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A radiographic evaluation of graft height changes after maxillary sinus floor augmentation with two different grafting materials

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Abstract
Aim: This study was designed to compare vertical dimensional changes in graft height in rabbits treated with two different grafting materials used in maxillary sinus augmentation.
Methods: This study consisted of 12 male New Zealand rabbits, weighing between 2.7 and 3.3 kg each. Twenty-four maxillary sinus floor elevation operations were performed, two on each animal. Each was then repaired with porous titanium granules (PTGs) or a demineralized human bone allograft (DHBA) in putty form. Following the surgery, cone beam tomography (CBCT) was used to determine the height of the grafted maxillary sinuses in the PTGs and DHBA groups (A^0 and B^0, respectively). The animals were sacrificed 6 weeks postoperatively. The augmented sinuses in the PTG and DHBA groups (A^1 and B^1, respectively) were examined 6 weeks postoperatively using CBCT.
Results: The augmented height in A^1 group was higher than that in the B^1 group 6 weeks after the surgery. There was a significant difference in the graft resorption rates of the A and B groups. ($P<0.05$).
Conclusions: In this study, PTG was not resorbed and was able to withstand the air pressure inside the maxillary sinus. Thus, the initial graft height was not greatly reduced at the end of the experiment.

Keywords: Maxillary sinus, bone substitute, dental implant

1. Introduction

Alveolar bone resorption due to the loss of teeth and maxillary sinus pneumatization reduces the distance between the floor of the maxillary sinus and alveolar crest [1]. Maxillary sinus floor elevation (MSFE) is a frequently performed surgical procedure to restore insufficient jaw bone height in the posterior maxilla allowing dental implant placement [2]. Lift procedures have been used in severely resorbed maxillary sinuses to aid the placement of osteointegrated implants. In lift procedures, graft materials are used to contribute to osteogenesis and support the augmented space [3]. Autologous bone is the golden standard for clinical bone augmentation in MSFE, because it has osteoconductive as well as osteoinductive properties, contains osteogenic cells, and does not evoke immunogenic responses. Drawbacks of using autologous bone are, for example, limited availability of bone grafts and morbidity at the donor site [4] Several graft materials are available, among them allogeneic, xenogeneic, and various alloplastic synthetically derived materials. Alloplastic and allogenic bone grafts serve as mechanical spacers, effectively preventing soft tissue in-growth [5] Porous titanium granules (PTGs) are an alternative graft material used to augment osseous defects in maxillofacial surgery. Initially utilized in orthopedics, PTGs were later used to stabilize tibial plateau fractures and prosthetic reoperations in femoral stem fixations [6]. Titanium granules are composed of irregular and porous granules obtained from commercially pure titanium. Theoretically, these granules interlock with one another when implanted, producing a continuous structure [7]. Demineralized human bone allograft (DHBA) is made from ground cortical bone and is rich in osteoinductive proteins [8]. The principal field of use is bone defect and cavity filling. Research aimed at improving the intraoperative handling characteristics of DHBA has resulted in a variety of novel forms, including putty, powder, flex, gel, and paste [9, 10].
The aim of this study was to compare vertical dimensional changes in graft height in a 6-week follow-up of maxillary sinus augmentation in rabbits treated with two different grafting materials: PTGs and a DHBA in putty form.

2. Materials and methods

The study obtained the approval of the Institutional Ethics Review Committee for Animal Research at Ordu University (AU-2011.4.1/12), Turkey. Twelve male New Zealand rabbits, weighing between 2.7 and 3.3 kg each were used in the experiments. Twenty-four maxillary sinus floor elevation operations were performed, two on each animal. These were then repaired using PTGs (group A) or DHBA in putty form (group B). The groups underwent the same surgical procedures and evaluations.

Anesthesia and surgical technique were applied in the same way as the work done by Ayrancı *et al.* [11] Briefly, a midline incision of approximately 50 mm was made at the midline of the nasal dorsum. Two windows were outlined in the nasal bone, using a low-speed round burr then the sinus membrane was released from the floor and lateral and medial walls of the antrum. One sinus was filled with the PTGs (Natix, Tigran Technologies AB, Malmö, Sweden), and the other sinus was filled with DHBA in putty form (Berkeley Advanced Biomaterials Inc. Berkeley, USA) (Figs. 1. A–B). Following surgery, cone beam computed tomography (CBCT) (NewTom FP, Quantitative Radiology, Verona, Italy) was used to determine the height of the grafted maxillary sinuses in the PTG and DHBA groups (A^0 and B^0, respectively), as shown in Figure 2A. All animals received intramuscular ceftriaxone (25 mg/kg) and intracutaneous carprofen (4 mg/kg) twice daily for 3 days. The animals were individually housed at a temperature of 20 ± 1 °C under a light-dark cycle of 12 hours and the humidity was maintained at 50 ± 5%. The animals were given a soft diet and water ad libitum. No postoperative complications were observed during the 6-week observation period and recovery progressed normally in all animals. Six weeks after the surgery, the rabbits were euthanized with high-dose ketamine. The remaining graft height in the PTG and DHBA groups (A^1 and B^1, respectively) was then measured using CBCT (Fig. 2B).

