Sinus Floor Elevation with Biphasic Calcium Phosphate or Deproteinized Bovine Bone Mineral: Clinical and Histomorphometric Outcomes of a Randomized Controlled Clinical Trial

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Abstract: PURPOSE To clinically and histomorphometrically compare a biphasic calcium phosphate (BCP) and deproteinized bovine bone mineral (DBBM) for sinus floor elevation. MATERIALS AND METHODS Sinus floor elevation procedures (lateral window) were performed randomly applying either BCP (test) or DBBM (control). At 6 months, bone biopsy specimens were harvested and dental implants were placed. The proportions of new bone, residual grafting material, and nonmineralized soft tissue were calculated. Four months after implant placement, the prosthetic reconstructions were inserted and the implant survival was assessed. RESULTS Fifty-one patients were treated; 25 were randomly allocated to the BCP group and 26 to the DBBM group. After 6 months in 50 patients, bone biopsy specimens could be harvested, and a total of 121 implants could be placed subsequently. The histomorphometric analysis revealed a comparable percentage of new bone in both groups (BCP 35.9%, DBBM 35.4%; P > .05). The remaining grafting material was significantly lower with BCP (25.3%) compared with DBBM (45.9%; P < .001). Nonmineralized tissue was significantly higher for the BCP group (38.1%) compared with the DBBM group (18.2%; P < .001). The implant survival rate at loading was assessed at the level of the patients (96.0% for BCP and 88.8% for DBBM; P > .05) and at the level of the implants (96.9% for BCP and 94.7% for DBBM; P > .05). CONCLUSION Grafting with DBBM or BCP showed similar percentages of new bone 6 months after sinus floor elevation. Implant survival presented no significant difference until loading.

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Sinus floor elevation with biphasic calcium phosphate or deproteinized bovine bone mineral: clinical and histomorphometric outcomes of a randomized controlled clinical trial.

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ABSTRACT

Objectives: to clinically and histomorphometrically compare a biphasic calcium phosphate (BCP) and deproteinized bovine bone mineral (DBBM) for sinus floor elevation.

Materials and methods: Sinus floor elevation procedures (lateral window) were performed randomly applying either BCP (test) or DBBM (control). At 6 months, bone biopsies were harvested and dental implants placed. The proportions of new bone, residual grafting material and non-mineralized soft tissue were calculated. Four months after implant placement the prosthetic reconstructions have been inserted and the implant survival was assessed.

Results: Fifty-one patients were treated: 25 were randomly allocated to the BCP and 26 to the DBBM group. After 6 months in 50 patients bone biopsies could be harvested and a total of 121 implants could be placed subsequently. The histomorphometric analysis revealed a comparable percentage of new bone in both groups (BCP 35.9%, DBBM 35.4%; $p>0.05$). Remaining grafting material was significantly lower with BCP (25.3%) compared to DBBM (45.9%; $p<0.001$). Non-mineralized tissue was significantly higher for BCP (38.1%) compared to the DBBM group (18.2%; $p<0.001$). The implant survival rate at loading was assessed at the level of the patients (96.0% for BCP and 88.8% for DBBM; $p>0.05$) and at the level of the implants (96.9% for BCP and 94.7% for DBBM; $p>0.05$)).

Conclusions: Grafting with DBBM or BCP showed similar percentage of new bone, 6 months after sinus floor elevation. Implant survival presented no significant difference until loading.

INTRODUCTION

Maxillary sinus floor elevation to allow the placement of endosseous implants has become a standard surgical procedure with reliable long-term results, both in clinical (1-5) and experimental studies (6-8).

Various grafting materials were proposed for this kind of bone augmentation procedure.

Autogenous bone grafts (autografts) have been recommended for the purpose of acting as an osteoinductive and osteoconductive scaffold (9). In order to avoid the necessity for a second site to harvest the autogenous bone and to reduce post-surgical morbidity, bone graft substitutes were later introduced. These materials can be classified as follows: allografts (from same species, human, but different individual), xenografts (from different species, usually from bovine origin) and alloplastic materials (synthetic origin)(4).

Deproteinized bovine bone mineral (DBBM) is one of the most commonly used and most documented bone substitute materials in dental surgery (4, 10). The combination of DBBM and resorbable collagen membranes have been found to be effective for bone augmentation in situations where dental implants are placed (11-13). DBBM materials have also been intensely investigated for sinus floor elevation, revealing excellent long-term results (3).

However, despite his good osteoconductive proprieties, DBBM doesn't present an osteoinductive potential(14).

As alternative, alloplastic materials can be used (4, 15). Biphasic calcium phosphate (BCP), consisting of hydroxyapatite (HA) and β-tricalcium phosphate (β-TCP), showed in a 60:40 ratio similar bone changes to standard graft material (15-17). At the same time, these allografts revealed a higher resorption rate during new bone formation(15, 16). Nevertheless, the subsequently placed implants present excellent survival and success rates (18).

A new second generation of BCP has been developed with a 10:90 HA/TCP ratio and controlled microporosity (mean micropore diameter 1 μm), which may have an important influence on the osteoconductivity of calcium phosphate ceramics (19, 20). This material has demonstrated high

biocompatibility and osteoconductivity (21, 22), as well as an osteoinductive potential(14, 23). Despite the promising preclinical studies, the available data on humans is still very little (24).

The aim of the present trial was to clinically and histomorphometrically analyze a second-generation biphasic calcium phosphate and a deproteinized bovine bone mineral 6 months after a sinus grafting procedure.

