**UNIVERSIDAD DE SEVILLA**

FACULTAD DE ODONTOLOGÍA

DOCTORADO DE CIENCIAS DE LA SALUD

# BIPHASIC CALCIUM PHOSPHATE WITH OR WITHOUT HYALURONIC ACID VS. DEPROTEINIZED BOVINE BONE MINERAL FOR MAXILLARY SINUS AUGMENTATION. A RANDOMIZED CLINICAL TRIAL.

Tesis Doctoral

**NICOLA ALBERTO VALENTE**

Sevilla, 2021

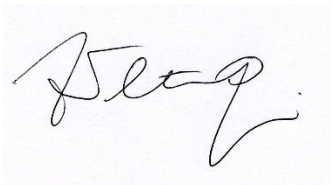
Sevilla, Enero de 2021

EUGENIO VELASCO ORTEGA, Profesor Titular de Odontología Integrada de Adultos y Gerodontología de la Facultad de Odontología y Director del Máster de Implantología Oral de la Universidad de Sevilla.

ANTONIO BARONE, Profesor Titular del Departamento de Cirugía de la Facultad de Odontologia de la Universidad de Pisa.

CERTIFICAN:

Que D. NICOLA ALBERTO VALENTE, inscrito en el programa de Doctorado de Ciencias de la Salud de la Universidad de Sevilla, ha realizado bajo su tutela y dirección el trabajo de investigación titulado **Biphasic calcium phosphate with or without hyaluronic acid vs. deproteinized bovine bone mineral for maxillary sinus augmentation. a randomized clinical trial**, que consideramos satisfactorio para optar al título de Doctor en Odontología**.**



Prof. Velasco Ortega Prof. Antonio Barone

# Table of contents

# 1. Introduction

The implant-prosthetic rehabilitation of the atrophic maxilla requires the creation of sufficient volumetric quantity of bone, through regeneration techniques, necessary to position the implants.

Although the purpose of the regenerations is the same in the upper and lower jaws, the techniques, and above all their predictability, vary greatly[1, 2]. Within this wide variability, the balance probably hangs in favor of the upper posterior maxilla[1]. Maxillary sinus augmentation (MSA) is probably the most predictable and best performing technique.[3-6]

The maxillary sinus floor elevation technique with lateral approach has been used in implant surgery for almost 40 years by now with high predictability of success. It was first described by Tatum in 1976, but first published by Boyne and James in 1980.[7, 8] In the first description of the technique present in the literature the sinus elevation was used not only to allow implant placement but, in 11 of the 14 cases reported, it was used in order to be able to reduce the posterior ridge, thus augmenting the inter-arch space and provide the patients with a conventional prosthesis.[8]

The technique has been further developed and modified during the years and its indications and directions have been refined by several authors. We distinguish now between a one stage procedure, with simultaneous insertion of the implants, or a two stage procedure, when the residual bone height is less then 4mm, but the basic procedure has essentially remained the same as was firstly described.[9]

Despite the wide spread use of this surgical technique and its relative safety there are several variables and possible complications that must be

taken into consideration in the planning and during the surgical procedure itself. One of these is the blood supply and vascularization of the sinus cavity and Schneider membrane and, in particular of the lateral maxillary wall, which is of crucial importance as a source of blood supply for our graft material and because the accidental severing of a vessel during the antrostomy can be, in some case, a significant intraoperatory complication.

This technique, however, requires that the clinician must have an absolute mastery of the maxillary sinus' anatomy before performing a sinus lift to avoid inconveniences that can prolong times of realization or even postpone execution, in rare cases. A large-caliber antral alveolus artery, for example, can cause intense bleeding, while the presence of a septum can cause the tearing of a sinus membrane.[10-12]

### 1.1. Anatomy of the maxillary sinus

The maxillary sinus is the largest of the paranasal sinuses, which also include the frontal, sphenoid and ethmoidal sinuses.

It usually occupies a large part of the body of the maxillary bone; it develops between the second and third month of intrauterine life, with a volume of 0.1-0.2 cubic centimeters at the time of birth. In fact, its size and its degree of pneumatization increase with the eruption of the first permanent elements, until it matures at the end of the adolescence period. A further widening can come with the persistence, on the medium/long term, of an edentulism in correspondence of the area of the floor of the sinus.

Classically, the maxillary sinus is described as a pyramidal pneumatic cavity. The base corresponds to the lateral wall of the nasal cavity, its upper part is where the opening of the maxillary sinus into the nasal meatus is located; There is also a mesio-buccal wall, depressed in correspondence with the canine fossa, a superior one which forms the floor of the orbit, a posterior one facing the anterior wall of the pterygomaxillary fossa. The last wall, as already mentioned, partly corresponds to the upper alveolar process and partly to the hard palate; in a dentate adult this is the most solid of the bony walls, even though it has recesses in correspondence with the roots of premolars and first molars; these will undergo a thinning with the progress of the individual's age.

The blood supply of the maxillary sinus is provided by three branches of the Maxillary Artery (MA): the Greater Palatine Artery, the Infraorbital Artery (IOA), and the Posterior Superior Alveolar Artery (PSAA). The PSAA originated from the MA while it passes through the pterygopalatine fossa. It descends to penetrate the maxillary tuberosity to give terminal alveolar and dental branches that supply blood for the posterior superior teeth, gingiva, and lining of the antrum. The IOA courses along the infraorbital groove and canal after its origin from the MA, then it exits from the infraorbital foramen on the facial aspect of the maxilla. Along the course through the infraorbital canal, the IOA gives origin to superior anterior alveolar branches that provides blood supply to the anterior teeth and lining of the antrum.[11, 13]

Usually the PSAA and the IOA form anastomoses inside and outside the bony lateral antral wall that supplies the Schneiderian membrane and the epiperiosteal vestibular tissues. According to the literature, an intraosseous

anastomosis is constantly present, while an extraosseous one is present in about 44% of the cases.[14, 15] Of particular importance is the intraosseous anastomosis, which is also called Alveolar Antral Artery (AAA). It was first described in 1934 and it passes through the area where the bony window is most frequently opened during sinus elevation.[16]

## 1.2. Biomaterials

In addition to being aimed at obtaining the quantity of bone necessary for implant positioning, the regenerative techniques should also allow gaining adequate bone quality, suitable not only for receiving the implant in its volume, but also for giving adequate stability immediately and over time. The biomaterial used can be a key factor in determining the quality of the regenerated bone. In general, there are two types of materials that can be used in MSA techniques: autogenous bone and bone substitutes, with the latter further distinguishable into allogeneic, xenogenic or synthetic. The exclusive use of autologous bone for MSA provides for the removal of large quantities of bone, with the need to open a second surgical site and a significantly increased morbidity and consequent discomfort of the patient. Bone substitutes avoid this inconvenience and, very often, are used in combination with limited quantities of autologous bone, taken in the proximity of the area to be regenerated, so as not to give up the osteogenic qualities of the latter. The bone substitute of choice in the MSA, perhaps because the most documented in the scientific literature, is the anorganic bovine bone mineral (ABBM).

There is a large amount of studies in the literature analyzing different materials used in the MSA procedure, in several of these studies different

biomaterials are often used in combination with others. For example, ABBM is often used with autologous bone (AB) or other biomaterials, calcium phosphate, in addition to being often used in combination with other materials, is sometimes used in its form of beta tricalcium phosphate (BCP) alone or, as in our study, mixed with hyaluronic acid (HA).[17]

The osteoconductive properties of ABBM and BCP have been widely documented with success for MSA in the literature. The BCP graft is used with a 90/10 ratio of beta-tricalcium phosphate and hydroxyapatite and has an osteoinductive potential demonstrating ectopic bone formation in skeletal sites in a rat muscle.[18]

In vitro and in vivo studies have demonstrated the possibility for HA to stimulate cell differentiation and tissue remodeling in the context of osteoblastic activity.[19-21] An in-vivo study from Sasaki also demonstrated the osteoinductive capacity of HA.[22]

Several studies have shown high implant survival rates following the utilization of bone substitutes in sinus grafting procedures.[6] BCP is an osteoconductive material that acts as a scaffold for new bone formation during the graft maturation period. Some authors found that the use of β-TCP as a bone substitute for repairing the alveolar cleft bone defects was successful.[23]

ABBM has been widely used for SA and has been proved to be very efficient for this procedure in comparison to AB.[24] This material has shown great osteoconductive properties and is able to maintain significant volume

of the augmented bone throughout the healing process and no more complications than with the use of AB were reported.[24]

The use of HA in addition to allogenic inorganic bone was described for SA procedure with good success rates in comparison to other biomaterials.[25]

Due to the thermal treatment to which they undergo, the mineral structure of animal derived bone grafts such as ABBM is crystalline, thus implying that it only dissolves very slowly under physiological conditions, way more slowly than the autogenous bone.[26] The extremely slow resorption rate of ABBM causes the permanence of graft granules even after a very long period thus increasing the potential infection risk.

TCP and hydroxyapatite with a ratio of 90%/10% might be a good alternative mixing the slow resorption rate of hydroxyapatite which guarantees the stability while the relatively fast resorption rate of TCP create an ideal environment for new vital bone formation.

