

ORIGINAL ARTICLE

Comparison between mineralized cancellous bone allograft and an alloplast material for sinus augmentation: A split mouth histomorphometric study

Roni Kolerman, DMD¹ | Joseph Nissan, DMD² | Marina Rahmanov, DMD² |
Hana Vered, MS³ | Omer Cohen, DMD, PhD¹ | Haim Tal, DMD, PhD¹

¹Department of Periodontology, The Maurice and Gabriela Goldschleger School of Dental Medicine, The Sackler Faculty of Medicine, Tel-Aviv University, Tel-Aviv, Israel

²Department of Oral Rehabilitation, The Maurice and Gabriela Goldschleger School of Dental Medicine, The Sackler Faculty of Medicine, Tel-Aviv University, Tel-Aviv, Israel

³Department of Oral Pathology, The Maurice and Gabriela Goldschleger School of Dental Medicine, The Sackler Faculty of Medicine, Tel-Aviv University, Tel-Aviv, Israel

Correspondence

Roni Kolerman, Department of Periodontology, The Maurice and Gabriela Goldschleger School of Dental Medicine, The Sackler Faculty of Medicine Tel-Aviv University, Klachkin 4, Israel.
Email: daniaron@netvision.net.il; kolerman@netvision.net.il

Funding information

MIS Israel

Abstract

Background: Several grafting materials have been used in sinus augmentation procedures including autogenous bone, demineralized freeze-dried bone, hydroxyapatite, β -tricalcium phosphate, anorganic deproteinized bovine bone, and combination of these and others. Yet, the issue of the optimal graft material for sinus floor augmentation is controversial.

Purpose: This prospective, randomized split-mouth study was undertaken to histomorphometrically compare a biphasic calcium phosphate (BCP) alloplastic bone substitute and a human bone mineral allograft (freeze-dried bone allograft, FDBA) in patients undergoing bilateral maxillary lateral sinus floor augmentation.

Material and methods: Apico-coronal core biopsies were harvested at 9 months from 26 bilateral sites in 13 treated patients. Specimens were processed for histological and histomorphometrical analyses.

Results: Newly formed bone (NB) was evident in all specimens with values of 27.5% and 24.0% at the FDBA and BCP sites, respectively ($P = .331$). The residual graft particle values were 12.5% and 25.4% ($P = .001$), and the connective tissue values were 60.0% and 50.6%, respectively. The osteoconductive value was 52.6% for the FDBA and 26.7% for the alloplast ($P = .001$). The values for the measured residual graft particles, connective tissue, and osteoconductivity, but not for NB, showed highly significant differences between the two groups. All sections in the alloplast material showed evidence of a light chronic inflammatory infiltrate, mainly comprising lymphocytes and multinucleated giant cells.

Conclusions: Both graft materials are suitable for sinus floor augmentation, with the allograft material being more osteoconductive.

KEYWORDS

biomaterials, bone substitute, sinus floor elevation

1 | INTRODUCTION

Alveolar bone resorption in the posterior maxilla is a common sequela of tooth loss and periodontal disease.¹ A lack of sufficient alveolar bone height in this area, especially below the maxillary sinus, often makes it impossible to place standard implants. The most common intervention currently used to increase bone height in this region is to

augment the maxillary sinus floor with autogenous bone grafts,^{2,3} a procedure referred to as "sinus floor elevation/augmentation." Tatum first described this procedure,⁴ and Boyne and James coined the term "sinus lift procedure" shortly thereafter and described the surgical intervention of raising the maxillary sinus floor by elevating the sinus mucosa and interposing bone grafts between the mucosa and bony sinus floor, resulting in adequate bone formation to anchor dental

implants of optimal length.⁵ Several systematic literature reviews demonstrated that the procedure may be safely applied in cases of posterior maxilla atrophy, leading to an implant survival rate of higher than 90% both in the short term and after more than 3 years of function.^{6–9} Many bone substitute materials have been used for grafting the cavity created during the maxillary sinus augmentation procedure. For bone regeneration to occur, biomaterials must be biocompatible, osseointegrative, and biodegradable. An optimal bone substitute biomaterial should act as a temporary scaffold for supporting the adhesion, growth, proliferation, and differentiation of the “seed” cells and should also degrade into nontoxic products that can be metabolized via physiological mechanisms.¹⁰ Osseointegrative materials (materials that enhance bone formation on their surface), including freeze-dried bone allograft (FDBA), demineralized freeze-dried bone allograft (DFDBA), xenografts produced from bovine bone (BB), porcine bone (PB), and alloplastic materials (β -tricalcium phosphate [TCP] and hydroxyapatite [HA]), as well as combinations such as that of HA and α - and β -tricalcium phosphate (β -TCP) may allow new bone formation, thus constituting a scaffold that serves to stabilize the bone clot and support bone growth during the early healing phase.¹¹ Recently, osteoinductive materials, including scaffolds enriched with recombinant osteoinductive factors (such as bone morphogenetic protein-2 [rhBMP-2]) have been claimed to enhance bone formation by stimulating the bone regeneration process.^{11,12} The discovery of some cases of human immunodeficiency virus (HIV) transmission to recipients of bone allograft products (DFDBA), the persistence of infectious diseases such as hepatitis via bone allograft,¹³ and the fear of bovine spongiform encephalopathy (BSE, “mad cow disease” applied for xenografts) transferring to humans¹⁴—although no documentation is available—have raised concerns about potential disease transmission from xenografts and allografts to man. Therefore, the use of alloplast materials has been considered a viable alternative that is well accepted by most patients. Bioceramics made from a mixture of HA and β -TCP have demonstrated satisfactory bioactivity and osteoconductivity.^{15–18}

Alloplastic bone substitute materials such as biphasic calcium phosphate (BCP) including HA and α -TCP and β -TCP have been shown to attract multinucleated giant cells (MNGCs) after implantation, although at different rates.¹⁹ These cells are thought to be responsible for the degradation of bone substitute materials and at the same time induce vascularization by releasing vascular endothelial growth factor and other biologically active compounds, such as chemokines.¹⁹ The number of these MNGCs at the implantation bed varies depending on the histochemical structure of the biomaterials.^{20,21}

Comparisons between demineralized and non-demineralized freeze-dried bone allografts (DFDBA, FDBA) have revealed that FDBA results in significantly more new bone growth over time than DFDBA.⁹

To the best of our knowledge, few randomized split-mouth histomorphometric studies have compared the use of FDBA and alloplast materials in human sinus augmentation procedures.⁹

Thus, the purpose of this study was to histologically and histomorphometrically compare the use of FDBA and a BCP alloplastic material

in sinus floor augmentation procedures. The hypothesis is that both materials facilitate similar amounts of new bone in the augmented sinuses.