MATERIALS AND METHODS

Study Design

This study was designed as a multicenter, randomized, controlled clinical study conducted at the clinic of reconstructive dentistry, University of Zurich, Zurich, Switzerland and in a private practice set up, Zentrum für Implantologie, Parodontologie und 3D- Diagnostik, Konstanz, Germany.

The clinical study protocol, all materials and procedures were approved by the respective local ethical committees (KEK-ZH-Nr. 2014-0478; F-2013-086-z). Prior to the start of the investigation, informed consent was obtained from all patients. The study was independently, externally monitored to ensure consistency and accuracy.

Subjects

Generally healthy female and male subjects, who were at least 18 years of age, with a need for either unilateral or bilateral sinus floor elevation procedures to place one or more dental implants (staged approach), were recruited.

The following further inclusion criteria were applied:

- 2 to 4 mm of residual bone height, measured on a recent available radiograph (panoramic radiograph or cone beam CT scan, <12 months old)
- Adequate oral hygiene (full mouth plaque index <25%)(25)
- Adequate inflammation control (full mouth bleeding on probing ≤25%)(26)

The defined exclusion criteria were:

- General contraindications for bone grafting and oral surgical procedures
- Local contraindications (e.g. sinus anomalies)
- Untreated periodontitis

- Participation in other investigational drug or device studies 30 days prior or during the conduct of the present trial.
- Antibiotic therapy within 28 days prior sinus floor elevation
- History of drug or alcohol abuse
- Smokers (> 10 cigarettes per day)
- Current pregnancy or breastfeeding

As a secondary exclusion criterion during the sinus floor elevation:

- Defects of the Schneiderian Membrane

Clinical protocol

Sinus floor elevation procedure

Preoperatively, patients received antibiotics and analgesics/antiphlogistics depending on the standard of care of the clinical study center. Furthermore, they rinsed with 0.2% chlorhexidine solution for 1 minute. The surgical procedure was performed using the lateral window technique (1), under local anesthesia.

First, a mucoperiosteal flap was elevated and a bony window was created on the lateral wall of the maxillary sinus using rotary as well as piezoelectric instruments. The Schneiderian membrane was then carefully elevated. In case of a membrane defect, the patient was excluded from further participation in the study (secondary exclusion criterion).

At this point in time, a sealed randomization envelope was opened to allocate the sinus to either one of the two following grafting materials:

- test group: biphasic calcium phosphate (BCP), HA/TCP 10:90 (Straumann® VivOss™, Institut Straumann AG, Basel, Switzerland)

- control group: deproteinized bovine bone mineral (DBBM) (Bio-Oss®, Geistlich AG, Wollhusen, Switzerland)

In cases with augmentation of both sinuses, one sinus was randomized as study sinus. If the sinus was randomized into the test group, the opposite sinus was augmented with DBBM and vice versa.

The selected bone substitute material was used for grafting the obtained space beneath the elevated mucosa. In both treatment groups**,** the bony window was then covered with a resorbable collagen membrane (Bio-Gide®, Geistlich AG, Wollhusen, Switzerland), followed by a tension-free flap closure using non-resorbable sutures.

Patients were instructed to rinse twice a day until suture removal with 0.2% chlorhexidine solution and received antibiotics (750 mg Amoxicillin Sandoz® or 300-600 mg Clindamycin Sandoz®; 3 times a day, for 6 days). Analgesics were prescribed (50 mg Voltaren®, max. 150 mg a day, or 400 mg Ibuprofen®, max. 1200 mg a day).

After a healing time of 1-2 weeks, sutures were removed, and the wound healing assessed (“very good”, “good”, “normal” or “impaired”).

Implant placement

After a healing time of 6 months (±7 days), implant placement was performed.

Prior to surgery, patients rinsed with 0.2% chlorhexidine solution for 1 minute. Under local anesthesia a mucoperiosteal flap was elevated. The implant bed preparation was performed first by means of a trephine burr with an inner diameter of 2.8 mm, in such a way that a bone biopsy could be harvested at the same time. Dental implant placement (Straumann Dental Implant System, SLActive®, Institut Straumann AG, Basel, Switzerland) was then completed according to the standard procedure of the clinic. The flap was closed tension-free with non-resorbable sutures. Analgesics (Voltaren®, max. 150 mg a day or Ibuprofen®, max. 1200 mg a day) and a 0.2% chlorhexidine solution (twice a day, until suture removal) were prescribed. After a healing time of 1-2 weeks, the sutures were removed and the wound healing evaluated ("very good", "good", "normal" or "impaired").

Four months after implant placement, the implants were initially loaded and the survival as well as the success rates (according to Buser (27)) were evaluated.

Histological procedure and histomorphometry

All procedures and analyses were executed by the same experienced histologist, which was blinded regarding the randomization group, at least until microscopy and analysis (recognition of the grafting material).

Samples that had been harvested with a trephine drill were fixed in buffered 4% formaldehyde solution. After dehydration in a series of graded ethanol solutions, the tissues were finally embedded in a methyl methacrylate solution (sigma-aldrich M55909-1L). With a diamond band saw (Exakt Apparatebau, Norderstedt, Germany) the blocks were cut into 200-µm-thick sections, which were then ground and polished to a thickness of 60-80 µm (Exakt Apparatebau) and stained with toluidine blue.