HA has been shown to stimulates post-operative neoangiogenesis in surgical wounds, thus significantly accelerating the healing process.[27, 28] Also Hyaluronic acid has a bacteriostatic effect on pathogens commonly found in gingival lesions and periodontal wounds.[29, 30] Application of HA during the surgical therapy may reduce bacterial contamination of surgical wound sites hence decreasing the risk of postsurgical infection and promoting more predictable regeneration.[29]

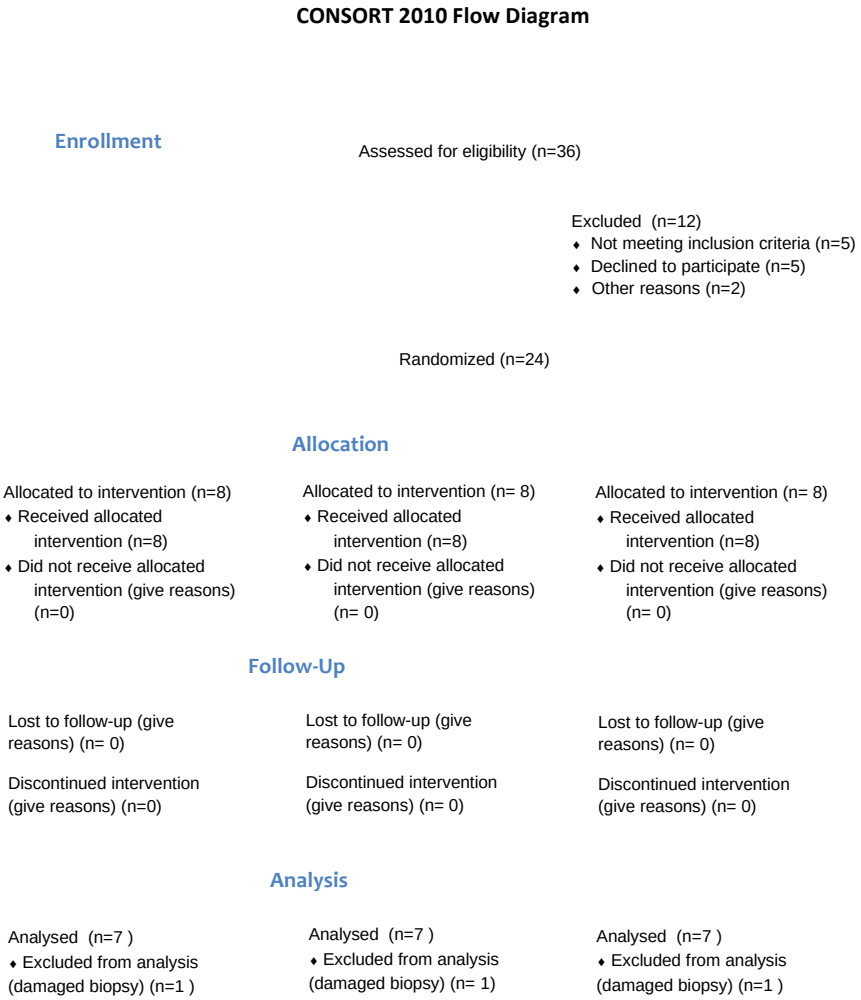
## 1.3. Aim

The present randomized controlled study aims to evaluate and compare, histomorphometrically and clinically three different bone substitutes such as: ABBM, Tricalcium Phosphate (TCP) with or without the addition of hydroxyapatite that were used for lateral MSA. This study population will be followed until the 3rd year. This study describes histological differences between the three groups at 9 months.

# 2. Patients and methods

## 2.1.Study Design

The present study was prepared in agreement with the CONSORT statements for improving the quality of reports of parallel group randomised trials (http://consort-statement.org/).




The patients were recruited, between march 2018 and February 2019 at the Dental School of the University of Geneva, Switzerland. All the patients

understood and signed an informed consent form for being enrolled in this trial. All patients received a staged approach, namely a MSA was first performed and after 9 months of healing the implants were inserted. The principles of the Declaration of Helsinki, as revised in the 2004, for biomedical research involving human subjects, the ICH-GCP or ISO EN 14155 (as far as applicable) as well as all national legal and regulatory requirements were followed; moreover, the study design was approved by the Ethical Committee of Geneva, Switzerland (Approval number: 2018-01183) and was registered in the database of the NIH ClinicalTrials.gov (NCT04506827).

All patients who required lateral MSA, who had 3 mm or less of residual bone crestal height in the posterior areas, who were 18 years old or older, who were able to understand and sign a consent form - were eligible for this study.

The exclusion criteria were:

- full contraindication to implant surgery,
- systemic diseases that could negatively influence wound healing;
- heavy smokers (more than 10 cigarettes/day);
- head and neck irradiation treatment;
- uncontrolled diabetes;
- chronic or acute sinus pathology;
- uncontrolled periodontal disease;
- full mouth plaque and bleeding score higher than 25%,
- tooth extractions in the previous 2 months

## 2.2.Randomization, allocation concealment, blinding, calibration

Randomization sequence was obtained using a random permutation sequence generator (Statistics Toolbox, MatLab 7.11, The MathWorks, Natick, MA) without any kind of minimization of confounding factors such as age, smoking habit, gender and augmentation area.

## 2.3.Primary and secondary outcomes

Primary outcome**:** Histomorphometric parameters of the augmented bone.

Secondary outcomes:

- Mean bone gain measured at 9 months through CBCT evaluation
- Intraoperative and post-operative complications
- Implant insertion torque measured in Ncm
- Early implant failure
- Patient related outcome measures (PROMs)

## 2.4.Interventions

### 2.4.1. Sinus augmentation

All the patients enrolled in the study received a session of oral hygiene and a periodontal examination before the surgical procedure to obtain a more favorable oral environment for wound healing. A CBCT was mandatory for all included cases to verify that the maxillary sinus was clear and that the residual bone height was 3 mm or less. A voxel size of 0.125 mm, with a set

of the parameters to 8.0 mA, 80 kV and an exposure time of 12-18 s were applied. CBCT radiographic measurments were carried out by GD following the protocol described in a series of studies in order to standardize the procedure.[31-34] All the enrolled patients received 2 gr. of amoxicillin and clavulanic acid (or 600 mg. of clindamycin for those who were allergic to penicillins) as a pre-medication one-hour before surgery. All patients rinsed for 1 minute with 0.2% chlorhexidine mouthwash (and twice a day for the following 3 weeks). Local anesthesia was administered, a mid-crestal incision with mesial and distal release were performed to access the lateral bone wall of the maxillary sinus, subsequently, ultrasound bone surgery (Piezosurgery ®, Carasco, Italy) with specific tips was used for the bone window osteotomy (Figure 1). The Schneiderian membrane was reflected and lifted up medially with flat sinus curettes. Once the sinus membrane was completely lifted a bioabsorbable pericardium membrane (Smartbrane, Regedent AG, Zurich, Switzerland) was applied to protect it (Figure 2).

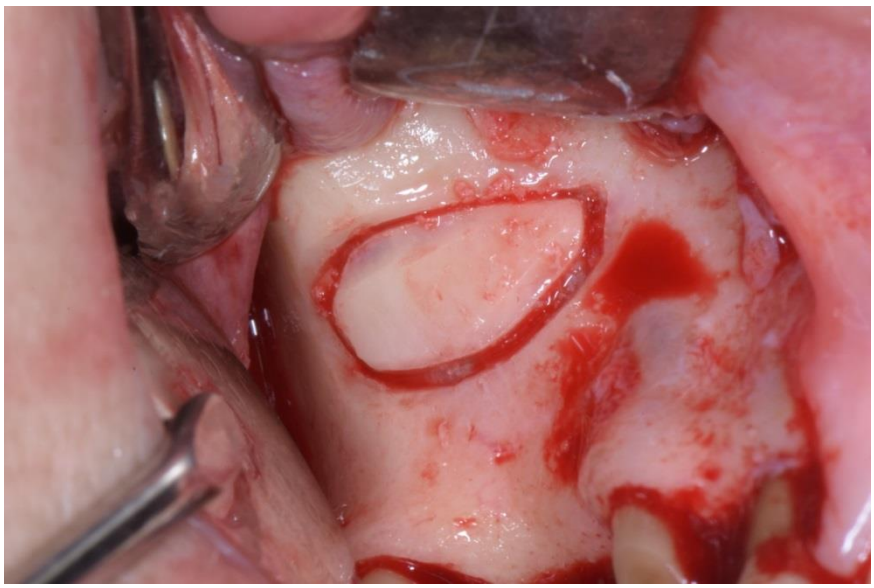


Figure 1: Bone window created with the aid of piezosurgical inserts

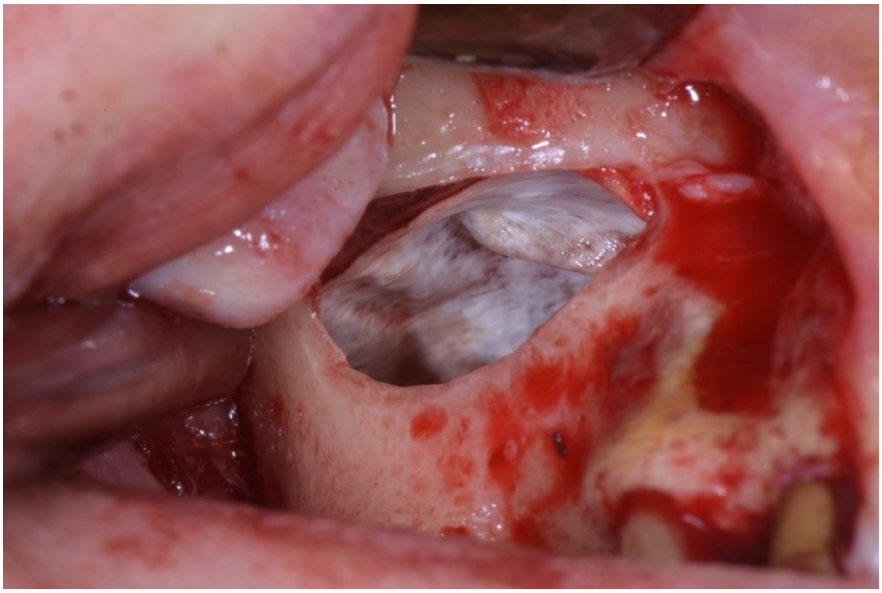


Figure 2: Sinus membrane elevated and protected with a bioabsorbable membrane

The randomization sealed envelopes were opened and the clinician allocated the patients to one of the three experimental groups:

1) Control group that received Demineralized Bovine Bone Mineral (Bio-Oss Cancellous, Geistlich, Wolhunsen, Switzerland);
2) Test group 1 that received TCP with particle size ranging from 250 to 1000 µm (Osopia, Regedent, Zurich, Switzerland);
3) Test group 2 that received TCP as in test group1 plus crosslinked Hyaluronic Acid (Hyadent BG, Regedent, Zurich, Switzerland) with a ratio 2 to 1.

