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The effects of implant surface roughness and surgical technique on implant fixation in an *in vitro* model

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Key words: implant, *In vitro* model, surface roughness, surgical technique

Abstract

Objectives: The aim of the present study was to determine the relationship between implant surface parameters, surgical approach and initial implant fixation.

Material and methods: Sixty tapered, conical, screw-shaped implants with machined or etched surface topography were implanted into the explanted femoral condyle of goats. The implant sites were prepared either by a conventional technique, by undersized preparation, or by the osteotome technique. Peak insertion & removal torque, bone-to-implant contacts (BIC) and morphological bone appearance were assessed by scanning electron microscope (SEM) and micro-computer tomography (micro-CT).

Results: Insertion and removal torque values were significantly higher for etched implants inserted with the undersized technique (115.2 ± 31.1 , 102.9 ± 36.4 Ncm) respectively. Also, the average BIC value was higher for the etched implants placed with the undersized technique (87.5 ± 5.6), which was statistically significant compared with machined and etched implants inserted by conventional technique.

Conclusion: In conclusion, this study shows that the surgical technique has a decisive effect on implant fixation (represented in this study by installation torque value/removal torque value and histomorphometric evaluation) in trabecular bone. Nevertheless, additional *in vivo* studies have to be done to prove the importance of surgical protocol for the final implant–bone response.

The final clinical success of oral implants is determined by various implant and non-implant-related parameters. Implant-related parameters are implant shape; implant surface configuration and implant surface composition. Non-implant-related parameters are mainly dealing with the skills of the surgeon, health condition of the patient and final loading protocol of the implant supported prosthetic construction (Albrektsson et al. 1981). In view of this, the goal of implant installation is to obtain a good primary stability of the implant in the alveolar jawbone. The establishment of a mechanically stable interface between the implant and bone prevents the devel-

opment of a fibrous tissue capsule. Subsequently, the implant surface parameters are supposed to stimulate the bone cell reaction resulting in an enhanced healing response and improved implant–bone contact (BIC). In view of this, there are numerous reports that suggest a beneficial effect of implant surface roughness on implant healing and survival (Wennerberg et al. 1997; Vercaigne et al. 2000a, 2000b; Ogawa & Nishimura 2003). Nevertheless, a recently performed systematic review (Shalabi et al. 2005) dealing with surface roughened implants in experimental animal studies showed that the effect of surface roughness on oral implant behavior

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is not as straightforward as claimed. Considering that the surgical technique varies between the different studies, the additional effect of surgery on the outcome of a lot of experimental implant studies is perhaps underestimated. This suggestion is supported by two recent studies that reported about 93–96% implant success in bone of poor quality and density. In these studies, the original Brånemark implant design was used that was neither surface roughened, oxidized, or coated with hydroxyapatite (HA.) Only the surgical routine was changed in that one author preferred to avoid drilling for the full length of the implant and the other drilled more narrow holes than generally recommended (Friberg et al. 1999; Bahat 2000). Apparently, such changes in surgical routine can be of paramount importance for clinical success. Still they do not rule out the possibility that hardware alterations may be of some significance as well [Albrektsson 2001]. Therefore, because of the controversial results of clinical data, it is currently impossible to determine whether one particular implant is superior to another or if one specific implant design has to be preferred [Puchades-Roman et al. 2000; van Steenberghe et al. 2000; Gotfredsen & Karlsson 2001; Engquist et al. 2002; Astrand et al. 2004; Wennstrom et al. 2004].

To obtain a better insight into the above mentioned contradictory data, we postulated the hypothesis that surgical planning and technique are one of the major parameters for the final implant–bone behavior and are perhaps more important than implant surface parameters, like roughness and composition.

As part of a consecutive series of studies, the purpose of this paper is to determine the relationship between implant surface parameters, surgical approach and initial implant fixation. For this purpose, an *in vitro* experiment was done in which machined and roughened implants were inserted in bone using different surgical protocols. The mechanical fixation as well as appearance of the implant–bone interface was evaluated.

Material and methods

Implants

Sixty tapered, conical, screw-shaped dental implants (Biocomp* Industries, Dordrecht,



Fig. 1. Machined and etched implants as used in the study.

the Netherlands) were used. All implants measured 10 mm in length and had a diameter of 4.6 mm (Fig. 1).