For histomorphometrical analysis, all sections were photographed with Microscope Leica DM6000 B (Leica Mikrosysteme, Wetzlar, Germany) and a digital camera (Leica DFC 450; Leica Mikrosysteme, Wetzlar, Germany). The digital images were first processed with Photoshop CS6 (Adobe Systems Incorporated, San Jose, USA) and successively evaluated with an analysis software (Leica Application Suite, V4.3, Leica Mikrosysteme, Wetzlar, Germany).

The area fraction of three different tissue components (bone, grafting material and connective tissue) was calculated in a region of interest, which had a total surface of 3.2 mm^2 and was located in the grafted portion of the samples.

Outcomes Measures

Histomorphometry

The primary outcome was the calculated ratio of new bone to remaining grafting material out of the histomorphometrical analysis.

Implant survival and implant success

Secondary outcome measures were implant survival and success, assessed at implant loading (4 months after implant placement).

Implant success was defined using the following criteria (according to Buser (27)) apply:

- Absence of persisting subjective discomfort such as pain, foreign body perception and or dysaesthesia (painful sensation)
- Absence of a recurrent peri-implant infection with suppuration (where an infection is termed recurrent if it is observed at two or more follow-up visits after treatment with systemic antibiotics)
- Absence of implant mobility on manual palpation

- Absence of any continuous peri-implant radiolucency

Statistical analysis

Sample size calculations were performed for a clinically relevant difference between test and comparator across a given range of standard deviations, with the two-sided unpaired t-test under a significance level of 5% and with a power of 80%. Based on pre-clinical data a difference of the means of the ratio of new bone to remaining grafting material of approximately 0.4 was expected in favor of the test treatment compared to the control. In order to show superiority, considering a common standard deviation of 0.5, 25 subjects per group were considered necessary to confirm a clinically relevant difference statistically.

RESULTS

In total 56 (66% female and 34% male) patients with a mean age of 59.3 (±10.8) years were enrolled in the two study centers (Zurich: 27; Konstanz: 29). One patient was excluded because he did not fulfil the inclusion criteria. Four further patients were excluded during the sinus floor elevation procedure, because they presented defects of the Schneiderian membrane (3 cases) or missing crestal bone (0 mm residual vertical height, 1 case). This resulted in 51 patients receiving a grafting material in the study maxillary sinus: 25 BCP (test group) and 26 DBBM (control). In addition, 15 patients were qualified for bilateral treatment and the contralateral sinus was grafted with the opposite material. At the point in time of the sinus floor elevation, the mean residual bone height was 3.7±1.4 mm in the BCP group and 3.9±1.8 in the DBBM group. The difference between the groups was not statistically significant (p=0.64).

During the healing period one patient in the control group was excluded from further participation in the study because he refused to pay for the treatment.

No difference in the wound healing could be observed between test and control (p=0.94).

Six months after sinus floor elevation (6.1±0.4 months overall; 6.2±0.4 BCP group, 6.1±0.4 DBBM group), 50 patients received a total of 121 primary stable implants. During implant bed preparation, 87 biopsies were harvested (Figure 2-4), of which 62 could be analyzed histomorphometrically (29 in the test group and 33 in the control group).

Histomorphometry

The ratio of “new bone” (%) to “bone grafting material” (%) was calculated (Table 1). In a non-parametric significance test, the median ratio in the BCP group (1.38) was significantly higher compared to the median ratio in the DBBM group (0.67; $p<0.001$). Mean values instead, due to the large standard deviation of BCP, were not significantly different (DBBM: 0.86±0.45 vs. BCP: 3.48±6.76; $p=0.064$).

The mean proportions of newly formed bone, as well as bone graft materials and non-mineralized tissue are reported in Figure 1. “New bone” was comparable in both treatment groups (BCP 35.9% vs. DBBM 35.4%; $p=0.845$). Remaining “bone grafting material” was significantly lower with BCP (25.3%) compared to DBBM (45.9%; $p<0.001$). The mean proportion of non-mineralized tissue in the biopsies was instead significantly higher for BCP (38.1%) compared to the DBBM group (18.2%; $p<0.001$).

Subgroup “bilateral”

Bilateral sinus floor elevation was performed in 15 patients (30%). In these cases, a “split-mouth design” was applied. The mean proportion of “new bone” was not statistically significantly different between the two groups (BCP 32.6%, DBBM 28.1%; $p=0.123$). Remaining “bone grafting material” was significantly lower with BCP (24.5%) compared to DBBM (42.6%; $p=0.001$), while “non-mineralized tissue” was instead significantly higher for the BCP group (42.2%), compared to DBBM (28.9%; $p=0.003$).

The ratio of new bone compared to bone grafting material for this subgroup is reported in Table 2. The outcome is comparable to that of the entire cohort: the median ratio in the BCP group (1.37) is significantly higher compared to the median ratio in the DBBM group (0.59) ($p=0.017$). The parametric comparison of means failed to show statistical significance ($p=0.106$).

Implant survival / success

After implant surgery, no difference in wound healing could be observed between groups (p=0.92).

Implants were loaded after 5.6±2.9 months (5.8±3.2 BCP group, 5.4±2.6 DBBM group) of healing. Out of 121 placed implants in 50 patients, 5 implants in 4 patients were lost before loading (early failures, 3.2±1.5 months after placement). On patient level, the overall implant survival rate was 92.3% (CI: 81.3-97.5). One patient receiving 2 implants into a test site lost both implants, 3 weeks after insertion. In two cases, two implants were inserted into control sites; both patients lost 1 of these implants 3 months afterwards. In another control case an implant was lost 4 months after placement; the implant was the only one inserted (in the same region). The resulting survival rate for the test group was 96.0% (CI: 78.9-99.9) and for the control group 88.8% (CI: 69.2-96.7).