The sinus was grafted with the biomaterial corresponding to the group, the bony window was repositioned, stabilized and covered with a resorbable pericardium membrane (Figures 3, 4).

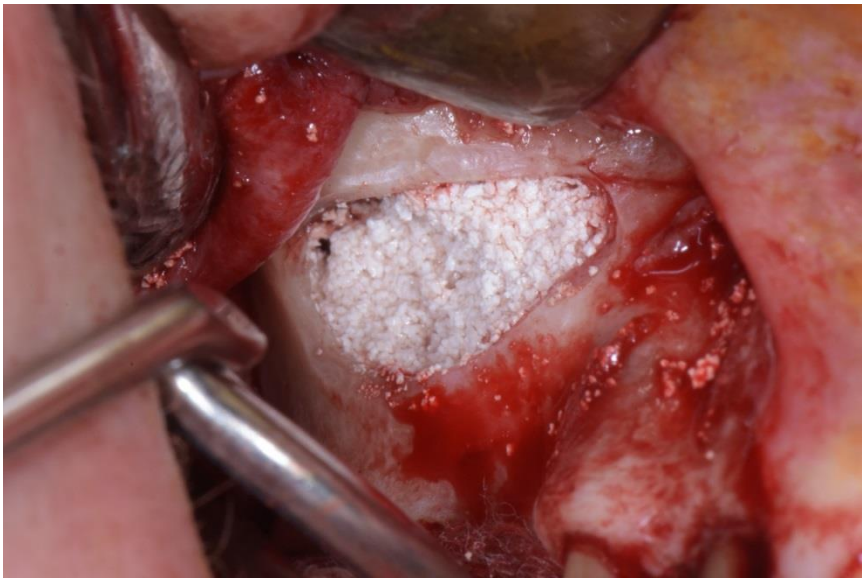


Figure 3: Sinus grafted with biomaterial

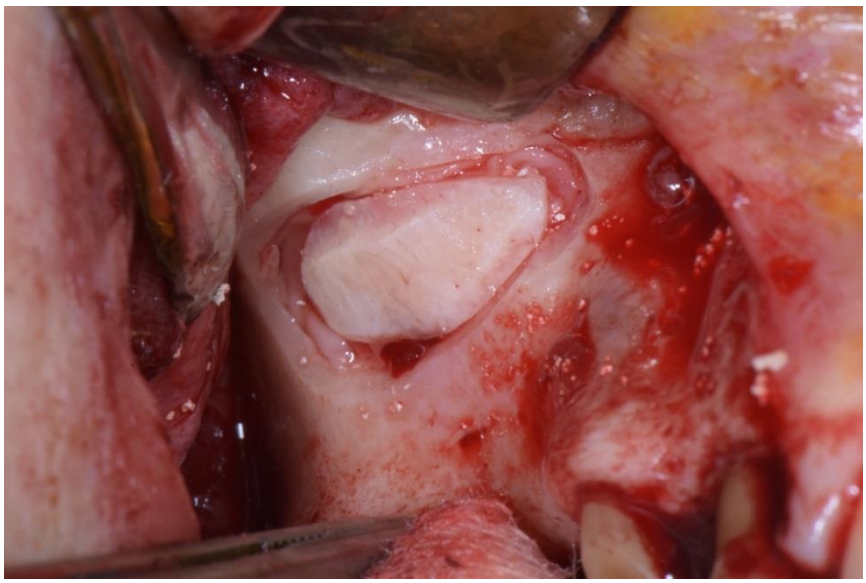


Figure 4: Bone window repositioned after completion of grafting

All patients were prescribed 1gr amoxicillin + clavulanic acid twice daily for 7 days (for those who were allergic to penicillin 300mg clindamycin 3 times daily for 7 days) as post-operative prophylaxis. All patients received 8mg of dexamethasone immediately after surgery and 4 mg of dexamethasone per day were prescribed for the following 5 days. Analgesics

were prescribed (1gr paracetamol 3 times daily) according to patients' need. The use of removable temporary restorations was not recommended during the healing period.

### 2.4.2. Implant placement

Nine months after the MSA a CBCT analysis was required to evaluate the total bone height gain.

Depending on the treatment plan, between one and three implants, bone level (BL) or tissue level (TL), of two different manufacturers (Institut Straumann AG, Basel, Switzerland and Sweden & Martina, Due Carrare, Italy) were positioned. While preparing the osteotomy implant site a bone biopsy was harvested using a trephine. The implant insertion torque was measured in Ncm for each implant.

### 2.4.3. Restorative phase and maintenance

After an osseointegration period of 3 months, the implants were restored. The prosthetic procedures were similar for all groups, impressions were taken with a polyether rubber material (Impregum®, Espe Dental AG, Seefeld, Germany) and a customized impression tray. Final Zirconia screw-retained restorations were delivered after a period of 2/3 weeks, peri-apical radiographs were taken and oral hygiene instructions were given to all participants in the study.

## 2.5. Patient reported outcome measures (PROMs)

Two weeks after MSA all patients answered a questionnaire regarding the oral health-related quality of life using the short-form Oral Health Impact Profile (OHIP-14).[35] This questionnaire consists of 14 questions each one formulated in the following way: "How often (impact item) because of problems with your teeth, mouth or dentures?" The impact items belonged to 7 different impact domains: 1. Functional limitations; 2. Physical pain; 3. Psychological discomfort; 4. Physical disability; 5. Psychological disability; 6. Social disability; 7. Handicap. Each question could be answered with a score from 0 to 4 corresponding to: 0 = never, 1 = hardly ever, 2 = occasionally, 3 = fairly often, and 4 = very often.

### 2.6. Complications

Possible complications were recorded at different time points of the study. Failure of the implant was considered as: any mobility of the implant, any infection that required removal, or any implant fracture. Implant success criteria were chosen according to Buser et al. 1990.[36]

### 2.7. Histologic procedures

Twenty-four bone biopsies, obtained from 24 maxillary sinuses, were fixed by immediate immersion in 10% buffered formalin and processed (Precise 1 Automated System; Assing, Rome, Italy) to obtain thin ground sections. The specimens were dehydrated in an ascending series of alcohol rinses and embedded in glycol-methacrylate resin (Technovit 7200 VLC; Kulzer, Wehrheim, Germany).

The specimens were sectioned, along their longitudinal axis, with a high precision diamond disk at about 150 µm and ground down to about 30 µm

with a specially designed grinding machine Precise 1 Automated System. Three slices were obtained from each specimen, subsequently stained with acid fuchsin and toluidine blue before the analysis. Histological analysis was carried out using a light microscope (Laborlux S, Leitz, Wetzlar, Germany) connected to a high-resolution video camera (3CCD, JVCKY-F55B, JVC, Yokohama, Japan) and interfaced with a monitor connected to a computer. This optical system was associated with a digitizing pad (Matrix Vision GmbH, Oppenweiler, Germany) and a histomorphometry software package with image capturing capabilities (Image-Pro Plus 4.5, Media Cybernetics Inc., Immagini & Computer Snc, Milano, Italy).

One single well-trained examiner, who was not involved in the surgical treatment, evaluated the histological results. Percentages of newly formed bone, marrow spaces and residual graft particles were reported.

### 2.8.Statistical analysis

Power analysis was employed to determine the sample size by using a 0.05 significance level and a power of 90%, based on the results reported in previous review paper concerning histomorphometric outcomes.[37] Sample size from 3 to 5 per treatment arm was required to detect significant difference between the bone substitute material groups by using secondary outcomes, that is, percentage of residual biomaterial and connective tissues. Sample size was increased with a factor 1.25 for possible drop-out of subjects. Final sample size per group ranged from 4 to 6 subjects.

Descriptive statistics were calculated, including means, standard deviations, medians, and confidence intervals. Age, baseline bone height, insertion torque, complications, bone gain and PROMs scores were analyzed

using the one-way analysis of variance (ANOVA) test, while the statistical analysis of the histomorphometric values for new bone, marrow spaces and residual graft particles was made using one-way multivariate analysis of variance (MANOVA). The level of significance was established at 5% (P = .05), and the analysis was carried out using statistic soft- ware (SPSS version 26, IBM).

# 3. Results

Thirty-six patients were considered eligible, 12 patients were excluded from the study for the following reasons: 5 patients refused to give their consent to participate in a randomized clinical trial, 3 patients had a thickening of the sinus mucosa that required additional examinations, 2 patients were under treatment with Direct Oral Anticoagulants and 2 patients after being enrolled and treated had to move to another city due to their job.

Baseline values for age, gender, initial bone height and number of implants placed are summarized in table 1.

Table 1 – Baseline parameters

| Parameter | Test 1 (BCP) | Test 2 (BCP + Hyaluronic acid) | Control (ABBM) | Group comparison |
|---|---|---|---|---|
| Age (mean ± SD) | 57.63 ± 13.97 | 60.63 ± 11.21 | 49.5 ± 11.28 | P = 0.194 |
| Gender ratio (m/f) | 3/5 | 3/5 | 3/5 | |
| Initial bone height (mm) ± SD | 2.5 ± 0.38 | 2.63 ± 0.23 | 2.69 ± 0.46 | P = 0.591 |
| Total implants | 16 | 13 | 15 | |

## 3.1. Primary Outcomes

### 3.1.1. Histological results

Three biopsy samples were not analyzed (one for each group) because the biopsies were damaged during their removal from the trephine. A total of 21 biopsies were examined, specifically 7 for each group.
All biopsy samples at low magnification showed a certain amount of new bone formation.

#### 3.1.1.1. Control (ABBM)

All samples, belonging to the control group, showed two different portions: the preexisting bone could be seen at the bottom of the samples (crestal portion), while at the top of the sample (apical portion) some residual particles can be observed (figure 5). Residual graft particles (RGP) were completely surrounded by newly formed bone in the area close to the preexisting bone with a thickening the cortical bone layer (figure 6), while the particles were only partially surrounded by new bone in the areas located more apically. Notably, the most apical portion of the samples showed less newly formed bone with a predominance of non-mineralized tissue between the residual graft particles (figure 7).