The implants were divided into two groups with each a different surface roughness, i.e. turned, machined and roughened (by grit-blasting and additional acid etching).

A Universal Surface Tester (UST[®], Innowep GmbH, Würzburg, Germany) was used to characterize the surface topographies of the implants. This equipment includes a diamond stylus, consisting of a 60° cone, which is moved across a surface with a load of 10 mN. For the topographical analysis of the implants, the roughness (R_a) of the threaded area of three screws from each group were selected at random and measured.

Bone specimens

Drilling specimens were collected from the femoral condyle of goats. This bone consists mainly of trabecular bone with a thin shell of cortical bone. The bone pieces were obtained within 2 h of animal's death and stored on ice at 4°C during transportation from the central animal facility to the laboratory.

Implant installation

All bone preparations for implant installation were performed with a very gentle surgical technique by using a dental drill (KaVo EWL Dental GmbH, Biberach, Germany) at a rotational speed of 2000 rpm. The drill was fixed in a drill standard with copious external cooling.

Three different surgical approaches were applied for the installation of the implants:

Approach 1: a press fit technique (according to the protocol of the manufacturer). Drilling started with a rosen drill (diameter

3 mm), a lindemann drill (diameter 2.3 mm for the tip) and pilot drill (diameter 2.55 mm). Subsequently, the hole was widened by a consecutive series of drills, i.e. 3.4, 4, and 4.6 mm drill in diameter. Installation of the implants was done using a Digital[®] torque gauge instrument (MARK-10 Corporation, New York, NY, USA) in order to measure the in screw torque value.

Approach 2: an undersized preparation. The same drilling sequence was used as for approach 1, but now the final drill was 4 mm in diameter. Subsequently, the 4.6 mm implant was installed with the torque instrument.

Approach 3: an undersized preparation was made in combination with the use of a spreader (or osteotome). The drilling was up to a drill with a diameter of 2.55 mm. Then, spreaders with a diameter of 3.4, 4, and 4.6 mm were used for the further preparation of the implant site. The length of the spreaders was 10 mm and each spreader was left for 1 min inside the preparation before the next diameter was used. Finally, the implant was installed manually with the torque instrument.

Mechanical testing

Eight specimens of each implant type and surgical approach were prepared to determine torque removal force. The torque-measurement device detects and registers the torque necessary to remove the implant.

The specimens were placed on a support jig for screw-out measurement. This support jig can be adapted in all directions, to put a perpendicular force on the implant. For the screw-out test, a slowly increasing torque (displacement 0.5 mm/min) was gradually applied to each implant until loosening. When the peak force representing implant loosening was reached, the torque-out procedure was finished.

Histological procedures and histomorphometric evaluation

After implant insertion, two implants of each approach and surface roughness were fixed by immersion in 4% buffered formalin. Then, these samples were dehydrated in a graded series of ethanol and embedded in methylmethacrylate. About 10- μ m-thick sections of the implants and surrounding tissues were prepared using a modified diamond blade sawing micro-

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tome technique. These sections were made parallel to the long axis of the implant surface. The sections were stained with methylene blue and basic fuchsin stains and the amount of bone contact to the implants was determined with help of a light microscope and an image analysis system (Leica Qwin Pro, Wetzlar, Germany).

Scanning electron microscope (SEM)

Following the torque measurement, specimens were fixed and dehydrated by a graded series of ethanol and embedded in methylmethacrylate. After polymerization the samples were hemi-sectioned perpendicular on the longitudinal axis of the implants. After polishing, the specimens were examined with a SEM (JEOL 6310, Jeol Ltd., Tokyo, Japan) in the back-scatter mode in order to analyze the bone appearance after the surgical procedure and the location of the fracture interface after the screw-out test.

Micro-computer tomography (micro-CT) evaluation

Also micro-CT equipment (SkyScan-1172, Skyscan n.v., Aartselaar, Belgium) was used for 3D analysis of bone deformation as a result of the implant insertion. For this procedure, polymeric (MMA) replicas were made of the machined and roughened implants. This was done because the use of pure titanium implants results in scattering of X-rays at the bone implant interface, which does not allow a proper analysis of this area. MMA resin implants were installed using the same approaches as for the

bulk titanium implants (four samples for each approach). After installation, specimens were cut into smaller size and placed in the middle of a cylindrical sample holder. High-resolution scanning and cone beam reconstruction were performed with a pixel size of 25.76 µm and a slice thickness of 25 µm. The number of slices was set at 472 to cover the entire length of the implant, i.e. 10.3 mm. This was in correspondence with the preliminary measured thickness of the bone specimen as measured with calipers and the interactive measurements of the cross-section of the sample. The micro-CT software was used to make a 3D reconstruction from the obtained set of scans.