2 | MATERIALS AND METHODS

2.1 | Study population

Thirteen adults (7 females, 6 males) aged 43–68 years (average 58 years) with no systemic disorders that could affect sinus augmentation surgery were selected from a pool of patients who required bilateral sinus lift procedures for posterior implant placement and comprised the study population (Table 1). The exclusion criteria used were chronic steroid therapy, uncontrolled diabetes, cardiovascular disease prohibiting extensive surgery, past head and neck irradiation, maxillary sinus cysts, active chronic sinusitis, smoking more than 10 cigarettes per day during the study period, or inability to perform proper oral hygiene.

One periodontist (RK) treated these patients between 2008 and 2013. Alternative treatment plans were discussed with each patient, and the patients and periodontist together selected the plan that required maxillary sinus elevation. All 13 patients presented with a moderate or severe atrophic posterior maxilla with residual alveolar bone <5 mm. The study participants signed an informed consent form, in which the procedure was explained in detail. The ethics committee of Tel-Aviv University approved the study protocol.

A staged approach was carried out at all 26 sites. Sinus floor augmentation procedures were followed by implant placement 9 months later. Mineralized 250–710 μ m FDBA (Raptos, Citageix, Laval, Canada) was randomly applied on one side and a synthetic HA and BCP 60:40 alloplast (4Bone SBS, Biomatlante, Vigneux de Bretagne, France) with a particle size of between 0.5 and 1 mm was applied on the contralateral side.

A thorough presurgical evaluation was carried out, including a full-mouth periodontal chart, occlusal analysis, study of the mounted casts and diagnostic wax-up, as well as initial periodontal therapy, including oral hygiene instructions and training as well as scaling and root planning where indicated. This was followed by additional periodontal therapy to reduce periodontal probing depth and bleeding on probing until a plaque index <10% was achieved. Computerized tomographic (CT) measurements showed a <5 mm bone height in both sinus floors in each patient.

2.2 | Surgical technique

Premedication with 8 mg dexamethasone (Rekah Pharm Ind, Holon, Israel) and 875 mg amoxicillin-clavulonate potassium (Augmentin—Glaxo Smith Klein, Brentford, UK) was administered 1 hour preoperatively. Immediately before surgery, the patients rinsed their mouths with 0.2% chlorhexidine for 1 minute (Tarodent Taro Pharm Ind, Haifa, Israel). The surgical procedures followed the technique described by Smiler and Holmes²² with only minor modifications as described by Kolerman and colleagues.^{15,23} Briefly, after exposure of the lateral bony wall of the antrum and demarcation of the window boundaries

TABLE 1 Histomorphometric data

Patient	Sex	Age (y)	Osteoconductive value (%)		Total mineralized tissue (%)		Residual graft (%)		Newly formed bone (%)	
			BCP	FDBA	BCP	FDBA	BCP	FDBA	BCP	FDBA
1	M	54	25.5	9.7	40.6	36.2	22.2	26.3	18.4	9.9
2	F	58	23.0	61.9	51.7	31.0	19.4	10.4	32.3	20.6
3	F	66	27.6	47.4	50.4	46.9	24.9	19.3	25.5	27.6
4	M	60	34.2	64.0	40.0	37.4	19.0	4.4	21.0	33.0
5	F	63	21.3	56.5	38.0	45.9	22.6	8.7	15.4	37.2
6	M	51	33.0	56.6	62.4	43.0	33.0	12.1	29.4	30.9
7	M	57	19.7	47.2	52.4	38.3	37.7	16.1	14.7	22.2
8	M	43	28.9	55.7	54.6	41.5	30.0	20.2	24.6	21.3
9	F	58	27.6	49.9	50.6	57.0	21.7	18.0	28.9	27.6
10	F	56	10.1	50.3	58.7	38.1	24.9	2.8	33.8	35.3
11	F	68	25.3	62.1	56.0	46.8	26.4	20.3	29.6	26.5
12	F	58	37.6	85.4	40.5	24.4	23.9	1.4	16.6	23.0
13	M	59	39.5	43.0	45.6	34.0	27.3	3.0	18.3	31.0
Mean		58	26.7 ^a	52.6	49.4 ^a	40.0	25.4 ^a	12.5	24.0	27.5
SD			7.9	16.9	7.8	8.3	5.5	8.1	6.8	8.1

^aStatistically significant.

with a 3-mm diamond bur, the lateral bony wall was completely removed, exposing the Schneiderian membrane. The membrane was gently separated and gently reflected using a broad flat curette (Kärmer-Nevins IMP6578, Hu-Friedy, Chicago, Illinois), creating a space beneath the elevated membrane and the peripheral bony walls.

A bioabsorbable porcine collagen barrier membrane (BioGide, Geistlich Pharma AG, Wolhusen, Switzerland) was placed under the membrane and was adapted to the membrane and to the peripheral bony walls. The established void was filled with either alloplast (4Bone SBS: Biomatlante, Vigneux de Bretagne, France) or FDBA (Raptos, Citigeix, Laval, Canada) in the left or the right sinus (this was determined by tossing a coin). An outer similar occlusive barrier membrane was applied to cover the entire external augmented site. Postoperatively, the same systemic antibiotics (Augmentin 875 mg twice daily) were administered for 1 week, and 4 mg dexamethasone was prescribed for 2 successive days. A generic nasal decongestant (Sinaf, Taro Pharm, Haifa Bay, Israel) was recommended. A 0.2% chlorhexidine gluconate solution was used 2 times per day for 2 weeks until suture removal.

