellous bone. After six weeks, three months, and six months, the removal torque for screw-shaped pure titanium implants placed in the femoral portion of the knee joint and the rabbit tibia (19) was compared to the tissue response to these implants, as measured by light microscopic morphometry on ground sections.

Whereas cortical bone developed around the tibial implants, most of the bone around the femoral intra-articular implants was cancellous. Over time, the torque required to remove the intra-articular implants rose, but not the torque required to remove the tibial implants. These results suggest that the amount of compact bone surrounding a titanium implant affects the resistance to unscrewing.

In a rabbit tibial metaphysis research, threaded hydroxyapatite-coated implants made of commercially pure (CP) titanium were used (20). The controls were uncoated titanium screw implants made by CP that were placed in the opposing leg. Histomorphometric analysis was performed on the semi-loaded implants 6 weeks and 1 year after placement. At six weeks of follow-up, there was increased direct bone contact with the hydroxyapatite-coated implants, though not significantly more. With the uncoated CP titanium controls, there was notably greater direct bone-to-implant contact one year after placement. Three distinct surface topographies (21) were explored for the surfaces, and screw-shaped implants were made with two blasted surfaces with varying surface roughness levels and one surface that was left as machined or turned. The bone reaction to the rotated implants and the response to the two blasted implant surfaces were compared after a year in rabbit bone. The two blasted surfaces showed statistically significant increases in removal torque and percentage of bone-to-metal contact, indicating firmer bone fixation.

Wennerberg examined the bone response to titanium screws that were sold commercially and had two distinct levels of surface roughness (22). After the surface roughness of the implants was assessed, they were implanted in rabbit tibiae and blasted with 25- and 250- $\mu\text{m}$  particles of aluminum oxide. Implants blasted with 25- $\mu\text{m}$  particles showed a considerably higher bone-to-metal contact after 4 weeks than implants blasted with 250- $\mu\text{m}$  particles. This study suggests a short-term disadvantage for bone tissue when surface roughness is greatly enhanced as opposed to moderately increased. A comparison was made (23) between the resistance to removal torque forces for two distinct surface textures of commercially pure titanium implants that were screw-shaped. After six weeks of implantation in rabbits, it was discovered that the removal torque for implants with rough surfaces was much higher than that of implants with smooth surfaces. Because thick sections are produced by the most commonly used histology techniques, it is challenging to collect quantitative and comprehensive information on the orientation and morphology of cells adhering to implants (24). Cell shape has not been ascertained using the laborious method of taking serial sections and creating a three-dimensional reconstruction. These kinds of studies are necessary because they would show how cells migrate and adhere to implant surfaces and how they should function once they are there.

## CONCLUSIONS

The results of our study showed that the sandblasted and acid-etched surfaces have greater osteoblast activity than the machined ones, which is statistically significant only during the later phases of healing. The sandblasted acid-etched surfaces promote, within the limits of this study, an increase in the number of osteoblasts in contact with the implant surface.

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