Statistical analysis

Mean values and standard deviations (SD) were calculated. One-way analysis of variance (ANOVA) was used for comparing the differences between groups. All calculations were performed through the use of GraphPad[®] Instat 3.05 software (GraphPad Software Inc., San Diego, CA, USA). Differences were considered as significant when $P < 0.05$. The Dixon outlier test was used to calculate and exclude outlying data from the dataset.

Results

Surface roughness measurements of implant surface

Surface topographic evaluation demonstrated that both experimental surfaces differed in surface roughness (Table 1). The machined surface showed an average surface roughness ($R_a = 0.45 \mu\text{m}$), which was significantly lower than the etched surface ($R_a = 1.47 \mu\text{m}$). The values for the

parameter R_{sk} showed that the two surfaces had a positively skewed surface; the surfaces consisted of more peaks than valleys. Although, the data appeared to indicate that the etched implants had a higher R_{sk} mean value, statistical testing revealed that the difference was not significant (Table 1).

Mechanical testing

The results of the torque measurements are listed in Table 2 and depicted in Fig. 2. Statistical testing showed that significant differences existed between the various surgical approaches (Table 2). The highest in screw and removal torque (115.2 and 102.9 N cm, respectively) values were always observed for the etched implants installed with Approach 2.

Histological and histomorphometric evaluation

The histological evaluation did not demonstrate a difference in bone contact within the same approaches between the various implants, i.e., no effect of implant surface properties (machined vs. roughened) was seen. For all surgical approaches the highest amount of bone-implant contact (BIC) was observed around the most coronal part of the implants.

Further in Approach 1 sections the trabecular bone was found to be mainly in contact with the top of the screw threads (Fig. 3). A lot of bone debris was seen at the apex of the implants (Fig. 4). Occasionally, bone debris was also present in the trabecular voids.

Approach 2 sections showed that in the trabecular bone area the major part of the implants was in close contact with bone.

Table 1. The mean \pm SD value of surface roughness parameters

	Machined	Etched	P-value
R_a	0.45 \pm 0.32	1.47 \pm 0.54	< 0.05
R_q	0.29 \pm 0.1	1.82 \pm 0.68	< 0.01
R_{sk}	0.08 \pm 0.35	0.29 \pm 0.35	> 0.05

Amplitude parameters (A. Wennerberg)

R_a – the arithmetic mean of the absolute values of the surface departures from a mean plane within the sampling area. It is measured in micrometers.

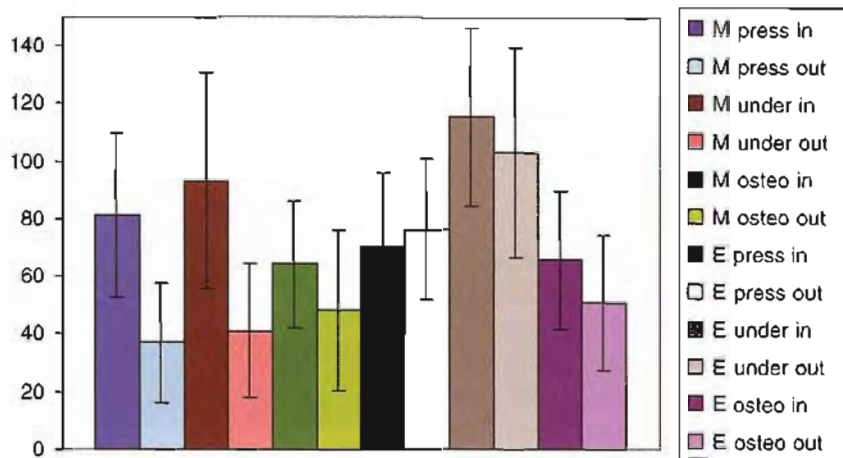
R_q – the root mean square value of the surface departures within the sample area. It has the statistical signification as the standard deviation of the height distribution.

R_{sk} – the measure of the symmetry of surface deviations about the mean plane. A negatively skewed surface has more valleys than peaks.

