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Implant surface characteristics influence the outcome of treatment of peri-implantitis: an experimental study in dogs

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***Abstract*:**

Aim: To analyse the effect of surgical treatment of peri-implantitis without systemic antibiotics at different types of implants.

Material and methods: Four implants repeesenting four different implant systems – turned (Biomet 3i), TiOblast (Astra Tech AB), SLA (Straumann AG) and TiUnite (Nobel Biocare AB) were placed in the left side of the mandible in six dogs, 3 months after tooth extraction. Experimental peri-implantitis was initiated by placement of ligatures and plaque formation. The ligatures were removed when about 40–50% of the supporting bone was lost. Four weeks later, surgical therapy including mechanical cleaning of implant surfaces was performed. No systemic antibiotics or local chemical antimicrobial therapy were used. After 5 months, block biopsies were obtained and prepared for histological analysis.

Results: Two of the TiUnite implants were lost after surgical therapy. Radiographic bone gain occurred at implants with turned, TiOblast and SLA surfaces, while at TiUnite implants additional bone loss was found after treatment. Resolution of peri-implantitis was achieved in tissues surrounding implants with turned and TiOblast surfaces.

Conclusion: Resolution of peri-implantitis following treatment without systemic or local antimicrobial therapy is possible but the outcome of treatment is influenced by implant surface characteristics.

Key words: bone level; dental implants; infection; inflammatory lesion; peri-implantitis; titanium; treatment

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Peri-implantitis is a common biological complication in implant therapy and is characterized by inflammatory lesions in peri-implant tissues and an associated loss of supporting bone (Zitzmann & Berglundh 2008). It is by definition an infectious disease and the inflammatory lesion in peri-implant tissues develops as a result of accumulation of bacteria on implant surfaces. In a consensus report from the Sixth European Workshop on Periodontology, it was stated that because the disease is caused by bacteria, treatment should include anti-infective measures (Lindhe & Meyle 2008).

Different protocols have been suggested in the treatment of peri-implantitis. Non-surgical procedures alone appear to be insufficient to resolve peri-implantitis lesions (Renvert et al. (2008), while surgical procedures may promote access for removal of the biofilm formed on the implant surface and thereby attain resolution. There is limited information on the long-term outcome of treatment of peri-implantitis. Claffey et al. (2008) in a review article reported that data obtained from case series and animal experiments indicate that no single cleaning method including chemical agents used during surgical treatment of peri-implantitis was proven

Conflict of interest and sources of funding statement

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to be superior. Furthermore, most studies on treatment of peri-implantitis have used systemic antibiotics as an adjunct to the surgical therapy. Thus, it remains to be demonstrated whether resolution of peri-implantitis lesions can be achieved after surgical treatment without systemic antibiotics.

The outcome of treatment of peri-implantitis at implants with different types of surfaces has been evaluated in animal experiments. While most studies focused on bone fill and potential re-osseointegration in bone defects (Jovanovic et al. 1993, Wetzel et al. 1999, Shibli et al. 2003, Parlar et al. 2009), Persson et al. (2001a) reported that resolution occurred following surgical treatment in combination with systemic antibiotics at implants with smooth (polished) and modified [sand-blasted and acid-etched (SLA)] surfaces.

We have reported previously on the influence of implant surfaces in spontaneous progression of experimental peri-implantitis around commercially available implants in dogs (Albouy et al. 2008, 2009). Using the same model, we now report on the outcome of surgical treatment of experimental implantitis. The aim of the present study was to analyse the effect of surgical treatment of experimental peri- implantitis without systemic antiobiotics at different types of implants.

Material and Methods

Animals

The regional Ethics Committee for Animal Research, Göteborg, Sweden, approved the study protocol. Six Labrador dogs about 1-year old were used. The outline of the experiment is presented in Fig 1. During all surgical procedures, general aneasthesia was induced with intravenously injected Propofol (10 mg/ml, 0.6 ml/kg) and sustained with N_2O:O_2 (1:1.5–2) and Isoflurane using endotracheal intubation.