At 9 months following the procedure, the exact location of the grafted/regenerated sites was examined and identified by CT scan (Figure 1). A 2- or 3-mm inner diameter (3- to 4-mm outer diameter) trephine bur (Biomet 3i, Palm Beach Gardens, Florida) depending on the upcoming implant to be inserted (3 outer diameters for 3.75 and 4.2 mm and 4 outer diameters for 5 mm implants) was used to harvest a bony core from the implant sites, followed by suitable drilling to the final dimensions of each osteotomy. Screw-type sandblasted acid-etched surface bone level titanium implants were used (Lance or Seven MIS implants, Bar Lev Industrial Zone, Israel). When two or more

implants were placed on one side of the jaw, the longer and more intact core specimen was processed for histomorphometric analysis. Cylindrical cores (2–3 mm in diameter, 7–13 mm in length) were harvested and fixed in 10% neutral buffered formalin for 76 hours, decalcified in 5% formic acid for 14 days, and embedded in paraffin. Three of

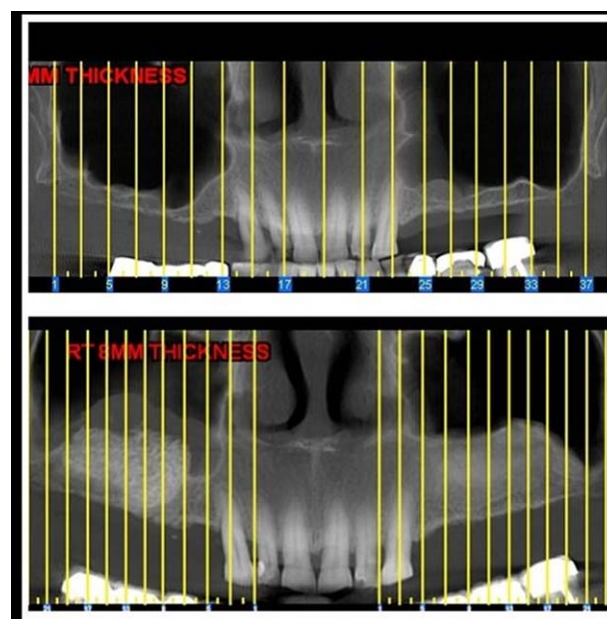


FIGURE 1 CT scan presenting severe bilateral atrophic posterior maxilla with residual alveolar bone <5 mm and a CT scan performed 9 months after bilateral sinus augmentation. The right sinus was augmented using BCP while the left by FDBA

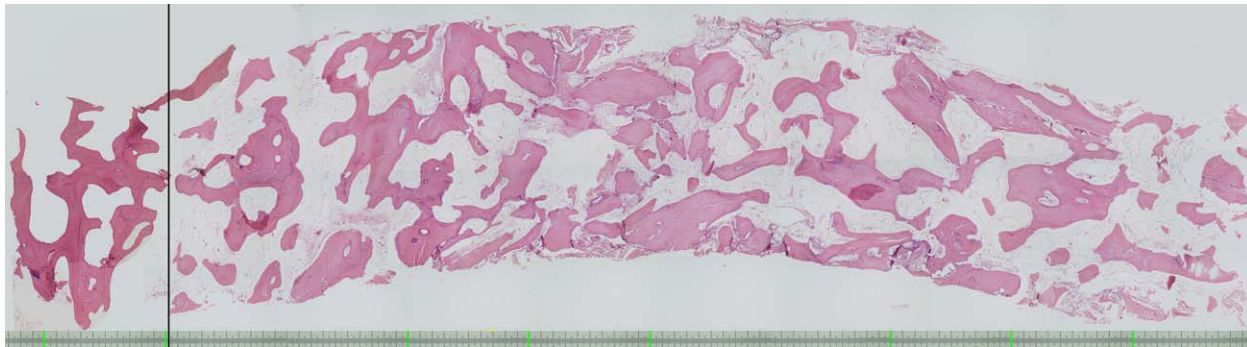


FIGURE 2 Histologic section at 9 months. FDDB particles are observed close to newly formed bone and in contact with connective tissue. The line demarcates the beginning of augmented bone (H&E, original magnification $\times 40$)

the most central 5- μm -thick sections were stained with hematoxylin-eosin (H&E) and Masson-Trichrome.

2.3 | Histomorphometric analysis

The stained specimens were photographed using a TCA-3 digital camera (Tucsen Imaging Technology Co, Province Jiayijie, Ltd, Shenzhen, Guangdong, China) Under a BH-2 light microscope (Olympus, Tokyo, Japan) at $\times 40$ magnification (Figures 2 and 3). BCP particles were identified by their typical morphology and color (Figure 4). The measurements on the allograft biopsies were performed under $\times 40$ magnification, and each biopsy was divided into 10–20 parts for calculation purposes. Each allograft specimen was divided into 40–50 parts ($\times 100$ magnification) to precisely identify graft particles that were similar to the newly formed bone and that were identified by the presence of empty lacunae and a delicate reversal line (Figure 5). Images were then processed to produce a segmented, pseudocolor image for identifying different tissue components. The processed images were analyzed using the Adobe Photoshop software (Adobe system software, Riverwalk, Citywest Business Park, Dublin, Ireland) to assess the percentages of the different components and the interface between the residual graft particles and the newly formed bone (Figure 6).

2.4 | Osteoconduction

After calibrating the system and digitizing the images, measurements were carried out using Bioquant Osteo 2009 version 9XP software (Bioquant Image Analysis Corporation, Nashville, Tennessee); the entire circumference of each section (containing new bone, graft particles, and soft connective tissue) was traced manually to create an individual region of interest. Interactive measurements of the areas of interest were obtained using image analysis. Histomorphometric analysis was used to calculate percentage bone-to-graft particle contact (BGC %) by dividing the length of graft particles in contact with the new bone perimeter by the circumference of the graft particle perimeter.¹⁵ Pristine (native) bone was identified according to the lack of graft material and was excluded from the analyzed data.