On implant base, the overall implant survival rate was 95.9% (CI: 90.4-98.5), 96.9% (CI: 88.7-99.8) for the test group and 94.7% (CI: 85.1-98.8) for the control group. No significant difference could be observed between the treatment groups, either patient based, nor implant based.

At loading, all implants (100%) were considered successful (according to Buser (27)).

DISCUSSION

The present randomized controlled clinical study showed a) no statistically significant difference in new bone formation between test and control groups 6 month after sinus floor elevation and b) similar survival and success of implants placed in primary augmented sinuses after a healing period of 4 months.

The significant difference of BCP over DBBM in the ratios of “new bone” to “bone grafting material” was mainly due to the higher proportion of “non-mineralized tissue” (BCP 38.1% vs. DBBM 18.2%) and lower percentage of remaining “bone grafting material” (BCP 25.3% vs. DBBM 45.9%) in the BCP group, whereas newly formed bone was similar in both groups (BCP 35.9% vs. DBBM 35.4%). This observation is consistent with the results of another randomized clinical trial comparing BCP to DBBM (15). In that particular study both groups showed similar amounts of newly formed bone 6 months after augmentation (BCP 21.6% vs. DBBM 19.8%), but significantly less remaining bone grafting material in the BCP group (BCP 26.6% vs. DBBM 37.7%) and more soft tissue components (BCP 46.47% vs. DBBM 40.4%). Another randomized, controlled investigation performed a histomorphometric comparison of vital bone formation, 6 to 8 months after bilateral sinus grafting with either BCP or DBBM (17). Similar to the present study, biopsies showed no significant difference in the average vital bone content (BCP 28.4% vs. DBBM 22.3%). The residual graft particles were in average 28.4% in the BCP and 26.0% in the DBBM biopsies, showing no statistically significant difference as well. Moreover, two recent systematic reviews, confirmed the non-significant difference in the amount of newly formed bone after sinus floor elevation with synthetic bone substitutes versus xenografts **(28, 29)**.

As mentioned above, in the present investigation, BCP showed a lower percentage of remaining "bone grafting material" compared to DBBM. Similar results were presented also by other clinical trials (15, 16). This could be explained with a higher resorption resistance of bovine bone substitutes, which has already been described in the literature (30-33). Consequently, the remaining integrated DBBM particles may contribute to the long-term volume stability of the augmented maxillary sinus. However, the clinical relevance of the remaining bone graft material is still questionable, since systematic reviews could not show any difference in implant treatment outcomes (4, 28).

In this clinical investigation a new BCP was used with an HA/TCP ratio of 10:90. This composite grafting material is similar to BCP 60:40 regarding the particle size, which varies between 500 and 1000 µm for both compositions (in comparison DBBM: 250–1000 µm). However, the materials are different, not only in the HA/TCP ratio, but also in the preparation processes. In particular, variation of the sintering temperature results in different microstructures(34, 35). Therefore, the interpretation and the comparison of the results with other clinical studies, investigating BCP with other compositions, is limited. Moreover, the present study reports the first clinical data on this grafting material in dental literature. Nevertheless, there are preclinical studies available which studied BCP in different animal models (14, 21, 22, 34). One preclinical study in the rabbit calvaria showed a higher percentage of mineralized new bone in defects filled with BCP 10:90 and BCP 60:40, compared to DBBM at 3 months. The difference was not statistically significant, and all grafting materials were comparable at 6 months. Both BCP compositions showed a higher percentage of non-mineralized tissue compared to DBBM after a healing of 6 months (22). This finding is similar to the results of the present investigation (BCP 10:90), as well as to the results of the above mentioned clinical sinus study comparing BCP 60:40 to DBBM (15).

In the current study, implants placed in augmented bone 6 months after sinus floor elevation showed survival rates of 96.0% in the BCP and 88.8% in the DBBM group. Three patients in the control group and only one in the test group experienced implant losses, explaining the apparently large difference in patient-based survival rates, which per contra was not statistically significant. The implant-based survival rates (96.9% BCP; 94.7% DBBM) appear to be in line with the ones reported in the literature. A systematic review presented a survival rate of implants placed in augmented maxillary sinuses of 96.5% after 3 years (implant based) (3). The use of a membrane over the lateral window seemed to increase the percentage to 98.3%. The failure rate during the healing phase (early failures) was 1.1% for rough surface implants. This parameter could offer a better comparison with the present study, since the survival rates are related to different observation periods. With an overall early failure rate of 4.1%, the current investigation reports an increased number of implant losses. The reason for this is difficult to elucidate. It has to be noted that the heterogeneity of protocols (one- vs two- staged), grafting materials, residual vertical bone in the systematic review may reduce the comparability with the present study. Patient related and local factors may influence the outcome. In particular, two out of the four patients, which experienced implant losses, had provisional removable partial dentures during the healing phase. From a practical clinical perspective premature loading (micromovements) may compromise the osseointegration. One may also speculate, that the lack of autogenous bone mixed to the bone substitute material may decelerate the new bone formation and, in some cases, possibly compromise the osseointegration. In order to support this speculation an animal study including 40 mini pigs, showed a significantly lower bone-to-implant contact 12 weeks after sinus floor elevation using DBBM alone, compared to autogenous bone in combination with DBBM (36). In contrast, a systematic review including only clinical studies comparing grafting with “DBBM alone” and “DBBM mixed with autogenous bone (AB), revealed no statistically significant difference in the 1-year survival rates of the implants (DBBM 96% vs. DBBM/AB 94%). Further, the addition of about

20% of autogenous bone to DBBM (80%) seemed not to improve new bone formation and bone-to-implant contact (37).

