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New laser-treated implant surfaces: A histologic and histomorphometric pilot study in rabbits

Abstract

Purpose: The purpose of this study was to confirm the validity of laser treated implant surfaces, with regard to high superficial purity preservation and to extremely regular and uniform roughness surfaces.

Methods: In this *in vivo* study, seven different laser treated implant surfaces were analyzed. A diode-pumped solid state source laser, in a Q-Switch output mode, was used at various wavelengths, which were chosen to generate surface irregularities of varying diameter, depth and pitch. Twenty one implants were placed in 11 New Zealand rabbits. Eight weeks after surgery, implants were harvested for histometric analysis: total, threads and body bone-to-implant, and bone-to-implant contacts were measured. The morphologic analysis of the surface was carried out using a Scanning Electron Microscope.

Results: Average bone-implant contact values were approximately 50% for all tested surfaces. Both total and threads values, within the same processing pattern group, had a high variance. Bone-implant contact thread and body variances were different, so that is possible that laser beam angle is able to modify the superficial roughness and thus the histological response.

Conclusions: Implants provided with pores of 20 and 25 μm achieved more than satisfactory bone-implant contact partial peaks. Further statistically significant experiments are needed in order to study, in depth, these surfaces.

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Implant surface structure represents one of the many factors affecting bone response, and therefore the osseointegration process [1]. To make possible a complete implant osseointegration into the bone, a wide contact surface is needed. To that end, since the diameter and length of an implant is limited for anatomical reasons, it becomes necessary to resort to micromorphologic surface modifications [2]. Implant surface treatments have the disadvantage, however, of contaminating the titanium surfaces, thereby interfering with the osseointegration process [3]. Common methods of producing surface roughness; for example, titanium plasma and aluminium oxide blasted surface methods, do not maintain optimal surface purity. Aluminium oxide-blasted implants cause surface contamination with aluminium particles, even when treated with hydrogen fluoride, which should improve titanium purity and therefore osseointegration [2]. Similar conclusions were reported by Olefjord and Hansson [4], who observed that implant surface contamination increased dissociation of the superficial titanium oxide layer, the stability of which is essential for a correct osseointegration process. These contaminating particles could exert proinflammatory effects by increasing the dissolution of the external titanium oxide layer [2].

In addition to surface purity, the other important feature to consider is implant surface micromorphology. Surface micromorphology refers to surface microporosity, which is required both to increase surface contact between bone and screw and to enhance surface conductivity [5]. It is well known that a rough surface allows for better osseointegration than a machined one [6]. It has been postulated that rough surfaces promote osteoconduction by increasing the surface area for fibrin attachment and providing surface features with which fibrin could become entrapped [5]. Thus, a rough surface is able to attach to osteogenic cells and initiate bone formation directly on the implant surface. On the other hand, if the implant surface is not able to retain the fibrin network, the osteogenic cells will rarely attach to its surface, and bone will form outward, taking more time [5]. This is most likely the case with the machined titanium surfaces, which show difficulties in achieving osseointegration in poor-quality bone [7,8]. These two variables, purity and micromorphology, are closely linked, because changes in the superficial structure, by means of material addition, lead inevitably to alterations in purity. For this reason, surface laser treatment appears a promising alternative.

Laser Surface Engineering (LSE) is a material processing system that exploits high-density laser beam to melt, heat or otherwise modify the external layers of materials [8]. This procedure can achieve extremely pure surfaces, as demonstrated in many studies [2] in which superficial compositions of laser-

treated implants and plasma-spray implants are compared. Laser-treated surfaces offer another important characteristic: whereas traditional techniques lead to surfaces of random roughness, laser treatment techniques yield irregularities that are extremely ordered and uniform. Many studies have demonstrated that cells participating in the osseointegration process prefer implant surfaces with strictly regular and ordered superficial roughness characteristics [9-12]. The work reported here summarizes the results from a pilot *in vivo* investigation on an animal model on the effect of laser parameters (wavelength, beam power, scanning motion, pulse frequency) on implant surfaces and the subsequent osseointegration process.