2.5 | Statistical analysis

Continuous variables are expressed as means \pm SD. The comparison between the two graft materials was examined using the nonparametric Wilcoxon Signed Rank Test due to the small sample size. Owing to the possible asymmetric distribution of the data, results were further analyzed using the Sign Test. In both tests, graft material was the

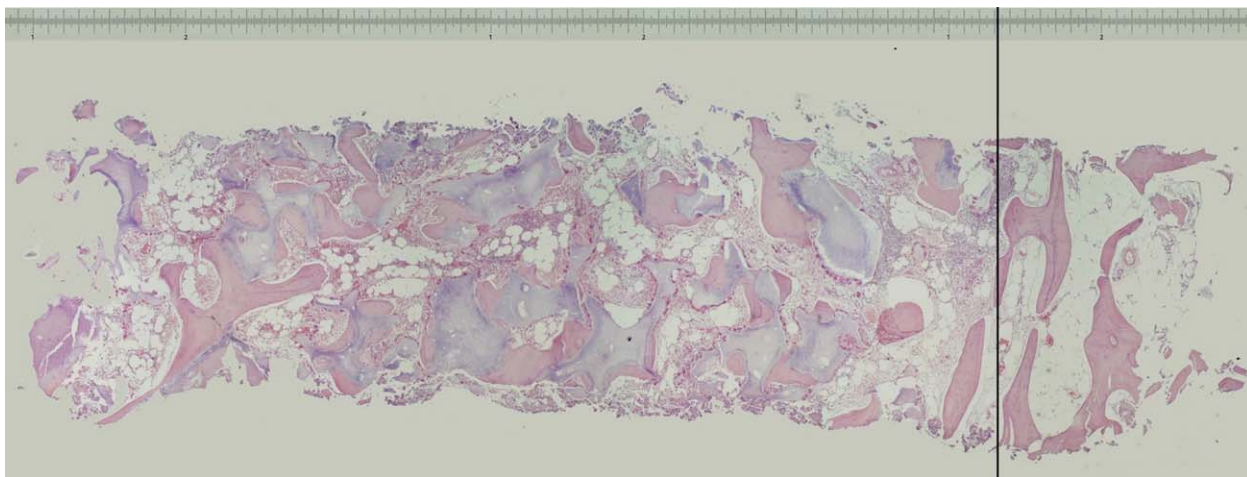


FIGURE 3 Histologic section at 9 months. BCP particles are observed close to newly formed bone and in contact with connective tissue (H&E, original magnification $\times 40$)

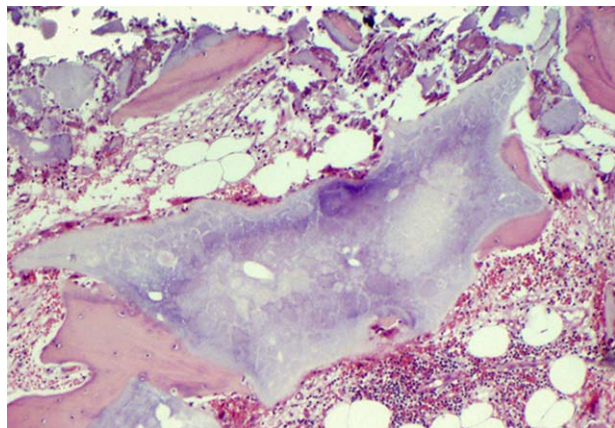


FIGURE 4 Histologic section showing BCP particles at 9 months (H&E, original magnification $\times 100$). Light chronic inflammatory infiltrate mainly lymphocytes cells and multinucleated giant cells. The intertrabecular marrow consisted of loose connective tissue and foci of adipose tissue

independent variable, while soft connective tissue, residual graft, new bone, and osseointegrative value served as dependent variables. All analyses were performed using IBM SPSS Statistics for Windows v24.0.1 (IBM Corp., Armonk, New York). A 2-tailed P value of $< .05$ was considered statistically significant.

3 | RESULTS

All 26 augmented sinuses provided ≥ 12 mm available bone for implant placement. Each augmented sinus provided at least one intact core, 2–3 mm in width and 7–13 mm in length. All specimens included both newly formed bone and, with four exceptions, pristine bone.

3.1 | Histology

Biopsy specimens contained both pristine and newly formed bone. The transition between the two was traced by a straight-line perpendicular

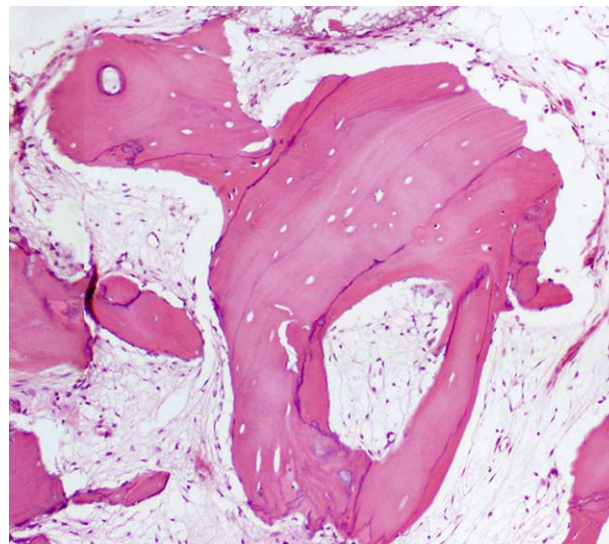


FIGURE 5 Histologic section showing FDDB particles at 9 months. The particles are embedded in the regenerated bone and in contact with loose connective tissue (H&E, original magnification $\times 100$)

to the long axis of the biopsy according to the basal part of the first graft particles identified (Figures 2 and 3). The following histological observations relate exclusively to the augmented zone, which is coronal to this arbitrary borderline.

FDDB: Newly formed bone was evident in all augmented sites. Allograft particles were in intimate contact with newly formed bone, and partly surrounded by loose connective tissue (Figure 5). There was no evidence of inflammatory infiltrate.

BCP: Newly formed bone was evident in all specimens (Figure 4), however, all the sections showed evidence of—light chronic inflammatory infiltrate mainly lymphocytes cells and MNGCs (Figure 7). The inter-trabecular marrow consisted of loose connective tissue and foci of adipose tissue and small sized blood vessels. Pristine bone consisted of low-density spongiosa and well-vascularized bone marrow.