Surgery

All mandibular premolars and the three anterior premolars in both sides of the maxilla were extracted. After 3 months, mucoperiosteal flaps were elevated in both sides of the mandible. Four implants representing four different implant systems with different surface characteristics (implant group A, B, C, D; Table 1) were placed in each side of the mandible and in a randomized order. The implants in group A, B and D were provided with healing abutments, while healing caps were placed on the implants in group C. The flaps were adjusted and sutured around the neck of all implants. Radiographs were obtained after implant placement using a customized film holder (Hawe Super Bite, Hawe Neos Dental, Bioggio, Switzerland). The radiographs were analysed using an Olympus SZH10 stereo macroscope and digital images were obtained with a Leica DFC280 camera. Different landmarks were identified for each implant type. The abutment fixture junction was used as a reference point for implant categories A, B and D, while at the implants of type C the most apical point of the abutment/healing cap screw was identified. The vertical distance between the landmark and the marginal bone level was assessed at the mesial and distal aspects of each implant using the QWin software (Leica Qwin Standard V3.2.0, Leica Imaging Systems Ltd., Cambridge, UK). Double assessments with a 2-month interval were made by two examiners.

The sutures were removed after 2 weeks and a plaque control programme including daily cleaning of implants and teeth was initiated.

Experimental peri-implantitis

Three months after implant installation, experimental peri-implantitis was initiated (Baseline; Fig. 1). Thus, the oral hygiene procedures were abandoned, cotton ligatures were placed in a sub-marginal position around the neck portion of the implants (Lindhe et al. 1992) and a new set of radiographs was obtained. The ligatures were replaced at weeks 3, 6 and 9. At week 12, when about 40–50% of the supporting bone was lost, the ligatures were removed. In one side of the mandible, oral hygiene procedures including daily cleaning of implants using toothbrush was per-

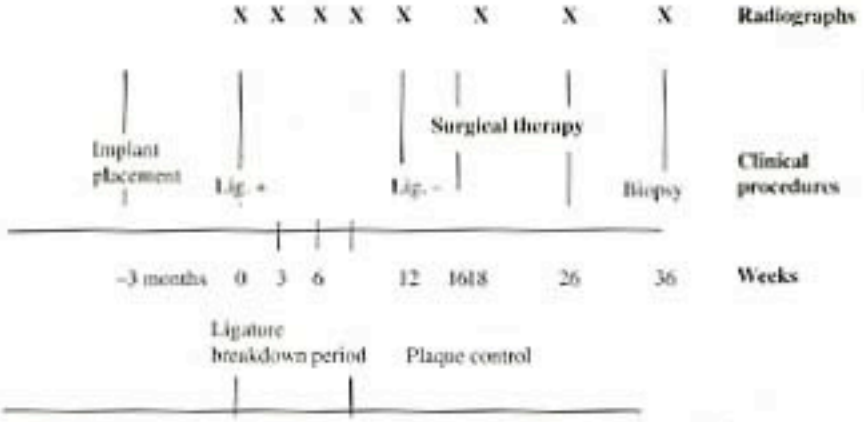

Fig 1. Outline of the experiment. Ligatures were placed at week 0 (Lig. +) and removed 12 weeks later (Lig. –). Radiographs (X) were obtained at weeks 0, 3, 6, 9, 12, 18, 26 and 36.

Table 1. Characteristics of implant types

Implant group	Surface	Name	Dimensions	Company
A	Turned	ICE Micro-Miniplant	3.25 × 10 mm	Biomet 3i, Palm Beach Gardens, FL, USA
B	TiOblast	MicroThread	3.5 × 11 mm	Astra Tech AB, Mölndal, Sweden
C	Sandblasted Large grit Acid Etched (SLA)	Standard plus implant NN	3.3 × 10 × mm	Straumann AG, Basel, Switzerland
D	TiUnite	MKIII Narrow Platform	3.3 × 10 mm	Nobel Biocare AB, Göteborg, Sweden

formed, while in the contra-lateral side no plaque control procedures were carried out. This study describes procedures and findings related to the implants receiving treatment, while results related to the untreated sites were reported previously (Albouy et al. 2008, 2009).