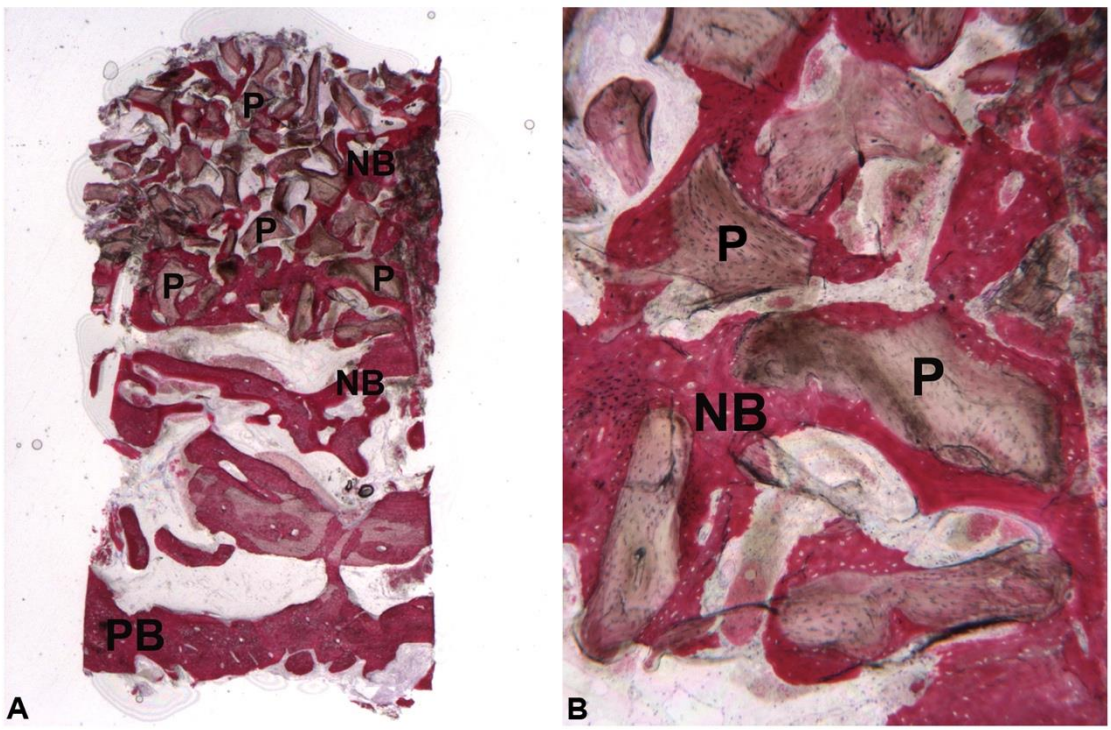



Figure 5: A) Control. Light microscopic ground sections of the samples showed the residual pre-existing bone (PB) and the regenerated area consisting of newly formed trabecular bone (NB) and residual biomaterial particles (P).
B) At higher-power magnification, the biomaterial particles (P) facing to pre-existing bone were surrounded by newly formed bone (NB). (Acid fuchsin-Toluidine blue 9X and 40X).

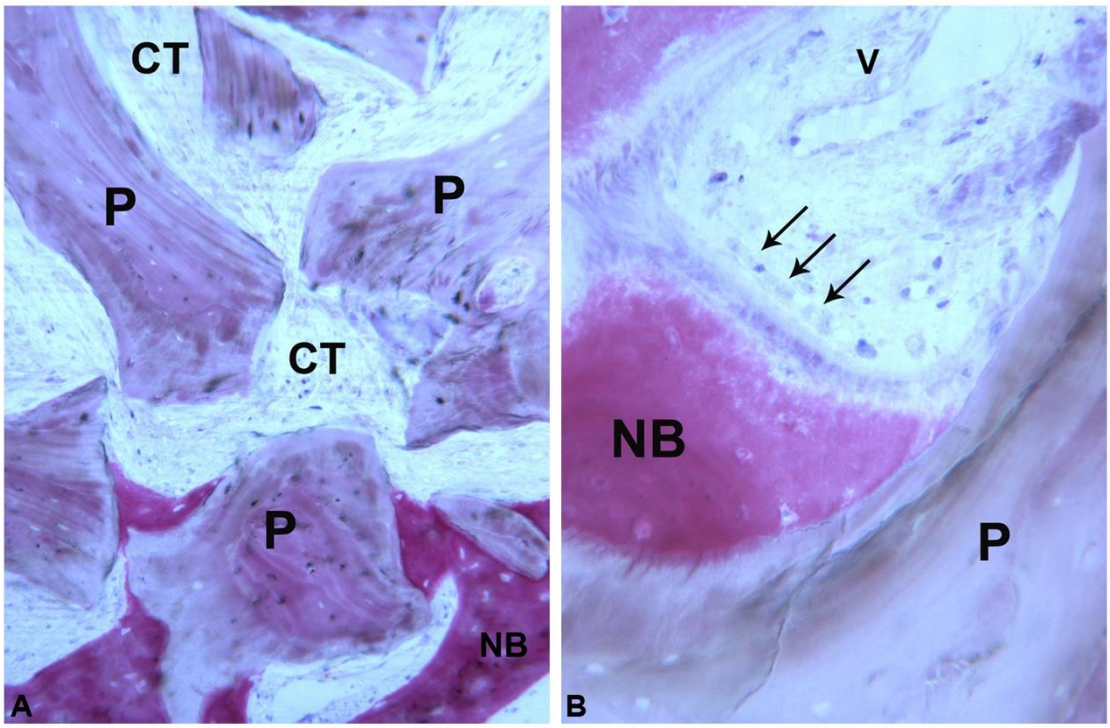



Figure 6: A) Control. Newly formed trabecular bone (NB) in the bone regenerated area was present. A portion of the residual biomaterial (P) was lined by new bone while the remaining part, was incorporated by the connective tissue (CT).
B) Control. Osteoblasts covering the woven bone (NB) close to the biomaterial (P) were seen, and microvessels (V) appeared in the tissue in both the bone and the particles.
(Acid fuchsin-Toluidine blue 100X and 200x).

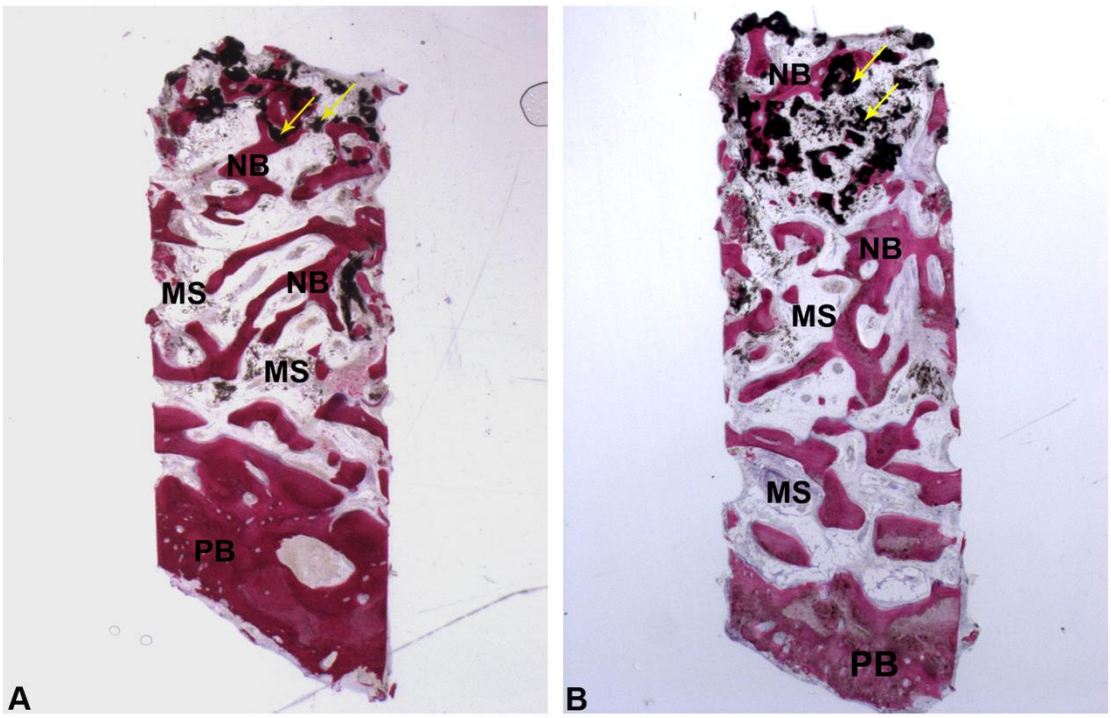



Figure 7: A, B) Test 1 and Test 2. Light microscopic ground sections showed the pre-existing bone (PB) at the bottom of the samples (occlusal region) while at the top, new bone trabeculae (NB) with large marrow spaces (MS) and a small amount of residual biomaterial were observed (yellow arrows). (Acid fuchsin-Toluidine blue 9X).

At higher magnification the osteocytes were entrapped by new bone, adjacent to the graft particles, new bone formation and osteoblastic activity were observed. Many blood vessels were adjacent to the bone formation areas (figure 8).