2.1 Tissue preparation

The maxilla was dissected and cut into smaller blocks in the sagittal and coronal planes, including the nasal and maxillary sinus, then fixed in the same solution for 48 hours at 4ºC, and decalcified with nitric acid. Specimens augmented with allograft in putty form were then embedded in paraffin and sliced into 5-µm sections and stained with hematoxylin-eosin.

2.2 Scanning electron microscopy

Augmented sinuses with porous titanium granules were stored at room temperature to dry, then mounted on metallic stubs, sputter-coated with gold, and examined using spectrum electron microscopy (SEM) (EVO LS10, Zeiss, Germany). SEM photomicrographs were taken at magnifications of X20 to assess the quality of the augmented spaces in the maxillary sinus (Fig. 3A).

2.3 Histomorphometric analysis

Measurements were taken from decalcified specimens using a personal computer-based image analysis system (Stereo-Investigator 7.0, USA). From the serial sections collected from each sample, four sections were randomly selected and analyzed manually. (Fig. 3B).

2.4 Statistical Analysis

The mean and standard deviation of the vertical height of the maxillary bone were assessed immediately after surgery and at the final follow-up observation in each group. The Student's independent *t*-test was employed to analyze differences in the resorption height of the grafted bone within the maxillary sinus according to the bone graft material. Significance was accepted at $P<0.05$. SPSS software (SPSS for Windows, release 11.0 versions, Copyright SPSS inc., NY. 2002) was used for the statistical analysis.

3. Results

In the PTG group (group A), the vertical height of the augmented graft material was 3.51 ± 0.25 mm immediately after the surgery (A^0) and 3.083 ± 0.213 mm at the final observation 6 weeks later (A^1). In the DHBA putty group (group B), the vertical height of the augmented graft material was 3.40 ± 0.23 mm immediately after the surgery (B^0) and 2.01 ± 0.50 mm at the final observation 6 weeks later (B^1) (Table 1). The augmented height in the PTG group (A^1) was higher than that in the DHBA group (B^1) 6 weeks after the surgery (Table 1). There was a significant difference in the graft resorption rates of the PTG and DBM groups (Table 2; $P<0.05$).

4. Discussion

Maxillary sinus floor augmentation is a feasible therapeutic procedure for increasing the height of the posterior maxilla in cases where there is inadequate bone for the placement of osteointegrated implants [12]. Various grafting materials have been used for sinus-floor augmentation, yielding different results [13-15]. Among these materials, autogenous bone graft is considered the best and most reliable for sinus augmentation, although its use is restricted by donor site morbidity, limited availability, and uncontrolled resorption, as concluded by previous studies [16, 17].

Animal models have been widely used in dental research to investigate the effects of different treatment modalities and to test materials, such as bone grafts [14, 18, 19]. The rabbit experimental model of maxillary sinus augmentation was first introduced by Watanabe *et al.* [20]. They stated that the advantages of the rabbit model were low cost, ease of experimentation, and easy distinction of membrane perforation. In addition, the ventilation system of rabbits is the same as that of humans, and rabbits have a well-defined ostium opening to their nasal cavities. Air pressure measurements of the nasal cavity and maxillary sinus with potent ostium are similar to those in humans for absolute pressures and synchronicity with respiratory cycle [21, 22].

For maxillary sinus augmentation graft materials to be practical, the structure should be able to withstand pressure caused by sinus pneumatization, especially in two-stage surgery. Previous studies reported that when uninterrupted pressure was applied to the grafting material, the air pressure in the maxillary sinus affected the sinus mucosa, resulting in changes in the healing of the augmented bone and bone structure [1, 23]

Asai *et al.* [1] reported that in rabbits without ostial occlusion, after 6 weeks, the augmented space was completely filled with an enlarged air space. They concluded that when present for long periods, such sinus air pressure had adverse effects on the mucous membrane. Therefore, in the present study, the experimental period lasted 6 weeks, and the animals were then sacrificed.

Sun *et al.* [14] reported that air pressure created movement in the maxillary sinus mucosa and that this movement, to which grafting materials in the sinus are continuously subjected, affected the augmented bone healing process and bone structure.

A previous study reported that repneumatization of the maxillary sinus was likely to occur within the first 2–3 years after surgery [24]. Johansson *et al*. [23] reported that in the first 6–7 months after sinus augmentation procedures using autologous grafts, the graft was resorbed at a rate as high as 47%.