Surgical treatment

Four weeks later, surgical therapy was performed in the implant regions that received plaque control after ligature removal. No antibiotics were provided before, during or after the surgical treatment. Full-thickness flaps were raised on the buccal and lingual aspects of the implants and granulation tissue was removed by curettes (Fig. 2). Mechanical cleaning of the implant surfaces was performed using gauzes impregnated with a sterile saline solution. Calculus on implant surfaces was chipped off with curettes. Profuse saline irrigation of the implants and the adjacent tissues was made before adjustment and suturing of the flaps around the neck portion of the implants. The sutures were removed 10 days after surgery and oral hygiene procedures were re-instituted and maintained during the subsequent 5-month period of the experiment.

Radiographic and clinical examinations of the implant sites were performed during the pre-experimental period with plaque formation and repeated placement of ligatures (weeks 3, 6, 9 and 12) and at weeks 18, 26 and 36 during the post-surgical treatment period (Fig. 1). The clinical examination included an assessment of plaque and visible signs of soft tissue inflammation (redness and swelling).

At week 36, the dogs were euthanized with a lethal dose of Sodium-Pentothal® and perfused through the carotid arteries with a fixative consisting of a mixture of 5% glutaraldehyde and 4% formaldehyde buffered to a pH of 7.2 Karnovsky (1965). The mandibles were retrieved and stored in the fixative.

Histological preparation and analysis

Tissue blocks containing the implant and the surrounding soft and hard tissues were dissected using a diamond saw (Exakt®, Apparatebau GmbH, Norderstedt, Germany) and processed for ground sectioning according to methods described by Donath and Breuner (1982). Each block was cut in a bucco-lingual plane using a cutting–grinding unit (Exakt®, Apparatebau GmbH). From each implant site, two central sections (buccal–lingual plane) were obtained and further reduced to a final thickness of about 20 μm using a micro-grinding unit (Exakt®, Apparatebau GmbH). The remaining mesial and distal portions were remounted and cut in a perpendicular (mesio-distal) direction and two central sections were prepared from each unit. The sections were stained in toluidine blue or fibrin stain of Ladewig (Donath & Breuner 1982).

The histological examinations were performed in a Leica DM-RBE® microscope (Leica Heidelberg, Germany) equipped with an image system (Q-500 MC, Leica). The area of the infiltrated connective tissue (ICT) and the distance between the apical border of the ICT and the peri-implant bone were assessed.

Data analysis

Mean values for all variables were calculated for each implant in each animal. Differences were analysed using analysis of variance (ANOVA) and the Student–Newman–Keuls test. The null hypothesis was rejected at $p < 0.05$.

Results

Clinical findings

The findings related to the implants scheduled for the continuous plaque formation after ligature removal are reported elsewhere (Albouy et al. 2008, 2009). Among the implants scheduled for surgical therapy of peri-implantitis, one implant of type D was lost 2 months after implant placement. Another two implants of type D were lost following surgical therapy due to continuous loss of supporting bone. These implants were lost on weeks 26 and 33, respectively, i.e. at 10 and 18 weeks after surgical therapy.

Plaque formation during experimental peri-implantitis resulted in overt signs of inflammation in the peri-implant mucosa of all implants. At sites exposed to plaque control after ligature removal, plaque was virtually absent, while signs of inflammation in the peri-implant mucosae remained at the examination performed before surgical therapy. The plaque control exercised during the post-surgical treatment period resulted in an improvement of clinical signs of inflammation at implants of types A, B and C, while at implants of type D swelling and redness in the peri-implant mucosa persisted (Fig. 3).

Radiographic findings

Radiographs from the different types of implants at 2 weeks after surgical therapy (week 18) and at the final examination and biopsy (week 36) are presented in Fig. 4. The results from the radiographic measurements are reported in Table 2 and Fig. 5. During the preparatory period of ligature-induced breakdown, bone loss varied between 3.81 and 3.92 mm. For the two implants that were lost after surgical therapy and before the final examination, the radiographic bone loss at the following examination interval was judged to encompass the entire remaining intra-osseous portion of the implant.