The short follow-up until implant loading can be considered the main limitation of the present investigation. The main focus and primary outcome of the study were however, linked to human bone biopsies and the histomorphometric analysis. Early failures during the healing phase after implant placement were also detected. However, no data has been collected on the stability of marginal bone as well as implant survival/success after loading. Considering the available data in the literature, comparable marginal bone levels as well as implant survival should be expected for both groups over time (28, 38). Nevertheless, the clinical significance of the different soft tissue and remaining grafting material proportions on the long-term outcome cannot be evaluated.

CONCLUSIONS

Sinus floor elevation with DBBM or BCP (10:90) for staged implant therapy showed similar percentage of new bone, 6 months after sinus floor elevation. No significant difference in implant survival and implant success until loading could be observed. Therefore, at least for an observation period of 6 months, BCP represent a valid synthetic alternative to DBBM for sinus floor elevation.

The impact of different proportions of soft tissue and residual bone substitute material on the long-term implant outcome remains unknown.

Acknowledgements and conflict of interest

The study was sponsored by Institut Straumann AG, Basel, Switzerland. The sponsor was not involved in the medical treatment, data management and analysis and monitoring of the study.

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References

1. Boyne PJ, James RA. Grafting of the maxillary sinus floor with autogenous marrow and bone. J Oral Surg 1980;38:613-616.

2. Valentini P, Abensur DJ. Maxillary sinus grafting with anorganic bovine bone: a clinical report of long-term results. Int J Oral Maxillofac Implants 2003;18:556-560.

3. Pjetursson BE, Tan WC, Zwahlen M, Lang NP. A systematic review of the success of sinus floor elevation and survival of implants inserted in combination with sinus floor elevation. J Clin Periodontol 2008;35:216-240.

4. Jensen SS, Terheyden H. Bone augmentation procedures in localized defects in the alveolar ridge: clinical results with different bone grafts and bone-substitute materials. Int J Oral Maxillofac Implants 2009;24 Suppl:218-236.

5. Del Fabbro M, Testori T, Francetti L, Weinstein R. Systematic review of survival rates for implants placed in the grafted maxillary sinus. Int J Periodontics Restorative Dent 2004;24:565-577.

6. Hurzeler MB, Quinones CR, Kirsch A, Schupbach P, Krausse A, Strub JR, et al. Maxillary sinus augmentation using different grafting materials and dental implants in monkeys. Part III. Evaluation of autogenous bone combined with porous hydroxyapatite. Clin Oral Implants Res 1997;8:401-411.

7. Quinones CR, Hurzeler MB, Schupbach P, Kirsch A, Blum P, Caffesse RG, et al. Maxillary sinus augmentation using different grafting materials and osseointegrated

dental implants in monkeys. Part II. Evaluation of porous hydroxyapatite as a grafting material. Clin Oral Implants Res 1997;8:487-496.

8. Hurzeler MB, Quinones CR, Kirsch A, Gloker C, Schupbach P, Strub JR, et al. Maxillary sinus augmentation using different grafting materials and dental implants in monkeys. Part I. Evaluation of anorganic bovine-derived bone matrix. Clin Oral Implants Res 1997;8:476-486.

9. Block MS, Kent JN. Sinus augmentation for dental implants: the use of autogenous bone. J Oral Maxillofac Surg 1997;55:1281-1286.

10. Kolk A, Handschel J, Drescher W, Rothamel D, Kloss F, Blessmann M, et al. Current trends and future perspectives of bone substitute materials - from space holders to innovative biomaterials. J Craniomaxillofac Surg 2012;40:706-718.

11. Zitzmann NU, Naef R, Scharer P. Resorbable versus nonresorbable membranes in combination with Bio-Oss for guided bone regeneration. Int J Oral Maxillofac Implants 1997;12:844-852.

12. Hammerle CH, Lang NP. Single stage surgery combining transmucosal implant placement with guided bone regeneration and bioresorbable materials. Clin Oral Implants Res 2001;12:9-18.

13. Jung RE, Fenner N, Hammerle CH, Zitzmann NU. Long-term outcome of implants placed with guided bone regeneration (GBR) using resorbable and non-resorbable membranes after 12-14 years. Clin Oral Implants Res 2013;24:1065-1073.

14. Miron RJ, Sculean A, Shuang Y, Bosshardt DD, Gruber R, Buser D, et al. Osteoinductive potential of a novel biphasic calcium phosphate bone graft in comparison with autographs, xenografts, and DFDBA. Clin Oral Implants Res 2016;27:668-675.

15. Cordaro L, Bosshardt DD, Palattella P, Rao W, Serino G, Chiapasco M. Maxillary sinus grafting with Bio-Oss or Straumann Bone Ceramic: histomorphometric results from a

randomized controlled multicenter clinical trial. Clin Oral Implants Res 2008;19:796-803.

16. Schmitt CM, Doering H, Schmidt T, Lutz R, Neukam FW, Schlegel KA. Histological results after maxillary sinus augmentation with Straumann(R) BoneCeramic, Bio-Oss(R), Puros(R), and autologous bone. A randomized controlled clinical trial. Clin Oral Implants Res 2013;24:576-585.