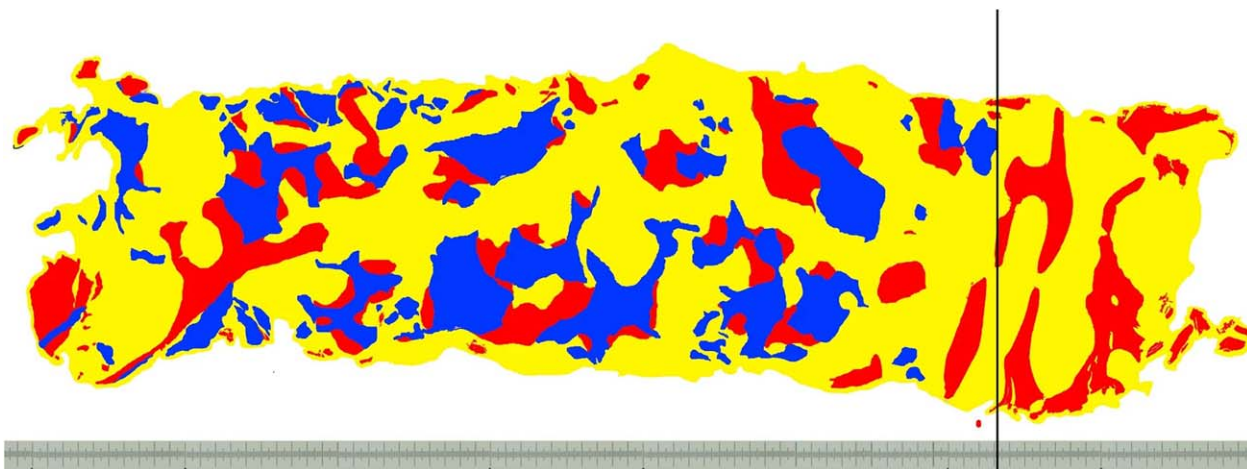


FIGURE 6 A segmented, pseudocolour image identifying BCP particles close to newly formed bone and in contact with connective tissue (H&E, original magnification $\times 100$). New bone (red), graft particles (blue), and connective tissue (yellow) are shown

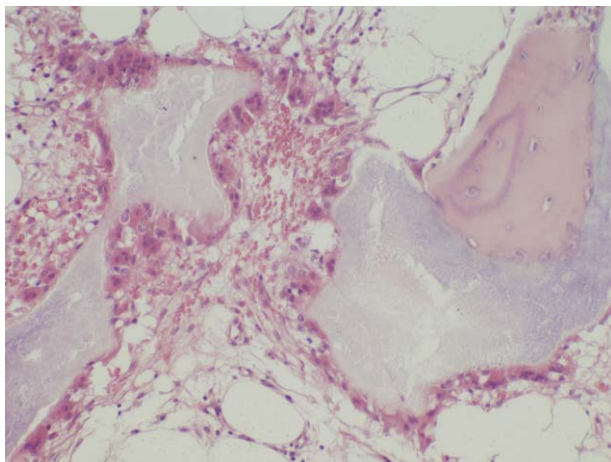


FIGURE 7 Intense chronic inflammatory infiltrate comprising mainly lymphocytes and multinucleated giant cells, around BCP particles

3.2 | Histomorphometry

Measurements were taken only from the core zone containing newly formed tissue and graft material. Graft particles were identified by their typical structure and color. New bone content ranged between 9.9%–37.2% with a mean of $27.5 \pm 8.1\%$ in the FDDBA specimens and 14.7%–33.8% with a mean of $24.0 \pm 6.8\%$ in the BCP specimens (Table 1). The mean percentage of marrow and connective tissue was $60 \pm 8.1\%$ for the FDDBA specimens and $50.6 \pm 7.8\%$ for the BCP specimens.

The comparison between the two materials appear in Table 2. The difference in new bone formation fraction was not statistically significant. The average percentage of residual graft was significantly lower in the FDDBA specimens (range: 1.4%–26.3%; mean: $12.5 \pm 8.1\%$) than in the BCP specimens (range: 19.4%–37.7%; mean: 25.4 ± 5.5). The mean osseointegrative value of the FDDBA specimens ($52.6\% \pm 16.9\%$) was significantly higher than for the BCP specimens ($26.7 \pm 7.9\%$). Total mineralized tissue (newly formed bone plus graft particles) accounted for 40.0% and 49.4% of the biopsy areas, respectively ($P = .008$ and $P = .022$ using the Wilcoxon Signed Ranks Test and the Sign Test, respectively) (Table 2).

4 | DISCUSSION

The present study is based on the results of a bilateral sinus floor augmentation procedure using a lateral wall technique with internal (beneath the Schneiderian membrane) and external (over the lateral window) collagen membrane placement. The objectives of this study were to evaluate and compare the healing response between two mineralized materials that were used for sinus lift procedures, FDDBA and β -TCP-HA (BCP).

It has been assumed that differences between the filler materials used in sinus lift procedures may modulate the quality and amount of newly formed bone. In the current study, both materials showed graft particles that were in direct contact with new bone. Bone growth in the FDDBA sites (27.5%) was comparable with that in the alloplast sites

TABLE 2 Comparison between FDDBA and BCP in 13 patients 9 months after bilateral sinus augmentation

	BCP	FDDBA	P^a	P^b
Osteoconductive value (%)	26.7 ± 7.9	52.6 ± 16.9	.001	.003
Total mineralized tissue (%)	49.4 ± 7.8	40.0 ± 8.3	.008	.022
Residual graft (%)	25.4 ± 5.5	12.5 ± 8.1	.001	.003
Newly formed bone (%)	24.0 ± 6.8	27.5 ± 8.1	.331	.581

^a P value by Wilcoxon signed rank test.

^b P value by sign test.

(24.0%). Our results regarding new bone formation in the BCP-grafted sides were comparable to the data published by Corbella and colleagues,⁹ who analyzed 11 articles that were published between 2001 and 2014, showing that average new bone growth was 26.3%. Those articles were divided into periods of short (<6 months, 23.1%) and long (>6 months, 29.4%) duration, and it was suggested that the potential maturation of BCP graft material may be time-dependent and that new bone formation increases mainly at the expense of the connective tissue compartment, while the residual graft particles fraction remains stable.⁹ The present results revealed that new bone formation on the FDDBA sides (27.5%) was less than that described by Cammack,²⁴ who used FDDBA (41.1%) and DFDBA (36%) for sinus lift procedures. In their study, the histomorphometric analysis included the summing of a set of polygons traced on the sample, which allowed the software to calculate a set of areas (ie, new bone, residual graft particles and soft tissue) as fractions of the total area of the sample. It appeared that DFDBA was resorbed more than FDDBA over time and that it induced less new bone than FDDBA, although no comparative studies were published.⁹ Another study,²⁵ in which a composite graft of DFDBA and BB was used in 20 patients, claimed that new bone was observed in contact with BB particles, while DFDBA particles were surrounded by connective tissue. The data obtained in the present study regarding new bone formation using FDDBA materials are similar to the data (27.2%–29.1%) that were reported by the same group using a different FDDBA material (Life Net Health, Virginia Beach, Virginia),^{23,26} although the histomorphometric technique used in the previous studies was point counting. The new bone formation fraction of FDDBA (27.5%) reported here is comparable to that (29%) presented by Scarano and colleagues,²⁷ although our bone samples were taken after 9 months and theirs after 6 months.