Radiographic bone gain was observed between week 12 (start of plaque control) and week 36 (final examination) at implants of type A (2.22 ± 1.49 mm), type B (1.59 ± 1.51 mm) and type C (0.89 ± 1.50 mm). At implants of type D, however, additional bone loss of

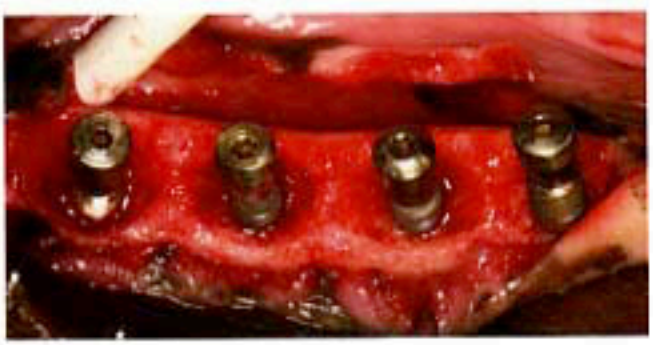

Fig. 2. Clinical photograph of implant sites exposed to surgical therapy. Note the osseous defects after the removal of the granulation tissue.

1.83 ± 2.37 mm occurred during the corresponding period. The difference between implant type D and implant type A was statistically significant.

Histologic findings

Ground sections produced from the different types of implants are presented in Fig. 6. The histological analysis revealed different treatment outcomes between implant types. Thus, in sections representing implant types A (turned surface) and B (TiOblast surface), no biofilm was detected on the implant surface. A thin barrier epithelium lined the marginal portion of the peri-implant mucosa next to the turned or TiOblast surface of these implants. Apical to the barrier epithelium, a zone of connective tissue that was rich in collagen but poor in vascular structures and cells faced the threaded portion of the implant. Small clusters of inflammatory cells were occasionally found in the marginal portion of the connective tissue around the implant types A and B.

The peri-implant mucosa formed around implants of type C (SLA surface) after surgical therapy contained inflammatory lesions of varying size. While few sites demonstrated clusters of inflammatory cells residing in the marginal portion of the connective tissue, the majority of specimens representing implant type C exhibited well-defined, moderately large inflammatory lesions that occupied a connective tissue compartment lateral to a barrier/pocket epithelium. The finding of inflammatory lesions in the soft tissues around implants of type C was consistently associated with the detection of a biofilm of the implant surface.

No signs of resolution following surgical treatment of peri-implantitis could be seen in sections representing the remaining sites of implant type D (TiUnite surface). Thus, the implant surface in this group was consistently covered by calculus and a biofilm and the adjacent mucosa harboured a large inflammatory cell infiltrate that extended from the margin of the soft tissue to the peri-implant bone. A pocket epithelium separated the inflammatory lesion from the biofilm in the marginal part of the mucosa, while in the apical portion the inflammatory cell infiltrate was in direct contact with the biofilm.

The results from the histometric measurements revealed that the size of the remaining inflammatory cell infiltrate (ICT) in the peri-implant soft tissues varied between 0.30 ± 0.45 and 0.49 ± 0.65 mm^2 in sites representing implant types A and B. The corresponding values for the specimens prepared from implant types C and D were considerably larger and amounted to 1.89 ± 2.33 (implant C) and 3.01 ± 1.34 mm^2 (implant D), respectively. The distance between the api-

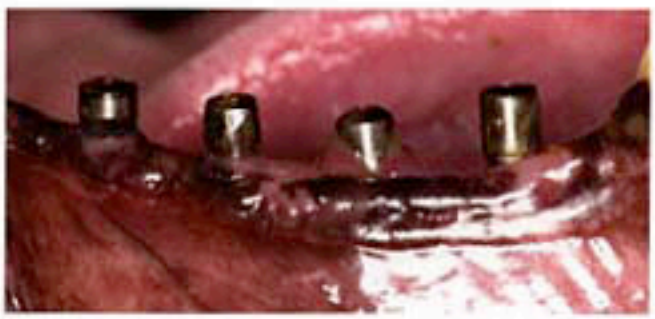

Fig. 5. Clinical photograph of the different implant types at the final examination (week 36). From left: implants B–D and A. Note the swelling around implant type D.