Materials and Methods

Laser-modified titanium implants

Eleven male New Zealand rabbits with an average weight of 4.0 kg were used in these studies. Twenty-one 3 mm x 6 mm laser-treated titanium implants were inserted. Using a pulsed, diode-pumped solid state (DPSS) source laser, in a Q-Switch output (Nuova Geass), the titanium implant surfaces were covered with a series of pores, each with a specific diameter and pitch. The laser beam power was set to generate pores of diameter between 5 and 40 μm , whereas scanning motion and pulse frequency were set in order to generate pores of pitch between 20 and 50 μm (Table 1). Morphologic analyses of the titanium implant surfaces were carried out using a Scanning Electron Microscope (SEM) (SEM LEO 420, Leo Electron Microscopy Ltd, Thornwood, NY, USA). Using the SEM, it was possible to verify the dimensions of and spaces between the pores. In the present study, seven different combinations of diameter and pitch of pores were tested. The purity of the implants was confirmed by an electron spectroscopy for chemical analysis (ESCA) carried out by Nobil Bio Recherche (Nobil Bio Recherche, Villafranca D'Asti, Italy) (Cassinelli & Morra; Reference nr: 2500700105 for Nuova Geass) and by energy-dispersive x-ray

TABLE 1: Processing pattern groups

Implant type	Processing pattern group	Depth	Pitch
Laser 5 μm , pitch 15 μm	1	5 μm	15 μm
Laser 10 μm , pitch 15 μm	2	10 μm	15 μm
Laser 20 μm , pitch 20 μm	3	20 μm	20 μm
Laser 20 μm , pitch 30 μm	4	20 μm	30 μm
Laser 25 μm , pitch 30 μm	5	25 μm	30 μm
Laser 30 μm , pitch 40 μm	6	30 μm	40 μm
Laser 40 μm , pitch 50 μm	7	40 μm	50 μm

spectroscopy (EDX) and x-ray photoelectron spectroscopy (XPS) analyses carried out by the Faculty of Engineering at the University of L'Aquila (L'Aquila; Italy) [13].

Experimental procedure

This study was approved by the Regional Ethics Committee for Laboratory Animal Research of the Catholic University of Rome. Twenty one experimental implants were placed in 11 rabbits: three implants for each experimental surface were tested. Surgery was executed under general anaesthesia, induced with a mixture of Ketavet 3 cc (Gellini-Intervet, Milan, Italy) and Domitor 1cc (Pfizer Inc., NY, USA). Implants were inserted under standard surgical protocol. In each rabbit, an incision was made by the distal femoral epiphysis. Two implants, randomly selected, were placed: one in the right distal femoral epiphysis, and the other in the left distal femoral epiphysis. There was one rabbit in which only one implant was inserted. Implant sites were prepared with a low round number drill (less than 800 rpm), and under continuous irrigation of physiological solution. After surgery, all implants showed primary stability. Teflon cover screws were applied on implant chambers (Nuova Geass, Udine, Italy)

(Fig. 1). Surgical sites were sutured with absorbable polyglycolic acid suture Dexon II (Kendall Company, Mansfield, MA, USA). Afterwards, implant sites were disinfected with a spray (Betadine, Asta Medica GmbH, Frankfurt, Germany) and subjected to antibiotic topic therapy with Kana spray (Gellini-Intervet, Milan, Italy). Rabbits underwent systemic antibiotic therapy using Enrofloxacin 1 ml, 5% (Baytril, Bayer, Leverkusen, Germany). Animals were sacrificed in a CO₂ gas chamber eight weeks after surgery.

Histologic and histomorphometric procedures

All bone specimens were immediately rinsed in saline, fixed in 10% neutral buffered formalin, and processed to obtain thin ground sections. Afterwards, they were dehydrated in an ascending series of alcohol rinses and then infiltrated with methacrylate (from 50% ethanol/resin to 100% resin, with each step lasting 24 hours). The photopolymerization was obtained using a 48 hour exposure to blue-light and the implants were oriented in order to visualize the two different sides at the same time. After polymerization, the blocks were ground to remove the excess of resin and to expose the implant and then were glued on plastic slides using a methacrylate-based glue. A Micromet high speed rotating blade microtome (Remet, Bologna, Italy) was used to separate the section from the block, thus obtaining a 250 µm thick section. The section was then ground down to about 40 µm using a LS-2 grinding machine (Remet, Bologna, Italy) equipped with waterproof grinding paper. Each section was polished with a polishing paper and 3 µm polishing cream. Buffered 10% toluidine-blue staining was used to analyze the different ages and remodeling pattern of the bone and basic-fuchsin was used to distinguish the fibrous tissue. On each specimen, the total, thread and body bone-to-implant contact (BIC), bone volume (BV) and expected bone-to-implant contact (EBC) percentages were estimated, and a statistical analysis was carried out. The histomorphometric analysis was performed by digitizing the images from the microscope using a JVC TK-C1380 Color Video Camera (JVC, Yokohama, Japan) and a frame grabber. The images were acquired with a 10x objective including the entire implant surface. Subsequently the digitized images were analyzed with image analysis software IAS 2000 (Delta, Sistemi, Italy). For each implant, the most central section was analyzed. Total BIC was estimated