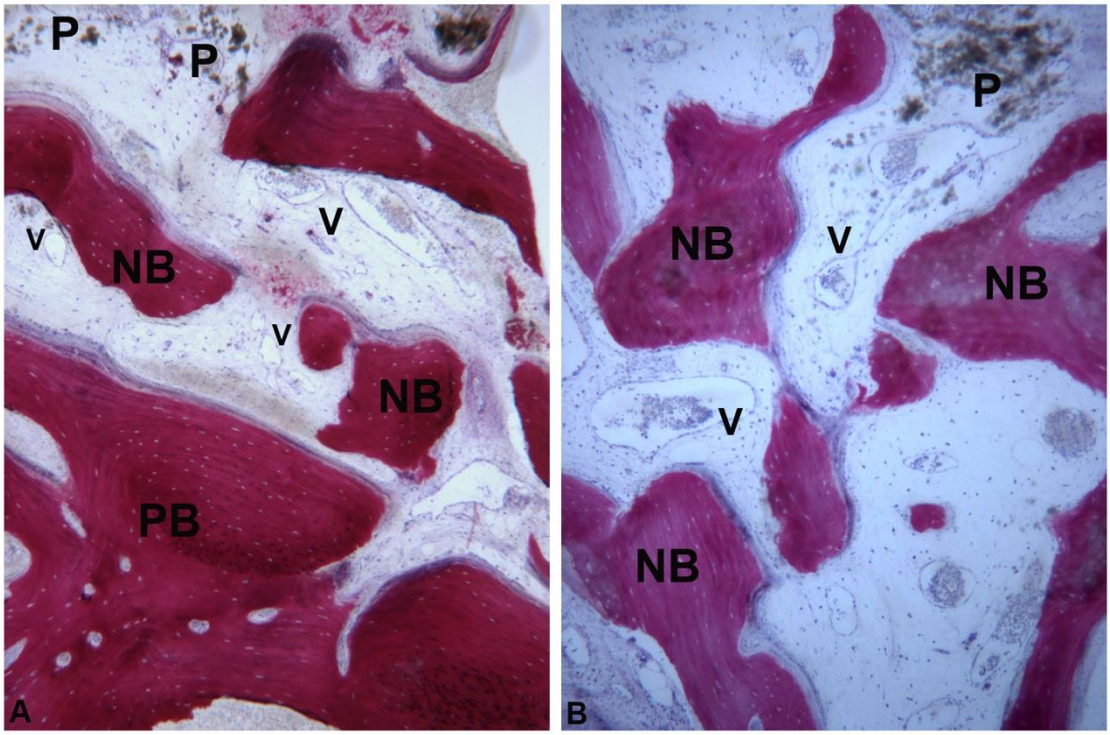



Figure 8: A)Test 1. Near to pre-existing bone (PB), newly formed bone (NB) was present, and a low amount of residual biomaterial (P) was detected. In this area, many blood vessels (V) were present.
B) The newly formed trabecular bone (NB) was surrounded by many blood vessels (V) and stromal cells. A small amount of residual biomaterials (P) was observed (Acid fuchsin-Toluidine blue 40X)

The percentages of new bone, residual biomaterials and non-mineralized tissues were 25.98%, 32.19% and 41.99% respectively (Table 2).

Table 2 – Histological results

| | Test 1 (BCP) | Test 2 (BCP + Hyaluronic acid) | Control (ABBM) | Group comparison |
|---|---|---|---|---|
| New Bone (%) ± SD | 23.85 ± 3.36 | 23.29 ± 2.01 | 25.97 ± 2.79 | P = 0.191 |
| Biomaterial (%) ± SD | 7.17 ± 4.37 | 7.47 ± 3.59 | 32.19 ± 1.52 | P < 0.000 |
| Non mineralized tissue (%) ± SD | 68.98 ± 7.40 | 69.80 ± 2.51 | 41.99 ± 3.44 | P < 0.000 |

### 3.1.1.2. Test 1(TPC) and 2 (TPC + Hyaluronic Acid)

Histological results were very similar for Test 1 and Test 2 groups. At low magnification, most of the samples showed the cortical bone at the bottom of the samples (crestal portion), while at the top (apical portion) of the sample the trabecular bone, was homogeneously distributed, with large marrow spaces and a small amount of RGP (figure 9). The biomaterial was almost completely resorbed in the areas adjacent to the pre-existing bone (figure 10), while the RGP were still present in the zones far from the cortical layer. In the areas far from the original bone the biomaterial particles showed signs of resorption, moreover, the RGP in some other areas were close to multinucleated cells (osteoclasts). Many blood vessels were present around RGP and osteoblasts.

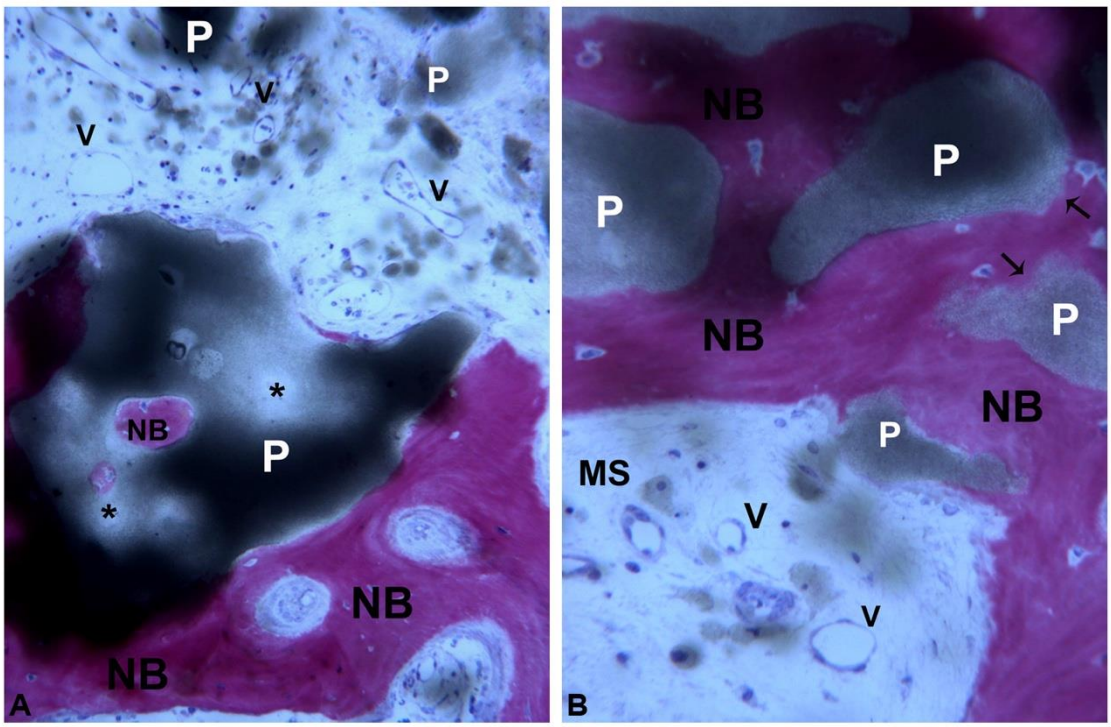



Figure 9: A)Test 1. Biomaterial particle (P) surrounded by newly formed bone (NB) was shown. Inside the particle, signs of bone degradation (*) and new bone formation (NB)

was observed. In the bone marrow many blood vessels (V) close to the residual biomaterial (P) were present.
B) Test 2. Some biomaterial particles (P) were incorporated in the newly formed bone (NB) and the shape of the particles revealed signs of resorption (black arrows). In the bone marrow (MS), the particles undergoing degradation (P) and some blood vessels (V) around residual biomaterial (P) were observed. (Acid fuchsin-Toluidine blue 100X)

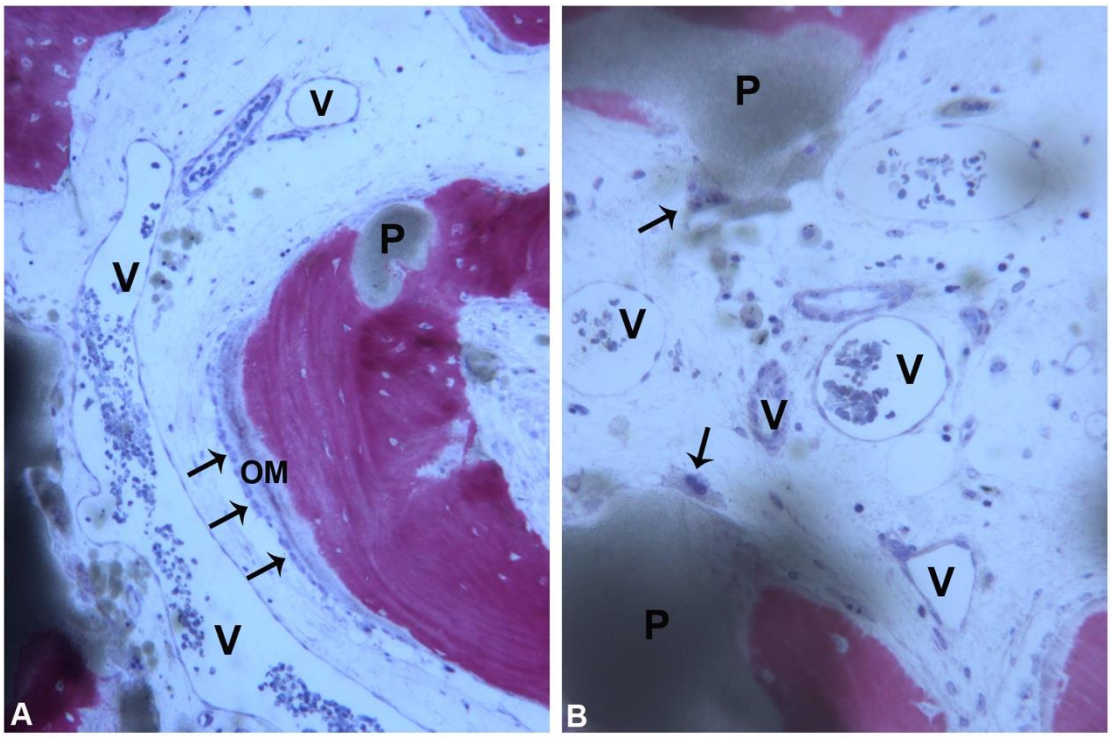



Figure 10: A) Test 1. Many small and large blood vessels (V) around residual biomaterial (P) and close to the osteoblasts (black arrows), deposting osteoid matrix (OM), were observed.
B) Test 2. Osteoclasts or multinucleated cells (black arrows) were also seen covering the biomaterial particles (P). Many blood vessels (V) between two residual biomaterial particles were present. (Acid fuchsin-Toluidine blue 100X and 200X)

In the Test group 1, the percentage of new bone, residual biomaterials and non-mineralized tissue was 23.85% 7.17%, and 68.98%, while in the Test group 2 it was 23.29%, 7.47% and 69.80% respectively (Table 2)

Considering the different histological components observed in biopsies, based on the different grafting materials, there was a statistically significant difference, F (6, 32) = 16.83, P < 0.0005; Wilk's Λ = 0.058, partial η2 = 0.759. When analyzing the between-subject effects, the graft material used had a statistically significant effect on both non-mineralized tissue (F [2, 18] = 72.003; P < .0005; partial η2 = 0.889), and residual graft particles (F [2, 18] = 126.268; P < .0005; partial η2 = 0.933), but not on new bone (F [2, 18] = 1.819; P = 0.191; partial η2 = 0.168), so the percentage of new bone was not statistically different between the three groups. The Tukey HSD post hoc test revealed that non-mineralized tissues were statistically significantly different between test 1 and control (P < 0.0005), test 2 and control (P < 0.0005), but not between test 1 and test 2 (P = 0.948). Similarly, RGP were statistically significantly different between test 1 and control (P < 0.0005), test 2 and control (P < .0005), but not between and test 1 and test 2 (P = 0.985).