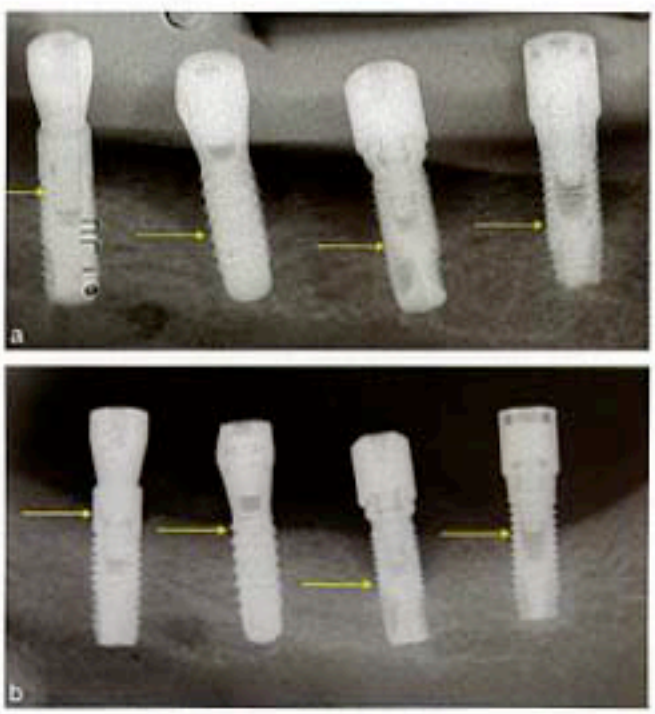

Fig. 4. Radiographs of the different implant types (a) at 2 weeks following surgical therapy (week 18) and (b) at the final examination (week 36) illustrated in Fig. 5. From left: implants B–D and A. Note the different bone level around implant type D at the final examination (b).

Table 2. Bone level alterations (mm) during the 24-week period following ligature removal/start of plaque control (12–36 weeks)

	Implant A	Implant B	Implant C	Implant D
Ligature removal/plaque control – biopsy (weeks 12–36)	2.22 (1.49)	1.59 (1.51)	0.89 (1.50)	−1.58 (2.61)*

Negative values indicate bone loss. Mean values and standard deviation (SD).
*$p < 0.05$ between implants D and A.

cal border of the ICT and the peri-implant bone was 0.99 ± 0.11 and 0.87 ± 0.51 mm at implant types A and B. In sites representing implant type C and D, the distances were shorter and measured 0.52 ± 0.71 and 0.14 ± 0.16 mm, respectively.

Discussion

This experimental study evaluated the effect of surgical therapy of experimental peri-implantitis without systemic antibiotics at different types of implants. Treatment resulted in improved clinical conditions at implants with turned, TiOblast and SLA surfaces, while at implants with a TiUnite surface, swelling and redness in the peri-implant mucosa persisted. In addition, two of the TiUnite implants were lost at 10 and 18 weeks after surgery, respectively. The radiographic analysis revealed that bone gain occurred at implants with turned, TiOblast and SLA surfaces following treatment, while at TiUnite implants, additional bone loss was found. The results from the histological analysis disclosed that resolution of peri-implantitis was achieved in tissues surrounding implants with turned and TiOblast surfaces. Remaining inflammatory lesions were found in SLA sites. No signs of resolution were seen in sections representing TiUnite implants. It is suggested that (i) resolution of peri-implantitis following treatment without systemic or local antimicrobial therapy is possible and that (ii) the outcome of therapy is influenced by implant surface characteristics.

The combined treatment strategy of systemic antibiotics and local, surgical therapy was demonstrated to be successful in resolving inflammation in several studies on experimental peri-implantitis.