FIGURE 1. Teflon cover screws applied

FIGURE 2. Thread and body areas over implant surface

TABLE 2. Histomorphometric evaluations relative to different laser spots diameter

Processing pattern group	5	10	20/20	20/30	25	30	40
Total BIC	51.1 ± 17.8	52.1 ± 2.2	51.5 ± 13.5	49.3 ± 19.7	49.4 ± 33.9	46.4 ± 4.4	49.5 ± 9.5
Threads BIC	49.0 ± 13.7	48.3 ± 4.6	53.4 ± 14.6	49.5 ± 16.1	46.0 ± 32.8	49.1 ± 3.8	48.4 ± 7.8
Body BIC	62.2 ± 45.4	45.4 ± 25.9	47.9 ± 7.8	55.2 ± 22.6	54.2 ± 32.0	56.2 ± 5.9	47.9 ± 12.7
Bone Volume	41.1 ± 4.3	34.5 ± 5.0	35.9 ± 5.9	35.6 ± 4.4	40.6 ± 18.4	33.2 ± 13.9	40.6 ± 0.8
EBC	33.2 ± 6.8	38.2 ± 2.5	33.3 ± 6.0	30.0 ± 9.6	31.1 ± 10.2	28.9 ± 12.0	35.3 ± 8.7
Δ (Tot BIC-EBC)	17.9	13.9	18.3	19.2	18.3	17.5	14.2

Percentage ± SD; BIC= bone-to-implant contact ; EBC= expected bone-to-implant contact

by counting the length of implant surface covered by bony trabeculae over the total length of the implant. Thread BIC was estimated by counting the length of implant surface covered by bony trabeculae exclusively over thread areas, and body BIC exclusively over body areas, as shown in Figure 2. Bone volume represents the amount of bone matrix measured over the entire microscopic field. This measurement was accomplished by outlining the bone islands and surfaces to determine the surface area of bone in each particular microscopic field, representing in clinical terms “bone quality”. Using the stereologic basis according to Parfitt [14], the two-dimensional measures of the bone surface area were expressed as a three-dimensional percentage volume of the total volume. According to Trisi *et al.* [15], expected bone-to-implant contact was determined by the “superimposition technique” and represent the BIC expected on the day of implant placement at a time when the implant surface had not yet generated any effect on bone growth. Using graphic software (Xres 2.0, Macromedia, city, country), images at 50x magnification were obtained of the implant and the bone just adjacent to the implant surface. The implant profile was superimposed on the blank bone image at three locations:

150, 500, and 1,000 μm lateral to the original implant surface and the mean BIC was calculated on this virtual implant interface. Because the locations of the superimposed images were so close to the actual implant profile, the BIC on this implant image could be considered the BIC that would be expected the day the implant was placed in this particular bone quality. The BIC was calculated on the virtual implant interface with the same technique used for the standard histologic images of the implant interface¹⁵.

Statistical analysis

On each specimen, the total, thread and body BIC, BV and EBC percentages were estimated.

The BIC values obtained for each of the seven laser diameters were summarized as mean and standard deviation (Table 2). Calculations for median and range produced very similar results (data not shown). To simplify interpretation, the BIC values were plotted versus laser spot diameter (Figure 3). Statistical significance was evaluated by Kruskal-Wallis one-way analysis of variance by ranks. Data were elaborated using SPSS®

FIGURE 3. (A) BIC % of total, thread and body values relative to different laser spots diameter. (B) Total BIC%, Expected bone-to-implant contact and their difference (Δ), bone volume values, relative to different laser spot diameters.