This study assessed vertical dimensional changes in a rabbit model subjected to maxillary sinus floor elevation procedures and then treated with two different grafting materials: PTGs and DHBA in putty form (Table 1).

Wohlfahrt *et al*. [6] studied the *in vivo* biological performance of PTGs in a tibial defect model in rabbits. They show that titanium had thrombogenic properties and therefore may result in increased osteogenesis due to the release of higher levels of growth factors into blood clots. In another study Jonsson and Miyöberg [25] demonstrated in clinical trials that porous titanium granules are more useful than autograft bone to fill the void created by reducing a collapsed fracture of the lateral tibial plateau.

In a recent pilot study, PTGs were used in sinus augmentation procedures in humans. In that study, Bystedt and Rasmusson [26] reported that titanium granules appeared to be a good bone graft substitute in sinus augmentation procedures.

Delgado-Ruiz *et al*. [27] reported that PTG particles must be covered by a membrane, especially when grafting larger defects because many particles were mobilized outside the grafted site. However, in many studies in the literature [14, 18, 20], in common with our study, the graft materials used in maxillary sinus augmentation were not protected by membranes.

Dursun *et al*.[28] reported that the findings of their studies suggest that PTG, a porous, permanent, nonresorrective bone substitute, may have a beneficial osteoconductive effect on the mechanical strength of the new bone formed in the augmented maxillary sinus.

Verket *et al*. [29] in their study compared to PTG and demineralized bovine bone mineral (DBBM) in experimental narrow marginal peri-implant bone defects with respect to early bone healing and implant stability. Showed a more coronal bone-implant contact in the DBMM group and exhibited more bone fill compared to the WPTG group and reported that the better mechanical properties observed for WPTG are negligible for early stability and osseointegration of implants.

Some studies showed that the morphological properties of PTGs seemed to aid bone formation [30, 31]. Turner *et al*. [32] reported that the characteristics of PTGs promoted blood clotting, resulting in bone cell proliferation, with increased numbers of platelets caused by clotting serving as a natural source of osteogenic molecules. DHBA contains active proteins, such as bone morphogenetic protein, transforming growth factor-beta, insulin-like growth factor, and fibroblast growth factor, all of which are indirectly involved in the bone healing process [33]. DHBA in putty form has osteoinductive and osteoconductive properties, with good handling capabilities. As a result, it has become increasingly popular in maxillary sinus augmentation [34]. But, in our study, the graft material can not with-stand sinus air pressure and not maintain the augmented space.

The present study mimicked two-stage surgery, in which there is no primary placement of implants and in which the required bone elevation is produced by raising the maxillary sinus base with graft materials. The PTGs were not resorbed in our study. Thus, the graft materials were able to withstand the air pressure inside the maxillary sinus, and they exhibited little loss in initial height at the end of the experiment (Table 2). Despite its osteogenic and osteoconductive properties and ease of application, the DHBA in putty form graft material failed to maintain the height augmentation achieved initially and was largely resorbed (Table 2). The results of the statistical analysis of the two graft materials showed that the initial and final values obtained using the PTGs were better than those achieved with the DHBA in putty form ($P<0.05$).

Table 1: Mean graft height (and SD) in millimeters in vertical graft height.

	Grafts	n	Graft Height Mean (mm)	SD
A^0	PTGs	12	3.51	0.25
B^0	DHBA	12	3.40	0.23
A^1	PTGs	12	3.08	0.21
B^1	DHBA	12	2.01	0.50

^{0} time graft augmentation
^{1} 6 weeks after sinus augmentation
A: PTGs, B: DHBA
PTGs: Porous titanium granules
DHBA: Demineralized human bone allograft
SD: Standard Deviation

Table 2: Mean decrease (and SD) in millimeters in vertical graft height

	n	Mean (mm)	SD
A^0-A^1	12	0,42	0,23
B^0-B^1	12	1,39	0,46

$P<0.001$
A^0-A^1: Mean decrease in milimeters in vertical graft height from immediately after surgery to end of experimental period for PTGs.
B^0-B^1: Mean decrease in milimeters in vertical graft height from immediately after surgery to end of experimental period for DBM.
PTGs: Porous titanium granules
DHBA: Demineralized human bone allograft
SD: Standard Deviation