## 3.2.Secondary Outcomes

### 3.2.1. Mean bone gain

Mean bone gain values, measured on CBCT, are shown in table 3 was 11.05 ± 1.5 mm for the control group, 12.25 ± 2.3 mm for the test 1 group, 11.04 ± 2.3 mm for the test 2 group, the difference observed between the groups was not statistically significant (F [2.20] = 0.881, P = 0.43) (Table 3).

Table 3 – Mean bone gain per group

| | Test 1 (BCP) | Test 2 (BCP + Hyaluronic acid) | Control (ABBM) | Group comparison |
|---|---|---|---|---|
| Mean bone gain (mm) ± SD | 12.25 ± 2.33 | 11.04 ± 2.29 | 11.05 ± 1.53 | P = 0.403 |

### 3.2.2. Implant insertion torque

A total of 44 implants, 15 in the control group, 16 in the test 1 group and 13 in the test 2 group, were placed. Precisely, 10 Bone Level and 34 Tissue Level implants were used, 16 Straumann(Bone and Tissue Level Implants, Straumann, Basel Switzerland)) and 28 Sweden & Martina (Premium/Prama, Sweden & Martina, Due Carrare, Italy) respectively. The mean insertion torque value was 41.9 ± 6.5 Ncm for the control group, 22.5 ± 5.3 N/cm for the test group 1 and 25.7 ± 6 N/cm for the test group 2. The difference between the groups was statistically significant (F [2.20] = 23.745, P <0.0005). The post hoc Tukey test revealed that the difference was statistically significant between the control group and the two test groups (P <0.0005), but not between the two test groups (P = 0.563).

### 3.2.3. Complications and implant failures

The number of complications detected, in the various phases of the study, was on average 1.33 ± 0.95 in total, and for the control, test 1 and test 2 groups respectively 0.88 ± 0.85, 1.38 ± 1.06 and 1.13 ± 0.99. No statistically significant difference was found between the groups in terms of

complications (F [2.21] = 0.535, P = 0.593). No implant failures were recorded at any stage.

### 3.2.4. Patient's reported outcome measures

Mean scores for the OHIP-14 questionnaire are reported in table 4, overall scores for control, test 1 and test 2 groups were respectively 8.63 ± 6.41, 12.13 ± 5.7 and 12.88 ± 6.23. The difference between the total OHIP-14 scores of the 3 groups was not statistically significant (F [2.21] = 1.098, P = 0.352). The only statistically significant differences between the three test groups were found for question 2 (P = 0.001), 3 (P = 0.038), and 12 (P = 0.002). The Tukey post hoc test shows the differences between the groups. For question 2, the control group showed a statistically significant difference compared to both the test group 1 (P = 0.001) and 2 (P = 0.021), while the test groups were not significantly different from each other (P = 0.329). The only statistically significant difference for question 3 was between the control group and the test 2 group (P = 0.044), albeit only slight. For question 12 the test 2 group showed a statistically significant difference from both the control group (P = 0.004) and the test group 1 (P = 0.004). Test group 2 and control group did not show statistically significantly differences from each other (P = 1).

Table 4 – OHIP-14 scores per group

| Impact item of the question | Test 1 (BCP) | Test 2 (BCP + Hyaluronic acid) | Control (ABBM) | Group comparison |
|---|---|---|---|---|
| Had trouble in pronouncing words | 0.38 ± 0.52 | 0.50 ± 0.53 | 0.50 ± 0.93 | P = 0.916 |
| Felt that sense of taste had worsened | 2.38 ± 0.52 | 1.75 ± 0.89 | 0.50 ± 1.07 | P = 0.001 |
| Had painful aching in your mouth | 1.25 ± 1.03 | 2.63 ± 1.30 | 1.00 ± 1.41 | P = 0.038 |
| Found it uncomfortable when eating food | 1.38 ± 1.06 | 0.88 ± 1.13 | 0.50 ± 0.76 | P = 0.233 |
| Been feeling self-conscious | 1.88 ± 1.64 | 1.38 ± 1.06 | 2.63 ± 1.68 | P = 0.263 |
| Have you felt tense | 0 ± 0 | 0 ± 0 | 0 ± 0 | |
| Diet has been unsatisfactory | 0 ± 0 | 0 ± 0 | 0 ± 0 | |
| Had to interrupt meals | 0.63 ± 0.92 | 0.50 ± 0.93 | 0.25 ± 0.46 | P = 0.639 |
| Found it difficult to relax | 0.50 ± 0.93 | 0.25 ± 0.70 | 0.38 ± 1.06 | P = 0.861 |
| Been a bit embarrassed | 0 ± 0 | 0 ± 0 | 0 ± 0 | |
| Been irritable with other people | 0 ± 0 | 0 ± 0 | 0 ± 0 | |
| Had difficulty during usual jobs | 0 ± 0 | 0.88 ± 0.83 | 0 ± 0 | P = 0.002 |
| Felt that life less satisfying | 0.63 ± 0.91 | 0.63 ± 0.74 | 0.75 ± 1.38 | P = 0.963 |
| Been totally unable to function | 0.25 ± 0.46 | 0.50 ± 0.93 | 0.88 ± 1.25 | P = 0.420 |
| Total | 12.12 ± 5.67 | 12.87 ± 6.27 | 8.62 ± 6.41 | P = 0.352 |

# 4. Discussion

While the percentage of new augmented bone in the three groups at histological exam wasn't statistically significant, the difference in the percentage of non-mineralized tissue and residual graft material was statistically significant.

It is not easy to analyze the results of a study on MSA, especially the histological ones, in fact it is almost impossible to find studies that make a comparative analysis between the exact same materials, different biomaterials are often used in combination with others. For example, ABBM is often used with autologous bone (AB) or other biomaterials, calcium phosphate, in addition to being often used in combination with other materials, is sometimes used in its form of beta tricalcium phosphate alone or, as in our study, mixed with HA.

A systematic review with meta-analysis of Danesh-Sani SA et al.[17] shows how, in studies using xenografts as grafting material, the histomorphometric results relating to new bone, residual biomaterial and non-mineralized tissue (26.82 ± 26.54, 29.33 ± 46.74, 44.86 ± 21.26), are very similar to the results that we obtained with ABBM, while the same cannot be said for the reported mean values relating to alloplastic materials. However, in the latter category the authors insert a very diverse variety of materials (TCP, BCP, bio-glass, corals, calcium carbonate, calcium sulphate, etc.), therefore, a real comparison with our results obtained with BCP is not possible.

A series of RCTs conducted by the same group of authors reported histological values obtained from biopsies collected with the same timing of our study, 9 months. The results obtained differ from ours but also in this

case the comparability is limited by the fact that xenograft was used, while in our study we also used TCP and HA.[38-40]

In the above-mentioned systematic review a comparative analysis was performed, based on histomorphometric outcomes between several graft biomaterials and consistent with our findings no statistically significant difference for newly formed bone was found when comparing xenografts with alloplastic biomaterials.[17] On the other hand, the authors of the systematic review did not find significant differences for residual graft materials and non-mineralized tissues; this was in contrast with our study, where the residual graft particles were significantly higher in the control group than in the 2 test groups, and the non-mineralized tissues were significantly lower in the control group when compared to the test groups. As already mentioned, however, a varied amount of different types of materials falls under the categories of xenografts and alloplastic materials.

Calcium phosphate has a greater tendency to reabsorb and leave less biomaterial residues as shown in our study and confirmed by a study from Boëck-Neto[41] in which the percentage of residual biomaterial, in the maxillary sinus, at histological control was 6.19%, although in this cited study new bone percentages were found higher than in ours, probably because Calcium phosphate was mixed with autologous bone in a 2:1 ratio.

A more recent study by Wagner et al.[42], similarly to our study, compares the use of BCP mixed with fibrin sealant with the use of autologous bone mixed with ABBM. In this latter study, the histological results relating to the ABBM + AB graft are very similar to ours obtained with ABBM only, conversely the results obtained with BCP + fibrin sealant show percentages

of new bone and non-mineralized tissue (20.6% and 54 %) similar to our study albeit slightly lower, and a percentage of residual biomaterial much higher than our data (25.4%).

In general, CaP-based grafting materials have a rather unpredictable rate of resorption, thus are less able to maintain the grafted volumes, and have a greater structural fragility, probably, as our results show, due to the greater presence of residual non-mineralized tissues[43]. The advantage of using these biomaterials is their ability to stimulate osteoblastic proliferation and differentiation thanks to their composition, structure and crystallinity which is similar to that of calcium hydroxyapatite, the main inorganic component of the bone.[44-46]

The other variable included in the second test group of this study was the addition of HA to BCP. The use of HA for bone regeneration still has a limited amount of evidence in the literature, however some results are promising. In vitro, Asparuhova et al.[21] showed that HA, when added to preosteoblastic mesenchymal stromal cells, strongly induces the growth of osteoprogenitors and the expression of genes encoding for bone matrix proteins. Mendes et al.[47], in an animal study on rats showed that HA alone in post-extraction sockets showed accelerated bone healing compared to the control at 21 days. Kim et al.[48] showed how HA in infected post-extraction sockets caused greater bone formation thanks to its osteoinductive but also bacteriostatic and anti-inflammatory properties.