Table 2. The mean \pm SD (N cm) of the removal torque value (RTV)

Group	Mean \pm SD	Comparison	P-value
M press in	81.2 \pm 28.4	M press out vs. E under out	< 0.001
M press out	50.2 \pm 41.9	M under out vs. E under out	< 0.01
M under in	93 \pm 37.4	M osteo in vs. E under in	< 0.01
M under out	54.3 \pm 42.7	M osteo out vs. E under out	< 0.01
M osteo in	64.3 \pm 21.6	E press in vs. E under in	< 0.05
M osteo out	48.4 \pm 27.9	E under in vs. E osteo in	< 0.01
E press in	70.8 \pm 25.7	E under out vs. E osteo out	< 0.05
E press out	76.6 \pm 24.4		
E under in	115.2 \pm 31.1		
E under out	102.9 \pm 36.4		
E osteo in	66 \pm 24.1		
E osteo out	51 \pm 23.6		

M, machined; E, etched; In, in-screw; Out, out-screw; Press, press fit; Under, under-sized; Osteo, osteotome.



M = Machined & E = Etched.

In = In-screw & Out = Out-screw.

Press = Press fit, Under = Under-sized, Osteo = Osteotome.

Fig. 2. Graph showing the removal torque values (mean ± SD) as obtained with the various approaches.

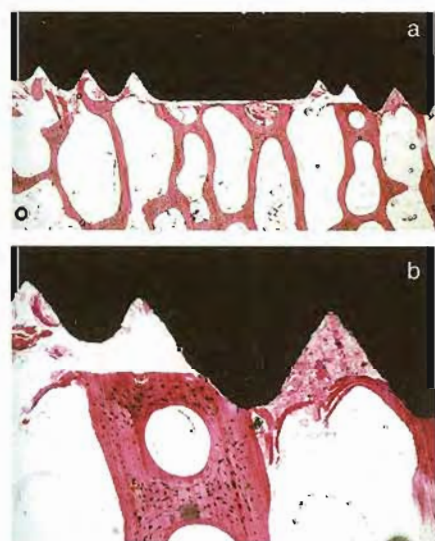


Fig. 3. Histological section of a roughened implant installed using the press-fit technique. (a). The bone trabeculae have only a limited contact with the implant surface (original magnification × 2); (b). Only occasionally bone debris was present in between the bone trabeculae (original magnification × 7).

The inner area of almost all screw threads was completely filled with bone. Also, a lot of fractured bone particles had penetrated in the trabecular voids (Fig. 5).

The light microscopical appearance of Approach 3 sections was in between both other approaches. Frequently, a lot of bone was observed in the inner area of the screw threads. Bone debris, i.e. small bone fragments were seen in the trabecular voids. Very occasionally (in two specimens), frac-



Fig. 4. Light micrograph of a roughened implant inserted with the press-fit approach. Fractured bone particles can be seen at the apex of the implant (original magnification × 2).

turing of bone trabeculae was observed. When present, this only occurred in trabeculae very close to the implant surface. Different from the other two approaches, a clear bone condensation was present at the implant apex (Fig. 6).

The histomorphometric evaluation showed (Table 3) that there was significantly less BIC for the machined press-fit inserted implants compared with the etched undersized and etched osteotome inserted implants. Statistical analysis also revealed that less BIC existed around etched press fit inserted implants compared with the two other approaches.

SEM

SEM confirmed our light microscopical observations. Similar to the histological



Fig. 5. Histological section showing the penetration of bone particles in the trabecular voids after installation of a machined implant using the undersized approach (original magnification × 7).

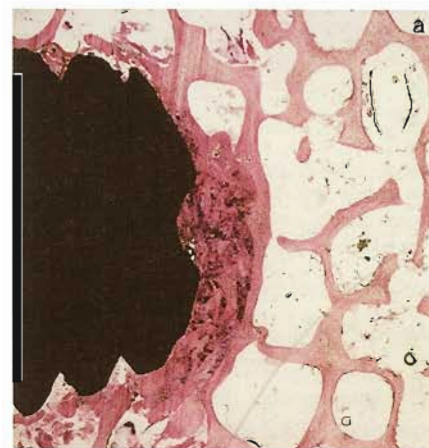


Fig. 6. Light micrograph showing bone densification at the apex of a roughened implant installed with the osteotome approach. (a) Original magnification × 2; (b) original magnification × 7.

sections, in approach 1 trabecular bone was seen to be in contact with only the tips of the screw threads. In approach 2, the majority of the screw threads were filled with bone tissue; a lot of debris was found in the trabecular voids as well as in the inner area of the screw threads (Fig. 7). Also in approach 3 some fractured bone trabeculae and debris was observed in the trabecular voids.