FIGURE 4. Histology of samples from seven pattern processing groups (10x, stained with Toluidine Blue). (A) LASER 5/15: This implant shows excellent osseointegration. It is surrounded almost entirely by bone trabeculae, although the bone contact is not uniform. (B) LASER 10/15: This implant shows good osseointegration, but it is not entirely surrounded by bone trabeculae. Trabeculae are well oriented and present a normal thickness and disposition. (C) LASER 20/20: Poor number of trabeculae around the screw. At higher magnification there is presence of a thin bone layer surrounding the titanium. (D) LASER 20/30: Good osseointegration; a large number of trabeculae visible. Trabeculae represent a normal thickness and orientation, but closer to the implant they tend to thicken. (E) LASER 25/30: High trabecular, connectivity and osseointegration. (F) LASER 30/40: Good osseointegration, with a large number of peri-implant trabeculae, presenting normal thickness and orientation. (G) LASER 40/50: A large number of peri-implant trabeculae is visible, presenting normal thickness and orientation. A thin bone layer coats titanium uniformly.

Advanced Statistical TM 13 (2004, Chicago, USA) software package.

Results

All the animals recovered well after surgery. No complications were evident during healing. At the sacrifice all the implants appeared stable.

Histologic observations for seven processing pattern groups

1. Laser 5/15 (Fig. 4a)

All implants in this group showed excellent osseointegration. They were surrounded almost entirely by bone trabeculae, of which the functional direction was not entirely normal. A thickening was observed around peri-implant trabeculae not

completely surrounding the screw. Bone contact was not uniform, and zone of direct contact and zone of medullary tissue interposition alternated.

The bone in direct contact with the screw was mainly of the composite type.

2. Laser 10/15 (Fig. 4b)

All implants in this group showed a good osseointegration, but they were not entirely surrounded by bone trabeculae. Trabeculae were well oriented and presented a normal thickness and appearance. Peri-implant thickening was not observed, but trabeculae made contact with the screw in the zone in which they met. Typical osseous slipping was not present, but zone of direct contact and contact zone of wide areas of medullary tissue alternated. Bone in direct contact with the screw was

mainly composite type, and almost present on the thread surface.

3. Laser 20/20 (Fig. 4c)

All implants appeared to show poor osseointegration, as few trabeculae were observed around the screw; however, at higher magnification, a thin bone layer was observed on the entire surface of the titanium implant, even if it was not in direct contact with surrounding bone trabeculae. In other words, it showed low connectivity. Peri-implant thickening was not observed, but there was contact of trabeculae with the screw in the zone in which they met. Typical osseous slipping was present, but the zone of direct contact and zone of medullary tissue interposition alternated. Bone in direct contact with the screw was mainly of the composite type, and almost present on thread surface.

4. Laser 20/30 (Fig. 4d)

All implants showed good osseointegration, with a large number of trabeculae surrounding the implants. Trabeculae presented with a normal thickness and orientation, but close to the implant they tended to thicken. Unlike the previous group, the thin bone layer surrounding the titanium implant was not particularly visible, because in the larger contact zones, normal trabeculae were present, with high levels of connectivity. Bone was in contact both in threads and in the implant body, even if zone of direct contact and thin contact zones of medullary tissue interposition alternated. For that reason the BIC level was quite low. Bone in direct contact with the screw was mainly of the composite type.

5. Laser 25/30 (Fig. 4e)

The implants in this group showed a high degree of variability, making analysis difficult. Trabecular connectivity and osseointegration levels were high in some sections of the implants, but in the others, either peri-implant trabeculae or connectivity were absent. Over the implant surface, the presence of a thin bone layer, not connected to the surrounding bone structure, was detected.

6. Laser 30/40 (Fig. 4f)

All implants in this group showed good osseointegration, with a large number of peri-implant trabeculae, presenting normal thickness and orientation. A thin bone layer coated the entire titanium implant, but the contact with titanium was not continuous, due to alternation of zones of direct contact and zones

of medullary tissue interposition. Bone was continuous around implant threads and body, where superficial roughnesses, due to surface treatment, were visible. Bone in direct contact with the screw was mainly of the composite type.

7. Laser 40/50 (Fig. 4g)

Results were comparable to Laser 30/40 group, described above.