17. Froum SJ, Wallace SS, Cho SC, Elian N, Tarnow DP. Histomorphometric comparison of a biphasic bone ceramic to anorganic bovine bone for sinus augmentation: 6- to 8-month postsurgical assessment of vital bone formation. A pilot study. Int J Periodontics Restorative Dent 2008;28:273-281.

18. Patel K, Mardas N, Donos N. Radiographic and clinical outcomes of implants placed in ridge preserved sites: a 12-month post-loading follow-up. Clin Oral Implants Res 2013;24:599-605.

19. Zhang K, Fan Y, Dunne N, Li X. Effect of microporosity on scaffolds for bone tissue engineering. Regen Biomater 2018;5:115-124.

20. Yuan H, Fernandes H, Habibovic P, de Boer J, Barradas AM, de Ruiter A, et al. Osteoinductive ceramics as a synthetic alternative to autologous bone grafting. Proc Natl Acad Sci U S A 2010;107:13614-13619.

21. Zhang Q, Jing D, Zhang Y, Miron RJ. Histomorphometric Study of New Bone Formation Comparing Defect Healing with Three Bone Grafting Materials: The Effect of Osteoporosis on Graft Consolidation. Int J Oral Maxillofac Implants 2018;33:645-652.

22. Yip I, Ma L, Mattheos N, Dard M, Lang NP. Defect healing with various bone substitutes. Clin Oral Implants Res 2015;26:606-614.

23. Shuang Y, Yizhen L, Zhang Y, Fujioka-Kobayashi M, Sculean A, Miron RJ. In vitro characterization of an osteoinductive biphasic calcium phosphate in combination with recombinant BMP2. BMC Oral Health 2016;17:35.

24. Barroso-Panella A, Gargallo-Albiol J, Hernandez-Alfaro F. Evaluation of Bone Stability and Esthetic Results After Immediate Implant Placement Using a Novel Synthetic Bone Substitute in the Anterior Zone: Results After 12 Months. Int J Periodontics Restorative Dent 2018;38:235-243.

25. O'Leary TJ, Drake RB, Naylor JE. The plaque control record. J Periodontol 1972;43:38.

26. Ainamo J, Bay I. Problems and proposals for recording gingivitis and plaque. Int Dent J 1975;25:229-235.

27. Buser D, Weber HP, Bragger U, Balsiger C. Tissue integration of one-stage ITI implants: 3-year results of a longitudinal study with Hollow-Cylinder and Hollow-Screw implants. Int J Oral Maxillofac Implants 1991;6:405-412.

28. Starch-Jensen T, Mordenfeld A, Becktor JP, Jensen SS. Maxillary Sinus Floor Augmentation With Synthetic Bone Substitutes Compared With Other Grafting Materials: A Systematic Review and Meta-analysis. Implant Dent 2018;27:363-374.

29. Wu J, Li B, Lin X. Histological outcomes of sinus augmentation for dental implants with calcium phosphate or deproteinized bovine bone: a systematic review and meta-analysis. Int J Oral Maxillofac Surg 2016;45:1471-1477.

30. Piattelli M, Favero GA, Scarano A, Orsini G, Piattelli A. Bone reactions to anorganic bovine bone (Bio-Oss) used in sinus augmentation procedures: A histologic long-term report of 20 cases in humans. International Journal of Oral & Maxillofacial Implants 1999;14:835-840.

31. Schlegel KA, Fichtner G, Schultze-Mosgau S, Wiltfang J. Histologic findings in sinus augmentation with autogenous bone chips versus a bovine bone substitute. Int J Oral Maxillofac Implants 2003;18:53-58.

32. Taylor JC, Cuff SE, Leger JPL, Morra A, Anderson GI. In vitro osteoclast resorption of bone substitute biomaterials used for implant site augmentations: A pilot study. International Journal of Oral & Maxillofacial Implants 2002;17:321-330.

33. Galindo-Moreno P, Hernandez-Cortes P, Mesa F, Carranza N, Juodzbalys G, Aguilar M, et al. Slow resorption of anorganic bovine bone by osteoclasts in maxillary sinus augmentation. Clin Implant Dent Relat Res 2013;15:858-866.

34. Dahlin C, Obrecht M, Dard M, Donos N. Bone tissue modelling and remodelling following guided bone regeneration in combination with biphasic calcium phosphate materials presenting different microporosity. Clin Oral Implants Res 2015;26:814-822.

35. Habibovic P, Yuan H, van den Doel M, Sees TM, van Blitterswijk CA, de Groot K. Relevance of osteoinductive biomaterials in critical-sized orthotopic defect. J Orthop Res 2006;24:867-876.

36. Jensen T, Schou S, Gundersen HJ, Forman JL, Terheyden H, Holmstrup P. Bone-to-implant contact after maxillary sinus floor augmentation with Bio-Oss and autogenous bone in different ratios in mini pigs. Clin Oral Implants Res 2013;24:635-644.

37. Jensen T, Schou S, Stavropoulos A, Terheyden H, Holmstrup P. Maxillary sinus floor augmentation with Bio-Oss or Bio-Oss mixed with autogenous bone as graft: a systematic review. Clin Oral Implants Res 2012;23:263-273.

38. Mordenfeld A, Lindgren C, Hallman M. Sinus Floor Augmentation Using Straumann(R) BoneCeramic and Bio-Oss(R) in a Split Mouth Design and Later Placement of Implants: A 5-Year Report from a Longitudinal Study. Clin Implant Dent Relat Res 2016;18:926-936.