Table 3. Bone-implant contact percentage

Group	Mean \pm SD	Comparison	P-value
M press	54.4 \pm 10	M press vs. E under	<0.01
M under	71 \pm 10.2	M press vs. E osteo	<0.05
M osteo	65.9 \pm 8.7	E press vs. E under	<0.001
E press	48.7 \pm 2.8	E press vs. E osteo	<0.01
E under	87.5 \pm 5.6		
E osteo	77.6 \pm 16.6		

M, machined; E, etched; In, in-screw; Out, out-screw; Press, press fit; Under, under-sized; Osteo, osteotome.

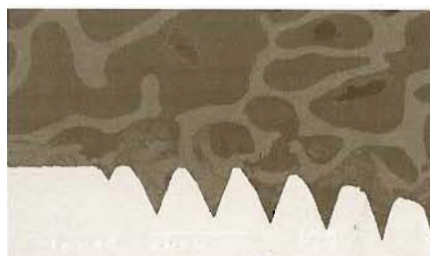


Fig. 7. Scanning electron microscope micrograph of an etched implant showing that the majority of screw threads are filled with bone trabeculae when the undersized implant installation approach is used.

Micro-CT evaluation

By doing 3D reconstruction of the micro-CT images, the three approaches were compared (Fig. 8). Similar observations were done as with the light microscopical sectioning and SEM imaging. Approach 1 and 3 resulted in well-defined defects, in which the cavity walls could be recognized very clearly. In approach 3, more bone debris appeared to be present at the coronal side of the bone defect. In approach 2, the defect wall was very difficult to recognize. A lot of small bone particles were present at the implant-bone interfaces, which were interspersed in the trabecular bone voids.

Discussion

In the present *in vitro* study we focused on the relationship between implant surface parameters, surgical approach and initial implant fixation. Our major finding is that the combination of implant surface roughness with undersized bone drilling results in the highest in- and out-screw values. Additionally, histological and micro-CT analysis showed that undersized drilling gives the highest amount of initial BIC. Combination of all these data indeed confirms that surgical approach has a high impact on the stability and fixation of oral implants. This corroborates with a recent

study of Buchter et al. (2003), who also evaluated *in vitro* implant stability of microrough implants inserted into the mandibular bone of minipigs by using a conventional burr technique, burr technique with additional thread cutting and osteotome technique. Resonance frequency analysis and removal torque were significantly better for the conventional burr technique compared with the osteotome technique. Our data appear to reduce the benefit of the osteotome technique for the installation of oral implants in bone of low density. However, we have to notice that the 'experimental' osteotome technique as used by us deviates from the original method as described by Summers (1994a, 1994b, 1994c), who did not use any pre-drilling at all. This might explain the lack of initially increased fixation in our study. We have to notice that the use of osteotomes is complex and requires additional training and skills from the surgeon. A proper orientation of the osteotome instrument is essential for the final shape of the created hole and the fit of the implant. For example, jiggling of the instrument during the bone densification procedure has to be prevented, because this can result in extensive widening of the hole in the apical implant area causing decreased implant fixation. Further, a recent study of Buchter et al. (2005) failed also to show a beneficial effect of the osteotome technique on the initial stability of implants, who claimed that this is because of the occurrence of microfractures in the peri-implant bone. Despite the agreement in results, this kind of microfracturing was not a constant observation in our histological sections, because it only occurred in a very limited way in only two of the samples.