Histomorphometric results

Results concerning osseointegration are reported in the Table 2. As shown by statistical means and by histological pictures, there were no relevant differences between the seven different processing pattern groups. The average total BIC values were approximately 50% for all test surfaces. In addition, thread BIC values did not mirror the body BIC values. EBC values are always lower than total BIC percentages: from 28.9% for Laser 30 group, to 38.2% for Laser 10 group. Total BIC % – EBC % (Δ values) are different for all tested groups. The best result was found in Laser 20/30 group, where $\Delta=19.2$. These results were plotted in Figure 3 and 4. Figure 3 shows a trend of slightly increasing BIC values with increasing laser spot diameter total, thread and body subsamples. Figure 4 shows the modifications of total, actual contact obtained (BIC), expected contact (EBC) and their differences (Δ). The laser diameter ≤ 10 and 40 were discordant. While the laser diameters 20/20 and 20/30 were constant, indicating a similar degree of osseointegration, the difference between the actual contact obtained and the expected contact was approximately 20%.

Scanning electron microscopy

Laser-induced modifications to titanium implant surfaces were characterized by their regularity and homogeneity due to the smooth pseudospherical cavities obtained with the laser surface treatment.

1. Laser 5/15 (Fig. 5a, 5b)

The laser source was adjusted to obtain pores that had a diameter of 5 μm and pitch (distance between centers of pores) of 15 μm . The fused border of the pore had greater visibility than in other patterns and it covered part of the interspace between the pores.

2. Laser 10/15 (Fig. 5c, 5d)

This pattern was created by adjusting the laser to obtain pores 10 μm in diameter and 15 μm in pitch. In this case, the fused

FIGURE 5. SEM of titanium inserts from seven pattern process groups. (A,B) Laser 5/15: Fused border of the pores has greater visibility than in other patterns and it covers part of the interspace between the pores (Mag.: 500 x; 5.00 k x). (C,D) Laser 10/15: Fused border of the pores has a more fragmented aspect because the fused material is distributed on a longer circumference (Mag.: 500 x; 5.00 k x). (E,F) Laser 20/20: Diameter of the pores equaled the pitch, and the pores were tightly packed (Mag.: 1.00 k x; 5.00 k x). (G,H) Laser 20/30: Fused material was minimized because of the long circumference of the pores and the spacing between them (Mag.: 1.50 k x; 5.00 k x). (I,J) Laser 25/30: For this pattern, diameter was 25 micrometers and pitch was 30 micrometers (Mag.: 500 x; 2.50 k x). (K,L) Laser 30/40: For this pattern, diameter was 30 micrometers and pitch 40 micrometers (Mag.: 500 x; 2.50 k x). (M,N) Laser 40/50: In this pattern, diameter was 40 micrometers and pitch 50 micrometers (Mag.: 500 x; 500 x).

border of the pores had a more fragmented appearance because the fused material was distributed on a longer circumference and this effect tended to produce droplets that adhered to the surface of the titanium implant.

3. Laser 20/20 (Fig. 5e, 5f)

In this case, the diameter of the pores equaled the pitch, and the pores were closely packed (no space left between them).

4. Laser 20/30 (Fig. 5g, 5h)

For this pattern, the diameter was 20 μm and the pitch was 30 μm . In this case, the laser source created holes with an average depth of 7 μm and the fused material was minimized because of the long circumference of the pores and the spacing between them. It was evident from the images that the laser did not provoke crack formation, which might alter the mechanical properties of the implant. The formation of cracks was prevented by the low volume of material affected by thermal effects around the pores – a characteristic of the laser used to create these patterns.

5. Laser 25/30 (Fig. 5i, 5j)

For this pattern, the pore diameter was 25 μm and pitch was 30 μm . The depth of the pores was the same as that observed with the 20/30 pattern (7 μm).

6. Laser 30/40 (Fig. 5k, 5l)

For this pattern, the diameter was 30 μm and pitch 40 μm . Again, the depth of the pores was 7 μm .

7. Laser 40/50 (Fig. 5m, 5n)

In this pattern, diameter was 40 μm and pitch 50 μm . Again, the depth of the pores was 7 μm .