FIGURES:

Figure 1: Mean proportions of newly formed bone, bone grafting materials and non-mineralized tissue. (BCP: biphasic calcium phosphate; DBBM: deproteinized bovine bone mineral)

Figure 2: Section of trephine burs and bone biopsies stained with toluidine blue. Magnification: 25x. DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate. Red frames: Magnification 50x (illustrated in Figures 3 and 4)

Figure 3: Histological section of bone grafted with DBBM, stained with toluidine blue. Magnification: 50x. DBBM: deproteinized bovine bone mineral; NB: new bone; NMT: non-mineralized tissue.

Figure 4: Histological section of bone grafted with BCP, stained with toluidine blue. Magnification: 50x. BCP: biphasic calcium phosphate; NB: new bone; NMT: non-mineralized tissue.

TABLES:

Table 1: Ratio of new bone (%) vs. bone grafting material (%) (DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate).

Table 2: Ratio of new bone (%) vs. bone grafting material (%) for the subgroup of patients with bilateral treatment (DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate).

Table 1: Ratio of new bone (%) vs. bone grafting material (%) (DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate).

	DBBM	**BCP**
mean (±standard deviation)	0.86 (±0.45)	3.48 (±6.76)
minimum	0.38	0.27
25% percentile	0.58	1.10
median	0.67	1.38
75% percentile	0.98	2.22
maximum	2.31	31.07
Biopsies	**N=33**	**N=29**

Table 2: Ratio of new bone (%) vs. bone grafting material (%) for the subgroup of patients with bilateral treatment (DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate).

	DBBM	**BCP**
mean (±standard deviation)	0.73 (±0.31)	4.08 (±7.74)
minimum	0.46	0.34
25% percentile	0.53	1.00
median	0.59	1.37
75% percentile	0.83	3.83
maximum	1.59	31.07
Biopsies	**N=15**	**N=15**

Figure 1: Mean proportions of newly formed bone, bone grafting materials and non-mineralized tissue (BCP: biphasic calcium phosphate; DBBM: deproteinized bovine bone mineral).

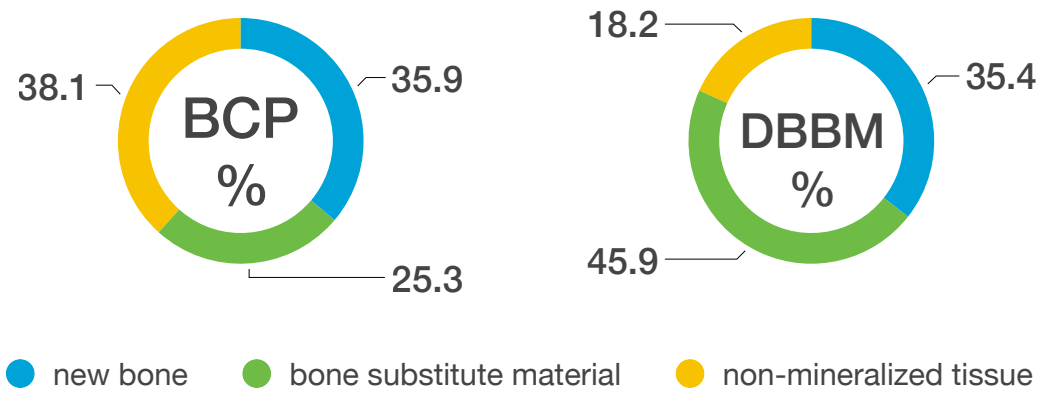

Figure 2: Section of trephine burs and bone biopsies stained with toluidine blue. Magnification: 25x. DBBM: deproteinized bovine bone mineral; BCP: biphasic calcium phosphate; NB: new bone; NMT: non-mineralized tissue. Red frames: Magnification 50x (illustrated in Figures 3 and 4).