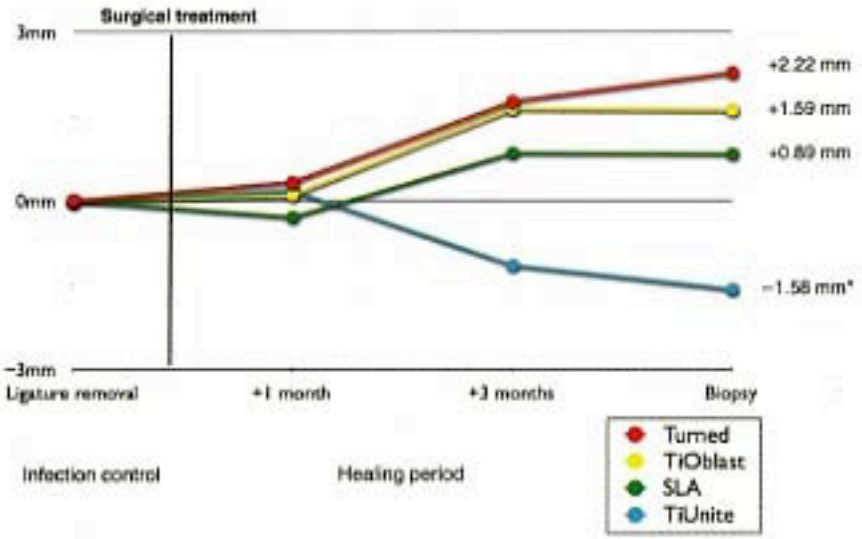

Fig. 5. Bone-level alterations during the period between ligature removal (12 weeks) and final examination (36 weeks).