Considering our findings also some comments have to be made on the final biological effect of implant surface morphology vs. surgical approach. Szmukler-Moncler



Fig. 8. Micro-computer tomography pictures showing the bone appearance after installation of the polymeric implant replica's. (a) Press-fit approach, (b) undersized approach, (c) osteotome approach.

et al. (2004a, 2004b) stated that it is bone interlocking at the interface that maintains the biological properties of textured surfaces, rather than a strong implant fixation *per se* and the pits carved during the etching process have a bone-interlocking capacity. Subsequently, osteogenesis at the bone-implant interface is supposed to be influenced by various mechanisms, including: (1) encouragement of endogenous ex-

pression of growth factors and cytokines by osteoblasts (Mustafa et al. 2002), (2) lower stimulation of osteocytes (the mechanoreceptor cell of bone), and (3) increased adhesion of monocytes/macrophages as well as blood platelets and their secretory profile (Park & Davies 2000; Park et al. 2001; Soskolne et al. 2002; Takebe et al. 2003). Recently, it has even been speculated that the bone healing events around implants follow a biological sequence that is not influenced to a great extent by the implant surface microtopography (Abrahamsson et al. 2004). Only the final outcome of the healing process would be determined by the implant surface texture, whereby micro-rough surfaces have to be considered as 'osteophilic'. The histology and micro-CT results of the current study leave space for a completely different explanation for increased bone formation. Because of the undersized and osteotome approach, a lot of small bone fragments are created and pressed in between the trabecular voids and in between the screw threads during implant installation. These bone fragments behave like a kind of autograft and can induce new bone formation. It can even be suggested that rough implant surfaces are more prone to adhesion of the bone fragments than machined implant surfaces resulting in an increased bone formation. Follow-up *in vivo* studies have to prove this hypothesis.

Finally, it is well known that the implantation of an implant in an undersized

implant socket alters the mechanical stresses in the peri-implant area in a significant manner compared with a press-fit inserted implant (Widmer et al. 1997; Taylor et al. 1998; Skalak & Zhao 2000). In the present study, the mismatch between implant and drill hole diameter was 0.6 mm. This contributed to the increased in- and out-screw values compared with the press-fit inserted roughened implants. Nevertheless, we do not know the magnitude of the stress as generated because of this specific size of mismatch. This lack of knowledge is significant, because high bone stresses can lead to increased bone remodeling and bone loss (Brunski 2003; Kitamura et al. 2004). All advices about size of the drill hole implant diameter and specific bone characteristics of the implantation site appear to be based on empirical experiences and lack clear scientific support. Therefore, studies have to be performed to the most optimal difference in diameter between drill holes and implant diameter and the subsequent biomechanical adaptation to stress (Hansson 2000, 2003).

In conclusion, this study shows that the surgical technique has a decisive effect on implant fixation (represented in this study by installation torque value/removal torque value and histomorphometric evaluation) in trabecular bone. Nevertheless, additional *in vivo* studies have to be done to prove the importance of surgical protocol for the final implant-bone response.

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要旨

目的:本研究の目的は、インプラント表面のパラメータと、外科術式及びインプラント初期固定との間の関係を明らかにすることであった。

材料と方法:山羊の大腿顆を摘出して、機械研磨またはエッチングした表面のテーパ-付きコニカル・スクリュー・インプラント60本を埋入した。インプラント骨床は、通法の術式、アンダーサイズ・テクニクまたはオステオトーム・テクニクによって形成した。最大埋入トルクと抜去トルク、骨-インプラントの接触面積(BIC)及び骨の形態を、走査電子顕微鏡(SEM)とマイクロ・コンピュータ断層撮影(マイクロCT)によって評価した。

結果:エッチング表面のインプラントをアンダーサイズ・テクニクによって埋入した時に、埋入と抜去トルクは各々最も高い値であった(115.2±31.1、102.9±38.4Ncm)。さらにBICの平均値はアンダーサイズ・テクニクで埋入したエッチング表面のインプラントが高く(87.5±5.6)、機械研磨のインプラント及び通法の術式で埋入したエッチング表面のインプラントに比べて、統計学的な有意差を示した。

結論:本研究は、外科術式が海綿骨内のインプラント固定に決定的な影響を及ぼすことを示した(本研究ではITV/RTVと組織形態計測学的評価によって表した)。しかし最終的なインプラント-骨の反応に関して外科的プロトコルの重要性を証明するには、さらに*in vivo*の研究を行う必要がある。