Three interesting clinical studies show the results of the use of HA in bone regeneration. Alcantara et al.[49], in a cone beam analysis, describe how HA, in post-extractive sockets, shows more bone volume at 30 days

compared to the control, but an insignificant difference at 90 days. Lorenz et al.[50] showed good bone formation induction ability of BCP+HA in post-extraction sockets at 4 months, however the study did not have a control group. Finally, in a study on lateral sinus augmentation, Knabe et al.[51], described good results, in terms of bone volumes obtained in regenerated sinuses, both for TCP alone and for TCP + HA at 6 months, but greater bone formation with TCP alone. However, in the two groups the TCP was not in the same form, being in granules in the TCP+HA group, as a putty scaffold in the TCP alone group, thus representing a major confounding factor.

As mentioned, the results are promising, but larger and better designed clinical studies are needed to draw definitive conclusions. From in vitro and animal studies we could say that HA probably plays an important role in accelerating bone formation in the initial phases, so in our study the 9-month follow-up is not able to highlight its beneficial effects. If this role were confirmed through clinical studies, shorter waiting times after bone grafting could be hypothesized for implant placement.

Comparing the bone height gain obtained after MSA with other studies present in the literature has a very relative value, in fact it is not only the material used that determines this outcome, but also and above all the technique used and other anatomical factors such as the width of the sinus. This was illustrated in a study by Stacchi et al.[52] which shows how the width of the maxillary sinus affects not only the final bone height gain, but also the quality, measured histomorphometrically, of the regenerated bone. Certainly the xenogenic grafts, having a greater persistence of residual material, guarantee a greater medium and long-term persistence of the volumes with greater increase in height, as demonstrated also by recent studies[53, 54], and

also have improved mechanical strength properties as demonstrated in this study by the insertion torque values[43]. Surely this data on the highest insertion torque in the ABBM group reconnects to the histological data that shows values on the total mineralized tissue (new bone and graft residues) that are significantly higher in the control group.

Overall, the incidence of complications was very low in all groups, with no complications in the implant and restorative phase and the few complications related to episodes of hematoma or swelling in the post-operative period after MSA and 6 perforations of the sinus membrane during MSA, all successfully repaired. This data is supported by similar evidence in the literature, in particular a systematic review with meta-analysis of Raghoebar et al.[55] which shows data similar to ours as seen in the studies included in the analysis.

The last of the parameters that this study proposed to evaluate was that related to the influence of oral health, in this case after an MSA procedure such, on the quality of life. We decided to do it with a questionnaire, adopted by many other studies, handed to the patient at a specific time point, but with questions that related to the entire previous period[35, 56-58]. The total scores of the OHIP-14 fpr the three groups were very low, showing that, despite its invasiveness, an MSA procedure does not have a negative impact on the quality of life of our patients. There was a statistically significant difference between the three groups only for three questions, however the scores were so low that reading this data in favor or against one of the three groups would represent, in our opinion, an interpretative distortion. In general, no question, for any group, scored higher than 2, which corresponds to "occasionally" and four questions, the numbers 6, 7, 10 and 11, received an average score of 0.

This confirms, therefore, the MSA as a comfortable procedure for the patient regardless of the material used for the graft.

# 5. Conclusions

1. MSA is a safe and predictable procedure from the biological and clinical point of view and with a high comfort perceived by the patient.

2. The use of ABBM or BCP has not influenced the outcomes in terms of bone gain in this study, with values that were comparable and not statistically significant when measured radiographically.

3. Histomorphometric results suggest better bone quality for ABBM with an higher mineral component visible in the histologic sections of the control group when compared with test groups 1 and 2 with a more accentuated non mineralized tissues component.

4. Implant insertion torque values suggest better mechanical resistance properties with the use of ABBM.

5. The addition of hyaluronic acid did not influence the outcomes in terms of bone gain or histologic results. Further analysis should be addressed at assessing the role of HA in the initial phases of bone formation

# 6. Bibliography

1. Aghaloo T, Moy P. Which Hard Tissue Augmentation Techniques Are the Most Successful in Furnishing Bony Support for Implant Placement? *Int J Oral Maxillofac Implants*. 2007;22(Suppl)

2. Elnayef B, Porta C, Suarez-Lopez Del Amo F, Mordini L, Gargallo-Albiol J, Hernandez-Alfaro F. The Fate of Lateral Ridge Augmentation: A Systematic Review and Meta-Analysis. *Int J Oral Maxillofac Implants*. May/Jun 2018;33(3):622-635. doi:10.11607/jomi.6290

3. Corbella S, Taschieri S, Del Fabbro M. Long-term outcomes for the treatment of atrophic posterior maxilla: a systematic review of literature. *Clin Implant Dent Relat Res*. Feb 2015;17(1):120-32. doi:10.1111/cid.12077

4. 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*. Jun 2013;24(6):635-44. doi:10.1111/j.1600-0501.2012.02438.x

5. Nkenke E, Stelzle F. Clinical outcomes of sinus floor augmentation for implant placement using autogenous bone or bone substitutes: a systematic review. *Clin Oral Implants Res*. Sep 2009;20 Suppl 4:124-33. doi:10.1111/j.1600-0501.2009.01776.x

6. 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*. Sep 2008;35(8 Suppl):216-40. doi:10.1111/j.1600-051X.2008.01272.x

7. Boyne PJ, James RA. Grafting of the maxillary sinus floor with autogenous marrow and bone. *J Oral Surg*. Aug 1980;38(8):613-6. doi:http://ci.nii.ac.jp/naid/10019575083/

8. Tatum H, Jr. Maxillary and sinus implant reconstructions. *Dent Clin North Am*. Apr 1986;30(2):207-29.

9. Misch CE. Maxillary sinus augmentation for endosteal implants: organized alternative treatment plans. *Int J Oral Implantol*. 1987;4(2):49-58.
10. Valente NA. Anatomical Considerations on the Alveolar Antral Artery as Related to the Sinus Augmentation Surgical Procedure. *Clin Implant Dent Relat Res*. Oct 2016;18(5):1042-1050. doi:10.1111/cid.12355
11. van den Bergh JP, ten Bruggenkate CM, Disch FJ, Tuinzing DB. Anatomical aspects of sinus floor elevations. *Clin Oral Implants Res*. Jun 2000;11(3):256-65. doi:10.1034/j.1600-0501.2000.011003256.x
12. Testori T, Weinstein T, Taschieri S, Wallace SS. Risk factors in lateral window sinus elevation surgery. *Periodontol 2000*. Oct 2019;81(1):91-123. doi:10.1111/prd.12286
13. Flanagan D. Arterial supply of maxillary sinus and potential for bleeding complication during lateral approach sinus elevation. *Implant Dent*. Dec 2005;14(4):336-8. doi:10.1097/01.id.0000188437.66363.7c
14. Solar P, Geyerhofer U, Traxler H, Windisch A, Ulm C, Watzek G. Blood supply to the maxillary sinus relevant to sinus floor elevation procedures. *Clin Oral Implants Res*. Feb 1999;10(1):34-44. doi:10.1034/j.1600-0501.1999.100105.x
15. Traxler H, Windisch A, Geyerhofer U, Surd R, Solar P, Firbas W. Arterial blood supply of the maxillary sinus. *Clin Anat*. 1999;12(6):417-21. doi:10.1002/(SICI)1098-2353(1999)12:6<417::AID-CA3>3.0.CO;2-W
16. Strong C. The Innervation and Vascular Supply of the Antrum: (Section of Laryngology). *Proc R Soc Med*. Apr 1934;27(6):745-51.
17. 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 Periodontal Res*. Jun 2017;52(3):301-312. doi:10.1111/jre.12402

18. Miron RJ, Zhang Q, Sculean A, et al. Osteoinductive potential of 4 commonly employed bone grafts. *Clin Oral Investig*. Nov 2016;20(8):2259-2265. doi:10.1007/s00784-016-1724-4

19. Schmidt JR, Vogel S, Moeller S, et al. Sulfated hyaluronic acid and dexamethasone possess a synergistic potential in the differentiation of osteoblasts from human bone marrow stromal cells. *J Cell Biochem*. Nov 28 2018;doi:10.1002/jcb.28158

20. Rony L, Aguado E, Pascaretti-Grizon F, Hubert L, Chappard D. Hyaluronic Acid Stimulates Osseointegration of beta-TCP in Young and Old Ewes. *Calcif Tissue Int*. Nov 2019;105(5):487-496. doi:10.1007/s00223-019-00589-y

21. Asparuhova MB, Chappuis V, Stahli A, Buser D, Sculean A. Role of hyaluronan in regulating self-renewal and osteogenic differentiation of mesenchymal stromal cells and pre-osteoblasts. *Clin Oral Investig*. Nov 2020;24(11):3923-3937. doi:10.1007/s00784-020-03259-8

22. Sasaki T, Watanabe C. Stimulation of osteoinduction in bone wound healing by high-molecular hyaluronic acid. *Bone*. Jan 1995;16(1):9-15. doi:10.1016/s8756-3282(94)00001-8

23. de Ruiter A, Janssen N, van Es R, et al. Micro-structured Beta-Tricalcium Phosphate for Repair of the Alveolar Cleft in Cleft Lip and Palate Patients: A Pilot Study. *Cleft Palate Craniofac J*. May 2015;52(3):336-40. doi:10.1597/13-260

24. Esposito M, Grusovin MG, Kwan S, Worthington HV, Coulthard P. Interventions for replacing missing teeth: bone augmentation techniques for dental implant treatment. *Cochrane Database Syst Rev*. Jul 16 2008;16(3):CD003607. doi:10.1002/14651858.CD003607.pub3

25. Schwartz Z, Goldstein M, Raviv E, Hirsch A, Ranly DM, Boyan BD. Clinical evaluation of demineralized bone allograft in a hyaluronic acid

carrier for sinus lift augmentation in humans: a computed tomography and histomorphometric study. *Clin Oral Implants Res*. Apr 2007;18(2):204-11. doi:10.1111/j.1600-0501.2006.01303.x

26. 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*. Mar 2012;23(3):263-73. doi:10.1111/j.1600-0501.2011.02168.x

27. King SR, Hickerson WL, Proctor KG. Beneficial actions of exogenous hyaluronic acid on wound healing. *Surgery*. Jan 1991;109(1):76-84.