Discussion

Even if achieving a good average BIC value, around 50%, seems to be a promising result, there were no relevant differences between tested implant surfaces. But many important considerations on this study are needed. First, BIC, both total and thread, within the same processing pattern group, had a high degree of variability, illustrated by high SD values (Table 2). This is due, in part, to the low number of specimens used in this pilot study. BIC thread variance did not mirror the BIC body variance. It is possible that laser beam angle is able to modify both the superficial roughness and, therefore, the histological response to this surface. This limit of the laser machine prototype was partially overcome by means of the setting-up of a definitive microroughening technique. Despite the small sample size, a decrease in osseointegration was observed with increasing pores dimension. Although it has been reported that the rate of BIC between titanium and bone depends more on the surface condition than on material composition [16], the peri-implant BV should be an important determinant of the percentage of BIC. Measuring the EBC, BIC values for a particular bone density type were found to be unaffected by the conductive properties of the implant surface. This ideal value can be used as a control and compared with the actual BIC for each particular type of surface. If an implant surface induces a BIC lower than that expected based on the peri-implant BV (EBC), this surface is defined as a nonconductive surface (actual BIC < EBC). If an implant surface induces the same BIC as the EBC based on the peri-implant BV (EBC), this surface is defined as a bone-neutral surface (actual BIC = EBC). If an implant surface induces a BIC higher than the EBC based on the periimplant BV, this surface is defined as a bone-conductive surface (BIC > EBC). Actual BIC values show that all tested surfaces are osteoconductive, because all EBC values were al-

ways lower than the BIC percentages. Indeed, osteoconductivity is the ability of an implant surface to conduct the growth of the bone over its surface. Similar studies confirm the reliability of implant surface laser treatment techniques [6,17-19]. Many studies stressed how laser treatment on smooth titanium implant surfaces induced a unique morphology, without cavity-like features or other morphological elements that could contain irremovable contaminations, such as traces of aluminium or silica left over from implant sandblasting [2,20]. These surface contaminations can be avoided by the use of laser treatment to process implant surfaces [20]. Similar conclusions were reported in one study of Gaggli *et al.* [2] where the laser processing procedure was described as a viable method of implant surface treatment, able to guarantee a high degree of titanium purity and thus a high degree of osseointegration. Previous methods of producing surface roughness, such as the titanium-plasma method and the aluminium oxide-blasted surface, did not result in optimal surface purity. Aluminium oxide-blasted implants caused surface contamination with aluminium particles, although subsequent treatment with hydrogen fluoride should guarantee a high degree of titanium purity and osseointegration. It is primarily the microscopic structure of the implant surface that is responsible for bone apposition to the implant. Other studies have shown that high-energy laser treated implants showed higher removal torque values than smooth, sandblasted, low energy laser and titanium oxide treated implants [6]. Minimum roughness dimensions of 20-50 μm are necessary for optimum osseointegration. Roughness of only 1-10 μm of dimension either did not improve torque value or made it worse [6]. A pilot study by Hallgren *et al.* [18] showed no biomechanical (removal torque) or histomorphometrical (BIC) differences between the test group (laser implants) and the control group (machined implants). In a follow-up study, the same authors compared the laser surfaces that gave the best results in the pilot study, with smooth titanium implants. The test group showed better outcomes in biomechanical tests: removal torque values was 52 N/cm in the laser treated implants vs. 35 N/cm in the control screws. Percentage BIC also demonstrated higher values in test implants than in control ones (40% in laser implants, 32% in machined implants). In a study by Cho and Jung [17], in which seven machined implants and seven laser-treated implants were compared (CSM implant, CSM, Daegu, Korea), better results were achieved in the test group than in the control group. Eight weeks after implant insertion, the average removal torque registration was 23.58 ± 3.71 N/cm for machined screws and 62.57 ± 10.44 N/cm for laser implants. Torque values showed statistically significant differences between the smooth implant

group and the laser-treated implant group ($p=0.00055$) (Wilcoxon's signed-rank test). In a study by Moritz *et al.* [19], a surface coated by a Bioactive Glass (BAG), via laser CO₂ processing, was analyzed. Cylindrical BAG-coated implants, used as test samples, and NaOH-treated, titanium-sandblasted implants, used as a control group, were inserted into the femoral epicondyles of six rabbits. After eight weeks of healing, surface-treated implants registered removal torque readings significantly higher than those in the control group. In the test group, even the quantity of new peri-implant bone was higher.

In conclusion, the purpose of this study has been to confirm the validity of implant surface treatment procedures, in compared with the EBC. These surfaces were all shown to be osteoconductive. Implants with pores of 20 and 25 μm achieved better performances than implants with other pore sizes, suggesting further experiments in order to confirm these preliminary data.

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