We used a mineralized material because of the relatively low osseointegrative property of DFDBA.^{25,28} It is noteworthy that while new bone formation fractions for FDDBA and BCP were similar (27.5% and 24%, respectively), the fractions of residual graft particles (12.5% vs 25.4%) were significantly different. The present residual BCP fraction of 25.4% was similar that of the graft material compartment using 4Bone reported previously (27.3%)¹⁵ as well as to the mean of 25.8% that was extrapolated from 11 studies using an HA-TCP formula.⁹ However, the scarcity of published comparative reports in this field imposes a difficulty in determining significant differences between the variously examined biomaterials in terms of new bone formation.

In the presence of adequate clot stability, a porous scaffold and integrity of the sinus membrane, the space between the sinus membrane and the residual peripheral bone may be repopulated by osteoprogenitor cells that induce bone growth independently of whether the biomaterial fills the available space.⁹ A number of studies reported that the sinus floor augmentation procedure might be successful even without grafting any material within the above-mentioned space.^{29,30}

Considerable data have been obtained on new bone formation in sinus floor elevation procedures; however, very few publications have dealt with the osseointegrative properties of the grafts.

Renno and Vivan^{21,31} suggested that for satisfactory biological interaction to occur, bone replacement graft materials must not only be biocompatible but must also present a high bioactive level resulting in over 50% of living tissue (new bone) being attached to a material surface. This attachment is established through the formation of a layer of biologically active hydroxycarbonate apatite at the material/tissue interface, quite similar to bone tissue apatite. In view of this assumption, the allograft fulfilled both eligible criteria: a. between 25% and 30% of new bone as proposed by Jensen²⁸ and b. a satisfactory bioactive level as reflected by its relatively high osteoconductive value.³¹

The use of the xenograft Bio-Oss has been associated with a new BGC of 34%-38%^{32,33} to 40.17%.³⁴ Nevertheless, a significantly higher value was measured (54.33%) using allografts (Puros).³³ The osteoconductive values of the FDDB surface obtained in the present work (52.6%), were higher than those of the alloplasts (26.7%), indicating that the integration of human particulate bone graft with the newly formed bone tissue is better than that obtained using a synthetic graft. Our data relating to bone-to-graft contact are consistent with those of Cordaro,³⁵ who reported a lower bone-to-implant contact ratio in a BCP graft (34%) compared to a BB mineral graft (48.2%). The direct comparison made in the present study, which takes into consideration the higher amount of residual BCP particles (25.4%) compared with FDDB particles (12.50%) and the relatively low wet ability/conductivity of the BCP, may indicate that the new bone has different mechanical properties. The allograft used incited no inflammatory infiltrate, similar to our previous observations,^{23,26} permitting a direct deposition of the newly formed bone on its surface. Recently accumulated evidence has put into question the role of large MNGCs in bone biomaterials.³⁶ MNGCs exist around the tissue/biomaterial interfaces of implanted medical devices and at injury sites.³⁶ MNGCs have also been seen in several tissues, where the size of the foreign particle is greater than that which will allow macrophage phagocytosis to occur.³⁷ Since then, macrophages have been suggested to fuse in response to larger-than-average particle sizes ("frustrated" macrophages).³⁷ Human histological samples in which bone grafting materials have been used for bone augmentation have consistently shown a substantially higher number of MNGCs around bone substitute material grafts in stable situations, that is, when they were harvested years after the original surgeries were performed.³⁸ These findings call into question the hypotheses that "foreign body reactions" may occur by demonstrating better and more stable long-term bone volume around certain bone grafts.³⁸ Furthermore, as in the present study, where residual graft BCP particles

accounted for 25.4% of the total tissue area, a selected class of bone substitutes (especially synthetic and BB substitutes) are routinely found to have significantly higher numbers of MNGCs.³⁹⁻⁴¹ These findings further question the role of MNGCs in biomaterials because these cells were once thought to only contribute to the foreign body reaction and, as documented recently, their phenotypes are implicated in wound healing and tissue regeneration and have the potential to degrade biomaterials.^{20,42}

It has been shown that the presence of MNGCs within biomaterial implantation beds is not only related to the type of bone substitute material used but also to the granule size of the bone substitute material used. Smaller xenogeneic bone substitute granules have been associated with MNGCs,⁴³ whereas larger granules are integrated within the implantation bed by means of mononuclear cell-triggered granulation tissue.¹⁹ These data can explain the absence of granulation tissue and the reactive giant cells that were noticed in all the BCP specimens in the present study because we used a small (0.5-1 mm) particle size.

It is therefore questionable whether the classic evaluation methods that consider single main criteria of a minimum of 25%-35% new bone formation as being acceptable²⁸ for the maintenance of osseointegration over time are valid.

The advantages of using a barrier membrane over the lateral bony window are well documented.^{44,45} However, no consensus has yet been reached on that issue, and some reports claim that such barriers provide no additional benefit.⁴⁶ In our opinion, the value of an internal collagen membrane beneath the schneiderian membrane has not attracted much attention. In the present clinical protocol, collagen membranes were placed beneath the reflected schneiderian membrane, even if the membrane appeared intact. Care was taken to avoid covering the peripheral bony walls to allow maximal vascularization of the grafted space. The effect of sinus membrane perforations upon implant success is still controversial.^{46,47} However, we believe that blood clot formation and subsequent bone growth are more predictable when an additional biological barrier is used. A perforated sinus membrane carries the risk of infection by exposing the healing site to the external respiratory system^{48,49}; therefore, the use of an internal membrane may help prevent the passage of both graft particles and bacterial contamination into and out of the sinus cavity via unseen potential small tears. It is noteworthy that although the revascularization of the sinus augmentation material is potentially delayed by covering an intact sinus membrane with resorbable membrane, a previous study⁵⁰ concluded that repairing perforations of the sinus membrane with a collagen membrane did not compromise the osseointegration of dental implants that were placed in the augmented maxillary sinus.