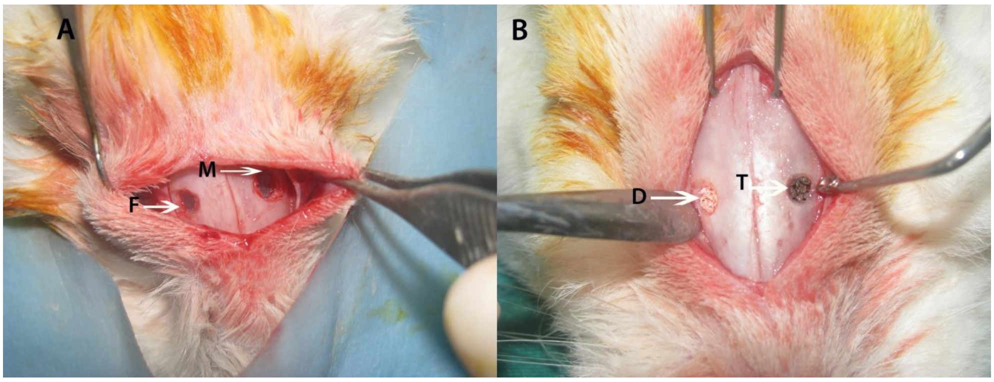

Fig 1A: Bony defects were made by round bur on the maxillary sinus anterior wall. **1B.** The defect sites were filled with grafting materials. *(F: Fenestration, M: Maxillary Sinus, D: Demineralized bone matrix, T: Porous titanium granules)*

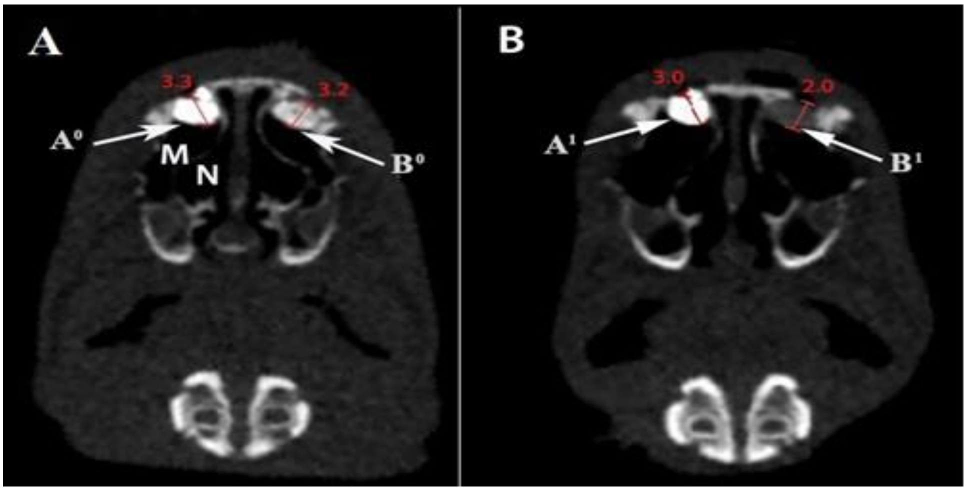

Fig 2A: Measurement of graft height from cone-beam computed tomography scan and comparison between groups using vertical sections at the immediately after surgery. **2B.** Measurement of graft height from cone-beam computed tomography scan and comparison between groups using vertical sections at the end of experimental period. *(M: Maxillary Sinus, N: Nose, A^{0-1}: Vertical heights of Porous titanium granules, B^{0-1}: Vertical heights of Demineralized human bone allograft)*

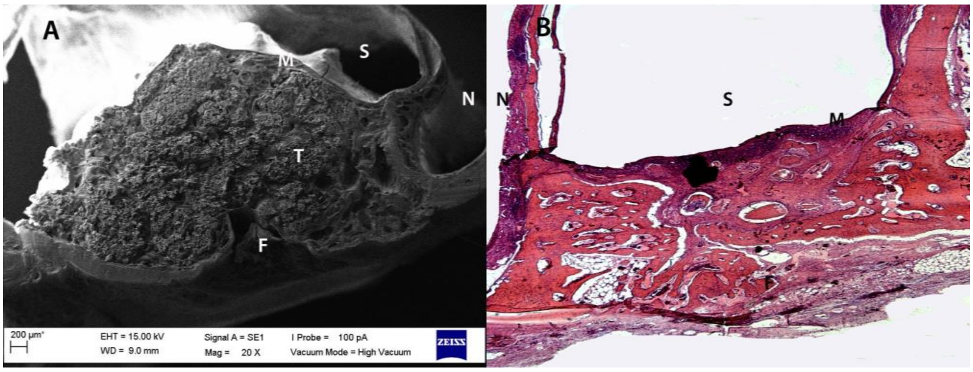

Fig 3A: Scanning electron microscopic images shows grafted area at the end of the experimental period. **3B.** Photomicrograph shows grafted region at the end of experimental period (x20) *(F: Fenestration, M: Maxillary Sinus Membrane, S: Maxillary Sinus, N: Nose, T: Porous titanium granules)*

5. Conclusions

PTGs were able to withstand the air pressure inside the maxillary sinus. Thus, at the end of the experiment, there was little loss in the initial increase in graft height achieved by the augmentation procedure.

6. Acknowledgments

The English in this document has been checked by at least two professional editors, both native speakers of English. For a certificate, please see:
https://www.scribendi.com/tracking?o=388563&k=vfTynXLxcVVc

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