28. Ortonne JP. [Comparative study of the activity of hyaluronic acid and dextranomer in the treatment of leg ulcers of venous origin]. *Ann Dermatol Venereol*. Mar 2001;Suppl:13-6. Etude comparative de l'activite de l'acide hyaluronique et du dextranomere dans le traitement des ulceres de jambe d'origine veineuse.

29. Pirnazar P, Wolinsky L, Nachnani S, Haake S, Pilloni A, Bernard GW. Bacteriostatic effects of hyaluronic acid. *J Periodontol*. Apr 1999;70(4):370-4. doi:10.1902/jop.1999.70.4.370

30. Carlson GA, Dragoo JL, Samimi B, et al. Bacteriostatic properties of biomatrices against common orthopaedic pathogens. *Biochem Biophys Res Commun*. Aug 20 2004;321(2):472-8. doi:10.1016/j.bbrc.2004.06.165

31. Imai H, Lang NP, Ferri M, Hirota A, Apaza Alccayhuaman KA, Botticelli D. Tomographic Assessment on the Influence of the Use of a Collagen Membrane on Dimensional Variations to Protect the Antrostomy After Maxillary Sinus Floor Augmentation: A Randomized Clinical Trial. *Int J Oral Maxillofac Implants*. Mar/Apr 2020;35(2):350-356. doi:10.11607/jomi.7843

32. Hirota A, Lang NP, Ferri M, Fortich Mesa N, Apaza Alccayhuaman KA, Botticelli D. Tomographic evaluation of the influence of the placement

of a collagen membrane subjacent to the sinus mucosa during maxillary sinus floor augmentation: a randomized clinical trial. *Int J Implant Dent*. Aug 19 2019;5(1):31. doi:10.1186/s40729-019-0183-5

33. Kawakami S, Lang NP, Ferri M, Apaza Alccayhuaman KA, Botticelli D. Influence of the height of the antrostomy in sinus floor elevation assessed by cone beam computed tomography- a randomized clinical trial. *Int J Oral Maxillofac Implants*. January/February 2019;34(1):223-232. doi:10.11607/jomi.7112

34. Kawakami S, Lang NP, Iida T, Ferri M, Apaza Alccayhuaman KA, Botticelli D. Influence of the position of the antrostomy in sinus floor elevation assessed with cone-beam computed tomography: A randomized clinical trial. *J Investig Clin Dent*. Nov 2018;9(4):e12362. doi:10.1111/jicd.12362

35. Slade GD. Derivation and validation of a short-form oral health impact profile. *Community Dent Oral Epidemiol*. Aug 1997;25(4):284-90. doi:10.1111/j.1600-0528.1997.tb00941.x

36. Buser D, Weber HP, Lang NP. Tissue integration of non-submerged implants. 1-year results of a prospective study with 100 ITI hollow-cylinder and hollow-screw implants. *Clin Oral Implants Res*. Dec 1990;1(1):33-40. doi:10.1034/j.1600-0501.1990.010105.x

37. 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*. Sep 2016;27(9):1106-22. doi:10.1111/clr.12702

38. Hirota A, Iezzi G, Piattelli A, et al. Influence of the position of the antrostomy in sinus floor elevation on the healing of mini-implants: a randomized clinical trial. *Oral Maxillofac Surg*. Sep 2020;24(3):299-308. doi:10.1007/s10006-020-00846-7

39. Imai H, Iezzi G, Piattelli A, Ferri M, Alccayhuaman KAA, Botticelli D. Influence of the Dimensions of the Antrostomy on Osseointegration of Mini-implants Placed in the Grafted Region After Sinus Floor Elevation: A Randomized Clinical Trial. *Int J Oral Maxillofac Implants*. May/Jun 2020;35(3):591-598. doi:10.11607/jomi.8005

40. Tanaka K, Iezzi G, Piattelli A, et al. Sinus Floor Elevation and Antrostomy Healing: A Histomorphometric Clinical Study in Humans. *Implant Dent*. Dec 2019; 28(6):537-542. doi: 10.1097/ ID.0000000000000932

41. Boeck-Neto RJ, Gabrielli MF, Shibli JA, Marcantonio E, Lia RC, Marcantonio E, Jr. Histomorphometric evaluation of human sinus floor augmentation healing responses to placement of calcium phosphate or Ricinus communis polymer associated with autogenous bone. *Clin Implant Dent Relat Res*. 2005;7(4):181-8. doi:10.1111/j.1708-8208.2005.tb00063.x

42. Wagner W, Wiltfang J, Pistner H, et al. Bone formation with a biphasic calcium phosphate combined with fibrin sealant in maxillary sinus floor elevation for delayed dental implant. *Clin Oral Implants Res*. Sep 2012;23(9):1112-7. doi:10.1111/j.1600-0501.2011.02275.x

43. Haugen HJ, Lyngstadaas SP, Rossi F, Perale G. Bone grafts: which is the ideal biomaterial? *J Clin Periodontol*. Jun 2019;46 Suppl 21:92-102. doi:10.1111/jcpe.13058

44. Park J. *Bioceramics: Properties, characterizations, and applications*. Springer Science & Business Media; 2008.

45. Huang Q, Wang L, Wang J. Mechanical properties of artificial materials for bone repair. *Journal of Shanghai Jiaotong University (Science)*. 2014;19(6):675–80.

46. Laurencin C, Khan Y, Veronick J. Bone graft substitutes: Past, present, and future. *Bone graft substitutes and bone regenerative engineering*. 2nd ed. ASTM International; 2014:1-9.

47. Mendes RM, Silva GA, Lima MF, et al. Sodium hyaluronate accelerates the healing process in tooth sockets of rats. *Arch Oral Biol*. Dec 2008;53(12):1155-62. doi:10.1016/j.archoralbio.2008.07.001

48. Kim JJ, Song HY, Ben Amara H, Kyung-Rim K, Koo KT. Hyaluronic Acid Improves Bone Formation in Extraction Sockets With Chronic Pathology: A Pilot Study in Dogs. *J Periodontol*. Jul 2016;87(7):790-5. doi:10.1902/jop.2016.150707

49. Alcantara CEP, Castro MAA, Noronha MS, et al. Hyaluronic acid accelerates bone repair in human dental sockets: a randomized triple-blind clinical trial. *Braz Oral Res*. 2018;32:e84. doi:10.1590/1807-3107bor-2018.vol32.0084

50. Lorenz J, Barbeck M, Kirkpatrick CJ, Sader R, Lerner H, Ghanaati S. Injectable Bone Substitute Material on the Basis of beta-TCP and Hyaluronan Achieves Complete Bone Regeneration While Undergoing Nearly Complete Degradation. *Int J Oral Maxillofac Implants*. May/June 2018;33(3):636-644. doi:10.11607/jomi.6026

51. Knabe C, Adel-Khattab D, Kluk E, Struck R, Stiller M. Effect of a Particulate and a Putty-Like Tricalcium Phosphate-Based Bone-grafting Material on Bone Formation, Volume Stability and Osteogenic Marker Expression after Bilateral Sinus Floor Augmentation in Humans. *J Funct Biomater*. Jul 29 2017;8(3)doi:10.3390/jfb8030031

52. Stacchi C, Lombardi T, Ottonelli R, Berton F, Perinetti G, Traini T. New bone formation after transcrestal sinus floor elevation was influenced by sinus cavity dimensions: A prospective histologic and histomorphometric

study. *Clin Oral Implants Res*. May 2018;29(5):465-479. doi:10.1111/clr.13144

53. Fouad W, Osman A, Atef M, Hakam M. Guided maxillary sinus floor elevation using deproteinized bovine bone versus graftless Schneiderian membrane elevation with simultaneous implant placement: Randomized clinical trial. *Clin Implant Dent Relat Res*. Jun 2018;20(3):424-433. doi:10.1111/cid.12601

54. Santagata M, Tozzi U, Tartaro G, et al. Maxillary Sinus Augmentation with Autologous and Heterologous Bone Graft: A Clinical and Radiographic Report of Immediate and Delayed Implant Placement. *J Maxillofac Oral Surg*. Dec 2014;13(4):401-8. doi:10.1007/s12663-013-0569-5

55. Raghoebar GM, Onclin P, Boven GC, Vissink A, Meijer HJA. Long-term effectiveness of maxillary sinus floor augmentation: A systematic review and meta-analysis. *J Clin Periodontol*. Jun 2019;46 Suppl 21:307-318. doi:10.1111/jcpe.13055

56. Younes F, Cosyn J, De Bruyckere T, Cleymaet R, Eghbali A. A 2-year prospective case series on volumetric changes, PROMs, and clinical outcomes following sinus floor elevation using deproteinized bovine bone mineral as filling material. *Clin Implant Dent Relat Res*. Apr 2019;21(2):301-309. doi:10.1111/cid.12730

57. Farzadmoghadam M, Mohammadi TM, Goudarzi R, Mohammadi M, Hasheminejad N. Is there a relationship between general and oral health-related quality of life in partially edentulous patients before and after implant treatment? A quasi-experimental study. *Clin Oral Implants Res*. Jun 2020;31(6):557-564. doi:10.1111/clr.13593

58. Zucoloto ML, Maroco J, Campos JA. Impact of oral health on health-related quality of life: a cross-sectional study. *BMC Oral Health*. May 12 2016;16(1):55. doi:10.1186/s12903-016-0211-2