5 | CONCLUSION

In summary, it may be concluded that although FDDB and BCP present similar amounts of new bone formation, the BCP showed a lower level of bioactivity as reflected by its reduced osteoconductive. The incited chronic inflammatory infiltrate in the BCP samples indicates that

further studies are necessary to identify the factors that result in this healing response when used in maxillary sinus augmentation and disclose its clinical meaning.

ACKNOWLEDGMENTS

This study was partially supported by the Gerald A. Niznick Chair of Implantology at Tel-Aviv University and (grant) by MIS Israel (Implants Technologies, Bar Lev Industrial Zone, Israel). The authors wish to thank Prof. Marilena Vered head of the Department of Oral Pathology at Tel-Aviv University for her help and Ms. Esther Eshkol for editorial assistance.

CONFLICT OF INTEREST

The authors have no conflict of interest.

REFERENCES

- [1] Pecora GE, de Leonardi D, Rocca CD, Cornolini R, Cortesini C. Short-term healing following the use of calcium sulfate as a grafting material for sinus augmentation: a clinical report. *Int J Oral Maxillofac Implants.* 1998;13:866–873.
- [2] Peleg M, Mazor Z, Chaushu G, Garg AK. Sinus floor augmentation with simultaneous implant placement in the severely atrophic maxilla. *J Periodontol.* 1998;69:1397–1403.
- [3] Wallace SS, Froum SJ. Effect of maxillary sinus augmentation on the survival of endosseous dental implants. A systematic review. *Ann Periodontol.* 2003;8:328–343.
- [4] Tatum OH. Maxillary sinus grafting for endosseous implants. In: Presented at the Annual Meeting of the Alabama Implant Study Group; 1977; Birmingham, AL.
- [5] Boyne PJ, James RA. Grafting of the maxillary sinus floor with autogenous marrow and bone. *J Oral Surg.* 1980;38:613–616.
- [6] del Fabbro M, Rosano G, Taschieri S. Implant survival rates after maxillary sinus augmentation. *Eur J Oral Sci.* 2008;116:497–506.
- [7] del Fabbro M, Wallace SS, Testori T. Long-term implant survival in the grafted maxillary sinus: a systematic review. *Int J Periodontics Restorative Dent.* 2013;33:773–783.
- [8] Esposito M, Grusovin MG, Rees J, et al. Interventions for replacing missing teeth: augmentation procedures of the maxillary sinus. *Cochrane Database Syst Rev.* 2014;5:CD008397.
- [9] Corbella S, Taschieri S, Weinstein R, del Fabbro M. Histomorphometric outcomes after lateral sinus floor elevation procedure: a systematic review of the literature and meta-analysis. *Clin Oral Implants Res.* 2016;27:1106–1122.
- [10] Yaszemski MJ, Payne RG, Hayes WC, Langer R, Mikos AG. Evolution of bone transplantation: molecular, cellular and tissue strategies to engineer human bone. *Biomaterials.* 1996;17:175–185.
- [11] Danesh-Sani SA, Engebretson SP, Janal MN. Histomorphometric results of different grafting materials and effect of healing time on bone maturation after sinus floor augmentation: a systematic review and meta-analysis. *J Periodontol Res.* 2016;52:301–312.
- [12] Miron RJ, Zhang YF. Osteoinduction: a review of old concepts with new standards. *J Dent Res.* 2012;91:736–744.
- [13] Mellonig JT. Donor selection, testing, and inactivation of the HIV virus in freeze-dried bone allografts. *Pract Periodontics Aesthet Dent.* 1995;7:13–22.
- [14] Sogal A, Tofe AJ. Risk assessment of bovine spongiform encephalopathy transmission through bone graft material derived from bovine bone used for dental applications. *J Periodontol.* 1999;70:1053–1063.
- [15] Kolerman R, Goshen G, Joseph N, Kozlovsky A, Shetty S, Tal H. Histomorphometric analysis of maxillary sinus augmentation using an alloplast bone substitute. *J Oral Maxillofac Surg.* 2012;70:1835–1843.
- [16] Mangano C, Perrotti V, Shibli JA, et al. Maxillary sinus grafting with biphasic calcium phosphate ceramics: clinical and histologic evaluation in man. *Int J Oral Maxillofac Implants.* 2013;28:51–56.
- [17] Mangano C, Sinjari B, Shibli JA, et al. A human clinical, histological, histomorphometrical, and radiographical study on biphasic HA-Beta-TCP 30/70 in maxillary sinus augmentation. *Clin Implant Dent Relat Res.* 2015;17:610–618.
- [18] Tosta M, Cortes AR, Correa L, Ddos SP, Tumenas I, Katchburian E. Histologic and histomorphometric evaluation of a synthetic bone substitute for maxillary sinus grafting in humans. *Clin Oral Implants Res.* 2013;24:866–870.
- [19] Ghanaati S, Barbeck M, Detsch R, et al. The chemical composition of synthetic bone substitutes influences tissue reactions *in vivo*: histological and histomorphometrical analysis of the cellular inflammatory response to hydroxyapatite, beta-tricalcium phosphate and biphasic calcium phosphate ceramics. *Biomed Mater.* 2012;7:015005.
- [20] Trindade R, Albrektsson T, Tengvall P, Wennerberg A. Foreign body reaction to biomaterials: on mechanisms for buildup and breakdown of osseointegration. *Clin Implant Dent Relat Res.* 2016;18:192–203.
- [21] Vivian RR, Mecca CE, Bigueti CC, et al. Experimental maxillary sinus augmentation using a highly bioactive glass ceramic. *J Mater Sci Mater Med.* 2016;27:41.
- [22] Smiler DG, Holmes RE. Sinus lift procedure using porous hydroxyapatite: a preliminary clinical report. *J Oral Implantol.* 1987;13:239–253.
- [23] Kolerman R, Samorodnitsky-Naveh GR, Barnea E, Tal H. Histomorphometric analysis of newly formed bone after bilateral maxillary sinus augmentation using two different osteoconductive materials and internal collagen membrane. *Int J Periodontics Restorative Dent.* 2012;32:e21–e28.
- [24] Cammack GV, Nevins M, Clem DS, Hatch JP, Mellonig JT. Histologic evaluation of mineralized and demineralized freeze-dried bone allograft for ridge and sinus augmentations. *Int J Periodontics Restorative Dent.* 2005;25:231–237.
- [25] Valentini P, Abensur D. Maxillary sinus floor elevation for implant placement with demineralized freeze-dried bone and bovine bone (Bio-Oss): a clinical study of 20 patients. *Int J Periodontics Restorative Dent.* 1997;17:232–241.
- [26] Kolerman R, Tal H, Moses O. Histomorphometric analysis of newly formed bone after maxillary sinus floor augmentation using ground cortical bone allograft and internal collagen membrane. *J Periodontol.* 2008;79:2104–2111.
- [27] Scarano A, Degidi M, Iezzi G, et al. Maxillary sinus augmentation with different biomaterials: a comparative histologic and histomorphometric study in man. *Implant Dent.* 2006;15:197–207.
- [28] Jensen OT, Shulman LB, Block MS, Iacono VJ. Report of the sinus consensus conference of 1996. *Int J Oral Maxillofac Implants.* 1998;13:11–45.
- [29] Cricchio G, Sennerby L, Lundgren S. Sinus bone formation and implant survival after sinus membrane elevation and implant placement: a 1- to 6-year follow-up study. *Clin Oral Implants Res.* 2011;22:1200–1212.