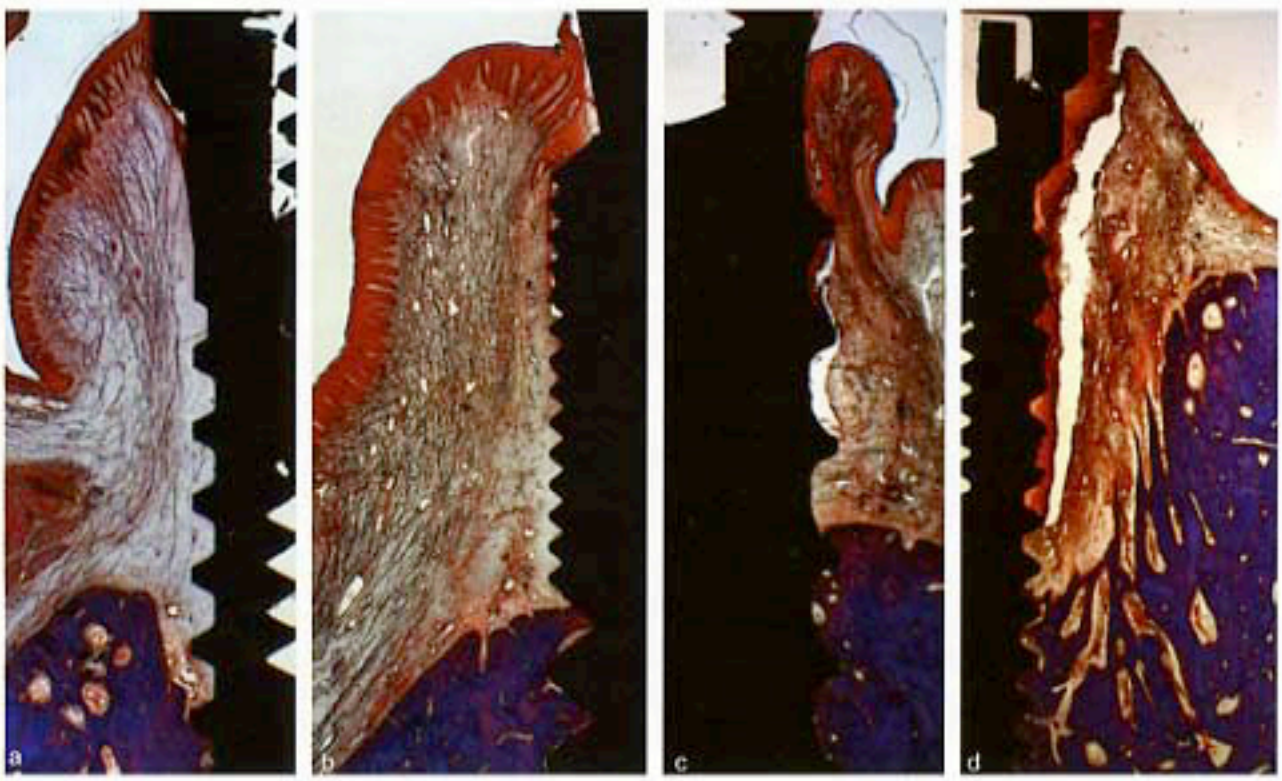

Fig. 6. Ground sections representing implant types A–D. Note the remaining inflammatory cell infiltrate in the tissues around implant type C (SLA surface) (c) and the large inflammatory lesion residing in the tissues around implant type D (TiUnite surface) (d). Fibrin stain of Ladewig.

The type of antibiotics used and cleansing methods applied during surgical treatment, however, varied between studies. Thus, Ericsson et al. (1996) performed surgical therapy of experimentally induced peri-implantitis in Labrador dogs. While all animals received systemic antibiotics with a combination of metronidazole and amoxicillin concomitant with the surgical procedure, local therapy including cleaning of implants with delmopinol was applied only to implants in one side of the mandible. It was reported that resolution of peri-implantitis lesions occurred around the implants receiving local therapy, whereas no signs of resolution were observed at the untreated implants. Ericsson et al. (1996) concluded that systemic antibiotic therapy alone is insufficient in resolving peri-implantitis lesions. Persson et al. (1999, 2001a,b) in studies on experimental peri-implantitis in Beagle dogs also used systemic antibiotics with a combination of metronidazole and amoxicillin. During surgical treatment of peri-implantitis, the implants were cleaned using cotton pellets soaked in saline. The histological analysis revealed that the inflammatory lesions had resolved and new bone formation had occurred in the previous defects. The combination of systemic antibiotics described above was also used by Schou et al. (2003) in a study on treatment of experimental peri-implantitis in monkeys. Local therapy was performed using different implant surface decontamination procedures including saline, chlorhexidine, citric acid and an air-powder abrasive unit. While no differences in treatment outcome were found between methods, Schou et al. (2003) suggested that the most simple technique should be applied. Wetzel et al. (1999) who evaluated treatment of experimental peri-implantitis in Beagle dogs applied a systemic antibiotic regimen consisting of metronidazole alone, while cleaning of implant surfaces was performed using chlorhexidine. In an experimental study in dogs, Schwarz et al. (2006) analysed the outcome of treatment of peri-implantitis using different implant surface decontamination procedures including metronidazole gel.

Taken together, the finding in the present study that resolution of peri-implantitis lesions was possible without systemic antibiotics brings new information and adds to our understanding of the importance of local therapy including biofilm removal at the affected implants in treatment of peri-implantitis.