pression of growth factors and cytokines by osteoblasts (Mustafa et al. 2002), (2) lower stimulation of osteocytes (the mechanoreceptor cell of bone), and (3) increased adhesion of monocytes/macrophages as well as blood platelets and their secretory profile (Park & Davies 2000; Park et al. 2001; Soskolne et al. 2002; Takebe et al. 2003). Recently, it has even been speculated that the bone healing events around implants follow a biological sequence that is not influenced to a great extent by the implant surface microtopography (Abrahamsson et al. 2004). Only the final outcome of the healing process would be determined by the implant surface texture, whereby micro-rough surfaces have to be considered as 'osteophilic'. The histology and micro-CT results of the current study leave space for a completely different explanation for increased bone formation. Because of the undersized and osteotome approach, a lot of small bone fragments are created and pressed in between the trabecular voids and in between the screw threads during implant installation. These bone fragments behave like a kind of autograft and can induce new bone formation. It can even be suggested that rough implant surfaces are more prone to adhesion of the bone fragments than machined implant surfaces resulting in an increased bone formation. Follow-up *in vivo* studies have to prove this hypothesis.

Finally, it is well known that the implantation of an implant in an undersized

implant socket alters the mechanical stresses in the peri-implant area in a significant manner compared with a press-fit inserted implant (Widmer et al. 1997; Taylor et al. 1998; Skalak & Zhao 2000). In the present study, the mismatch between implant and drill hole diameter was 0.6 mm. This contributed to the increased in- and out-screw values compared with the press-fit inserted roughened implants. Nevertheless, we do not know the magnitude of the stress as generated because of this specific size of mismatch. This lack of knowledge is significant, because high bone stresses can lead to increased bone remodeling and bone loss (Brunski 2003; Kitamura et al. 2004). All advices about size of the drill hole implant diameter and specific bone characteristics of the implantation site appear to be based on empirical experiences and lack clear scientific support. Therefore, studies have to be performed to the most optimal difference in diameter between drill holes and implant diameter and the subsequent biomechanical adaptation to stress (Hansson 2000, 2003).

In conclusion, this study shows that the surgical technique has a decisive effect on implant fixation (represented in this study by installation torque value/removal torque value and histomorphometric evaluation) in trabecular bone. Nevertheless, additional *in vivo* studies have to be done to prove the importance of surgical protocol for the final implant-bone response.

要旨

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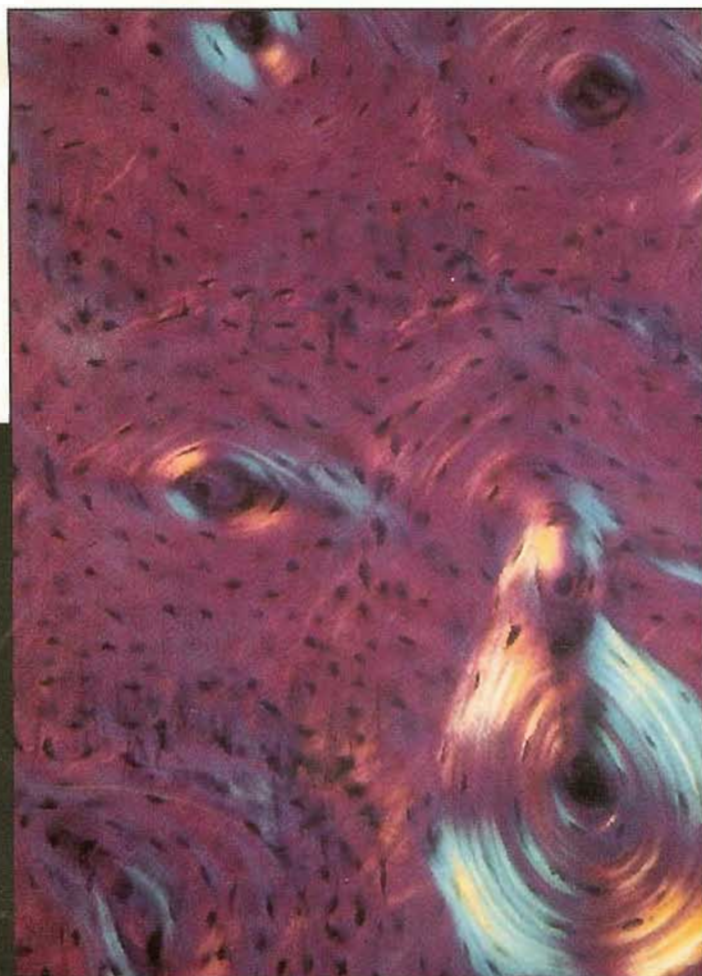
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