- [30] Moon JW, Sohn DS, Heo JU, Shin HI, Jung JK. New bone formation in the maxillary sinus using peripheral venous blood alone. *J Oral Maxillofac Surg.* 2011;69:2357–2367.
- [31] Renno AC, Bossini PS, Crovace MC, Rodrigues AC, Zanotto ED, Parizotto NA. Characterization and *in vivo* biological performance of biosilicate. *BioMed Res Int.* 2013;2013:141427.
- [32] Tadjoedin ES, de Lange GL, Bronckers AL, Lyaruu DM, Burger EH. Deproteinized cancellous bovine bone (Bio-Oss) as bone substitute for sinus floor elevation. A retrospective, histomorphometrical study of five cases. *J Clin Periodontol.* 2003;30:261–270.
- [33] Noubissi SS, Lozada JL, Boyne PJ, et al. Clinical, histologic, and histomorphometric evaluation of mineralized solvent-dehydrated bone allograft (Puros) in human maxillary sinus grafts. *J Oral Implantol.* 2005;31:171–179.
- [34] Proussaefs P, Lozada J, Kim J, Rohrer MD. Repair of the perforated sinus membrane with a resorbable collagen membrane: a human study. *Int J Oral Maxillofac Implants.* 2004;19:413–420.
- [35] 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. *Clinical Oral Implants Res.* 2008;19:796–803.
- [36] Miron RJ, Bosshardt DD. OsteoMacs: key players around bone biomaterials. *Biomaterials.* 2016;82:1–19.
- [37] Rao AJ, Gibon E, Ma T, et al. Revision joint replacement, wear particles, and macrophage polarization. *Acta Biomater.* 2012;8:2815–2823.
- [38] Jensen SS, Bosshardt DD, Gruber R, Buser D. Long-term stability of contour augmentation in the esthetic zone: histologic and histomorphometric evaluation of 12 human biopsies 14 to 80 months after augmentation. *J Periodontol.* 2014;85:1549–1556.
- [39] Bohner M. Resorbable biomaterials as bone graft substitutes. *Mater Today.* 2010;13:24–30.
- [40] Bohner M, Galea L, Doebelin N. Calcium phosphate bone graft substitutes: failures and hopes. *J Eur Ceram Soc.* 2012;32:2663–2671.
- [41] Kunert-Keil C, Scholz F, Gedrange T, Gredes T. Comparative study of biphasic calcium phosphate with beta-tricalcium phosphate in rat cranial defects—a molecular-biological and histological study. *Ann Anat.* 2015;199:79–84.
- [42] Miron RJ, Zohdi H, Fujioka-Kobayashi M, Bosshardt DD. Giant cells around bone biomaterials: osteoclasts or multi-nucleated giant cells?. *Acta Biomater.* 2016;46:15–28.
- [43] Barbeck M, Udeabor SE, Lorenz, et al. Induction of multinucleated giant cells in response to small sized bovine bone substitute (Bio-Oss) results in an enhanced early implantation bed vascularization. *Ann Maxillofac Surg.* 2014;4:150–157.
- [44] Tarnow DP, Wallace SS, Froum SJ, Rohrer MD, Cho SC. Histologic and clinical comparison of bilateral sinus floor elevations with and without barrier membrane placement in 12 patients: Part 3 of an ongoing prospective study. *Int J Periodontics Restorative Dent.* 2000;20:117–125.
- [45] Tawil G, Mawla M. Sinus floor elevation using a bovine bone mineral (Bio-Oss) with or without the concomitant use of a bilayered collagen barrier (Bio-Gide): a clinical report of immediate and delayed implant placement. *Int J Oral Maxillofac Implants.* 2001;16:713–721.
- [46] Fugazzotto PA, Vlassis J. A simplified classification and repair system for sinus membrane perforations. *J Periodontol.* 2003;74:1534–1541.
- [47] Avera SP, Stampely WA, McAllister BS. Histologic and clinical observations of resorbable and nonresorbable barrier membranes used in maxillary sinus graft containment. *Int J Oral Maxillofac Implants.* 1997;12:88–94.
- [48] Zijderveld SA, van den Bergh JP, Schulten EA, ten Bruggenkate CM. Anatomical and surgical findings and complications in 100 consecutive maxillary sinus floor elevation procedures. *J Oral Maxillofac Surg.* 2008;66:1426–1438.
- [49] Sakkas A, Konstantinidis I, Winter K, Schramm A, Wilde F. Effect of Schneiderian membrane perforation on sinus lift graft outcome using two different donor sites: a retrospective study of 105 maxillary sinus elevation procedures. *GMS Interdiscip Plast Reconstr Surg.* 2016;5:Doc11.
- [50] Karabuda C, Arisan V, Ozyuvaci H. Effects of sinus membrane perforations on the success of dental implants placed in the augmented sinus. *J Periodontol.* 2006;77:1991–1997.

How to cite this article: Kolerman R, Nissan J, Rahmanov M, Vered H, Cohen O, Tal H. Comparison between mineralized cancellous bone allograft and an alloplast material for sinus augmentation: A split mouth histomorphometric study. *Clin Implant Dent Relat Res.* 2017;00:1–9. <https://doi.org/10.1111/cid.12518>