In the present study, it was demonstrated that the implant surface characteristics influenced the outcome of treatment of experimental peri-implantitis. Not only the analysis of marginal bone-level changes after surgical therapy but also the evaluation of the histological sections revealed different results between the implant types. While further loss of bone support was prevented in implant type A, B and C, resolution of the peri-implantitis lesion was accomplished in sites representing implant types A and B. Previous reports on treatment of experimental peri-implantitis have included a number of different implant types. One of the most common implant types studied was the Brånemark implant with a turned surface. In the experimental studies described above (Ericsson et al. 1996, Persson et al. 1999, 2001a), resolution of the peri-implantitis lesion was observed in tissues surrounding Brånemark implants with a turned surface following surgical treatment. This finding is in agreement with results presented in the present study. Implant type A had a turned surface and all such sites demonstrated radiographic bone gain and resolution of the peri-implantitis lesion. While no previous documentation appears to be available on treatment of experimental peri-implantitis on implants with a TiOblast surface, the findings in the present study reveal that outcomes following treatment of peri-implantitis at implant type B (TiOblast surface) were similar to those obtained at implant type A (turned surface). Results from treatment of experimental peri-implantitis at implants with a SLA surface, however, were reported previously. Wetzel et al. (1999) analysed the outcome of treatment of experimental peri-implantitis around implants with SLA, TPS and smooth surfaces in Beagle dogs. While the degree of resolution of the inflammatory lesion in the peri-implant soft tissues was not addressed, it was reported that bone fill occurred in associated osseous defects around all types of implants following therapy. Persson et al. (2001a) studied the treatment of experimental peri-implantitis at implants with either an SLA surface or a smooth, polished surface. Resolution of peri-implantitis lesions was observed for all sites of both implant types. The results presented in the study by Persson et al. (2001a) are not entirely in agreement with data presented in the current study. Although no further loss of bone support was demonstrated, a remaining inflammatory lesion of varying size was observed in the peri-implant soft tissues around implants type C (SLA surface). The reason for the different results in terms of resolution of peri-implantitis lesion around implants with an SLA surface between the study by Persson et al. (2001a) and the present experiment is not understood. It should be pointed out, however, that systemic antibiotics were used as an adjunct to the local therapy in the study by Persson et al. (2001a), whereas no antibiotics were provided to the animals in the current trial.

The treatment outcome at implants type D (TiUnite surface) in the present study was entirely different from that observed at the other implant types. Two implants of type D were lost after surgical treatment of peri-implantitis and radiographic and histological analyses of remaining sites revealed that continuous bone loss occurred and that large inflammatory lesions persisted in the peri-implant soft tissues. While no previous documentation of treatment of experimental peri-implantitis appears to be available for implants with a TiUnite surface, the results presented in the present experiment indicate that implant surface characteristics may have a decisive influence on the outcome of treatment of peri-implantitis. It should be realized, however, that the results should be evaluated and interpreted from the prerequisites of the study and that the outcome from other or additional treatment protocols cannot be predicted. The mechanical biofilm removal procedures used in the present study were proven to be effective for implant types A, B and to some extent implant C, while at implant type D the treatment measures were insufficient.

The main objective in the present study was to evaluate resolution of peri-implantitis lesions following surgical treatment. The experimental outline and methods used for the analysis were designed accordingly. The radiographic bone gain observed for three of the implant types cannot therefore be evaluated in terms of the degree of the so-called re-osseointegration. Experimental studies aiming at investigating the potential re-osseointegration following treatment of peri-implantitis require assessments of important landmarks

that indicate the level of bone support at the time of treatment. Such landmarks were introduced in previous experiments by Wetzel et al. (1999) and Persson et al. (1999, 2001a) and included fluorochrome markers in bone tissue and titanium rings placed at the most apical position of the osseous defect. No attempts were made to assess re-osseointegration in the present study.

In summary, the results from the experimental study revealed that resolution of peri-implantitis following treatment without systemic or local antimicrobial therapy is possible and that the outcome of therapy is influenced by implant surface characteristics.

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Clinical Relevance

Scientific rationale for the study: It is not known if implant surface characteristics influence treatment outcome or if the adjunctive use of systemic antibiotics is required in surgical treatment of peri-implantitis.
Principal findings: Radiographic bone gain occurred at implants with turned, TiOblast and SLA surfaces, while at TiUnite implants additional bone loss was found after surgical treatment. Resolution of peri-implantitis was achieved in tissues surrounding implants with turned and TiOblast surfaces. Remaining inflammatory lesions were found in SLA sites. No signs of resolution were found in sections representing TiUnite implants.
Practical implications: The finding that resolution of peri-implantitis following treatment without systemic or local antimicrobial therapy is possible provides an ethical base to perform controlled clinical studies. A systemic antibiotic regimen may not always be required in the treatment of peri-implantitis. The finding that the outcome of treatment is influenced by implant surface characteristics points to the importance of risk assessments in treatment planning and the need to further investigate the problem related to decontamination of implant surfaces.