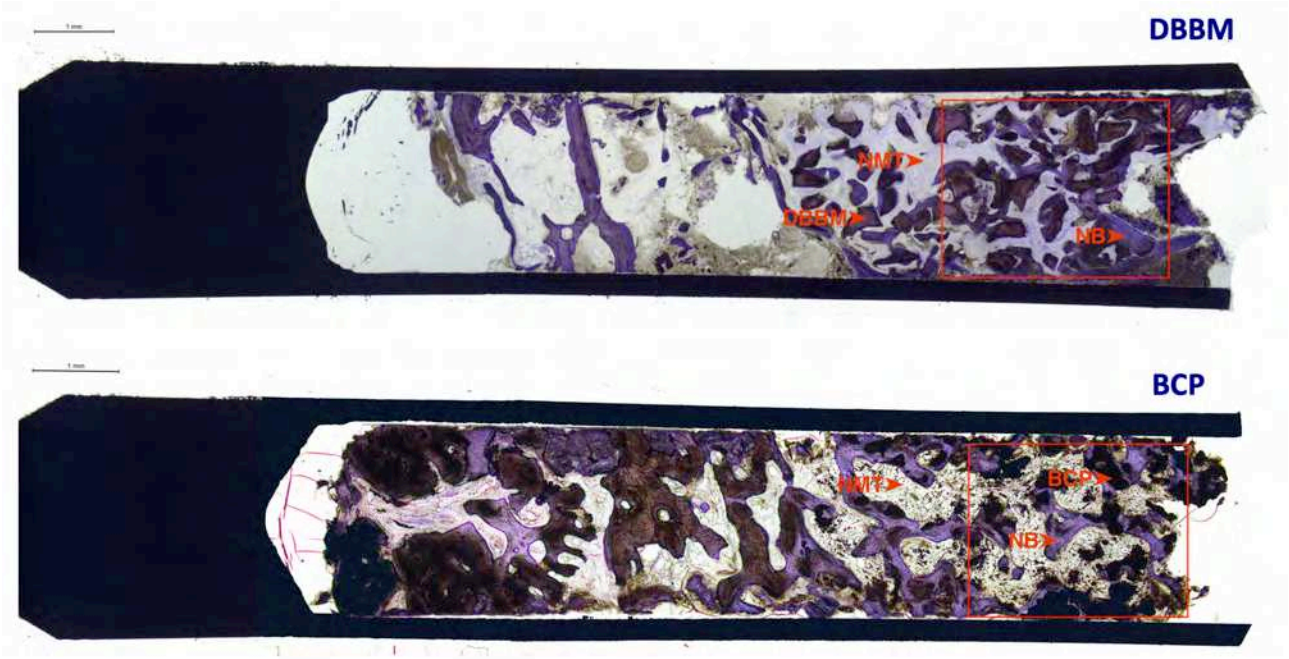

Figure 3: Histological section of bone grafted with DBBM, stained with toluidine blue. Magnification: 50x. DBBM: deproteinized bovine bone mineral; NB: new bone; NMT: non-mineralized tissue.

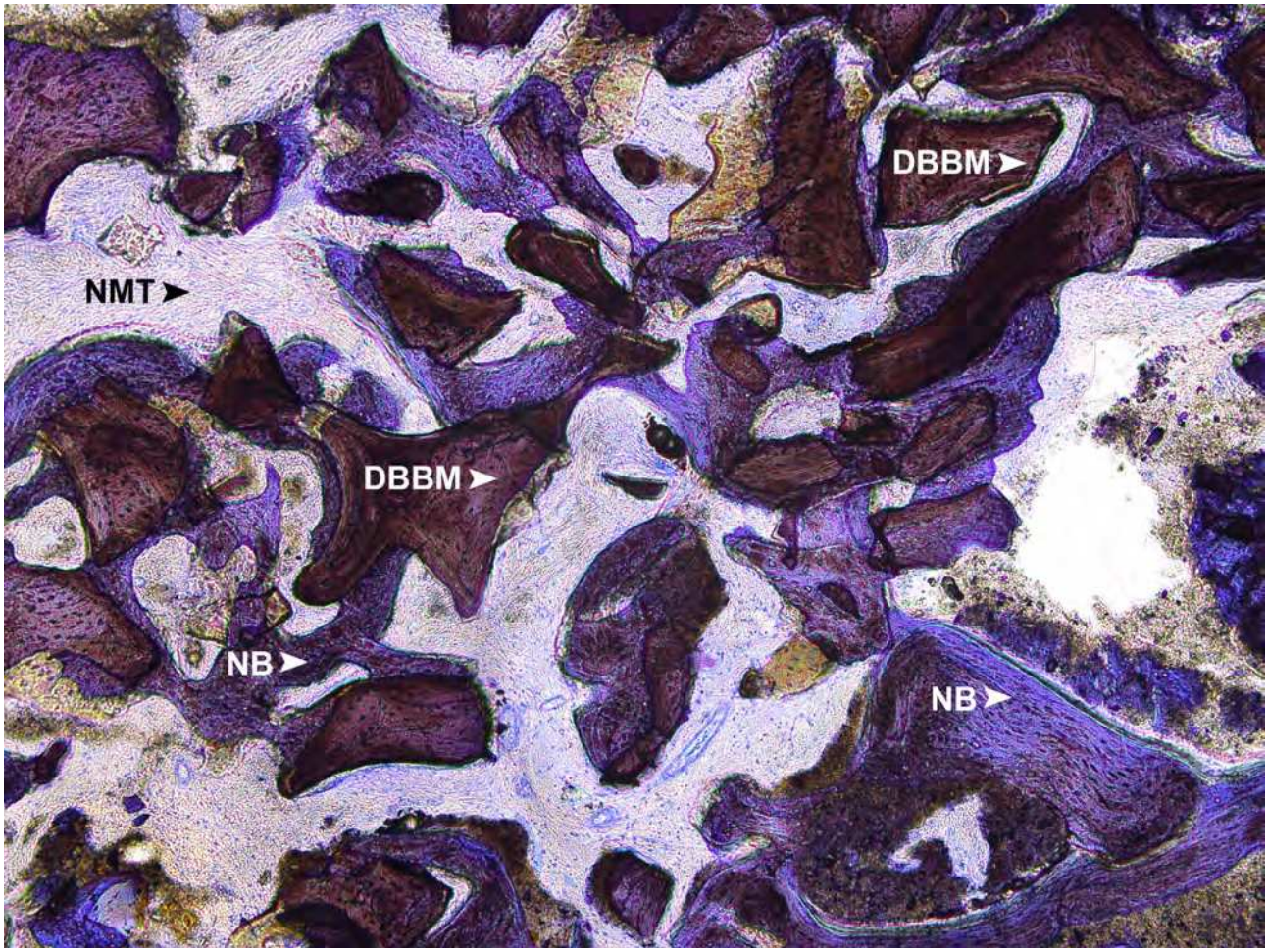

Figure 4: Histological section of bone grafted with BCP, stained with toluidine blue. Magnification: 50x. BCP: biphasic calcium phosphate; NB: new bone; NMT: non-mineralized tissue.