



Observational Study

HISTOMORPHOMETRIC ANALYSIS ON SOCKET PRESERVATION IN THE UPPER JAW USING A NEW XENOGRAFT MATERIAL

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ABSTRACT

After a dental extraction, a variable amount of bone resorption of the residual ridge is observed quantitatively and qualitatively. Alveolar Socket Preservation is a surgical technique that fills alveolar space with biomaterial to maintain alveolar ridge volume for subsequent implant insertion. The purpose of this study is to histologically analyze the healing process of the post-extraction alveoli in the upper jaws grafted with a new biomaterial. Five patients were enrolled in the study, all female, non-smokers, with no periodontal disease or diabetes, and not on any medication. The five treated sites were from three females mean age of 49 years. The two control sites were from two females mean age of 71 years. Test alveoli were packed with decellularized, and antigen-free bovine bone processed at low temperature (RE-BONE®; Ubgen, Padua, Italy) and then covered with a bovine-derived pericardium membrane (SHELTER® FAST; Ubgen, Padua, Italy). At 4 months, surgery for implant insertion was scheduled, and sampling was carried out to obtain bone to be histologically analyzed. The histomorphometric analysis showed an average increase of 6.3% of bone tissue in treated samples compared to controls, but no statistically significant differences were obtained due to the high standard deviation values. In our case series, the new biomaterial shows a good trend as regards the alveolar healing process. However, no conclusion can be drawn due to the limited sample. Therefore, additional studies with greater sample sizes are needed to obtain conclusive results.

KEYWORDS: *bone, graft, alveolus, maxilla, upper jaw*

INTRODUCTION

Alveolar bone is a “tooth-dependent” structure that develops during an eruption, its anatomy (height and thickness) is determined by the formation/presence of the teeth and their axis of eruption.

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Scientific evidence has shown the dynamic change of the tissues after tooth extraction (1, 2). In the first phase after tooth extraction, there is remodelling and resorption of the lingual and buccal walls due to the periodontal ligament's lack of nutritional support. Alveolar bone decreases by about 15% 6 months after extraction (3). Consequently, the size of the socket is reduced both vertically and horizontally. This dimensional change can lead to aesthetic and functional disadvantages, which reflect in a proper subsequent implant insertion. An adequate residual ridge width is one of the main prerogatives for success in long-term implant therapy. In addition, adequate bone volume is required for good soft tissue support (4).

The percentage of alveolar resorption after a tooth extraction is more influenced by bone thicknesses that are greater on the buccal side than on the lingual wall, and consequently, the greatest resorption is found vestibularly. This remodelling occurs in both the lower and upper arches. The most significant contraction occurs during the first month and stabilizes in six months (5).

Schropp et al. (6) studied cases and estimated a loss of ridge width of about 50% over a year. This study highlights the importance of maintaining bone volumes after tooth extraction, especially when dealing with aesthetic areas.

Among the different approaches proposed in the literature to preserve the edentulous ridge, animal-derived biomaterials associated with using barrier membranes were undoubtedly the most analyzed (7). The Alveolar Socket Preservation Technique (ASPT) is a regenerative technique used to minimize the dimensional changes of hard and soft tissues after a dental extraction (8).

ASPT inserts biomaterials in post-extractive alveolus in order to maintain the crest volume. In ASPT, the healing process is similar to that of untreated alveoli (9, 10). The use of membranes to cover the grafted alveolus aim to maintain the biomaterial in situ, or in an ungrafted site, to initially preserve the blood clot, thus excluding its colonization by epithelial cells. Recently a new xenograft has been introduced in the market. The previous report showed that it positively affects alveolus volume maintenance (11, 12).

Here a series of patients were enrolled to be treated with this new biomaterial to maintain crest volume. After 4 months, sampling was performed, and specimens were histologically analyzed to get more information as regards the healing process.

MATERIALS AND METHODS

Five patients who underwent tooth extraction on the upper jaw were enrolled on the study. The reasons for the dental extraction were vertical root fractures, destructive caries, and endodontically non-retractable teeth. All patients were women without periodontal disease or diabetes, non-smokers, and not taking medications such as bisphosphonates or immunosuppressants. The five sites treated with the ASPT were from three patients with a mean age of 49. The two control sites were from two subjects with a mean age of 71.

Surgical procedure

During extraction, an attempt was made to lift the flaps in the least invasive way possible to preserve the alveolus bone from further resorption due to surgical exposure. The alveolus was packed with decellularized and antigen-free bovine bone processed at low temperature (RE-BONE®; Ubgen, Padua, Italy) and then covered with a bovine-derived pericardium membrane (SHELTER® FAST; Ubgen, Padua, Italy). The two control sites were treated with the same surgical procedure but without using biomaterials, and the alveoli were left to heal spontaneously (Fig. 1-4).

Compression sutures were performed in monofilament in e-PTFE (Gore-Tex®) and removed after 10 days. An ice pack was maintained for a few hours. Anti-inflammatory therapy with Nimesulide was prescribed as well as soft and cold diet for at least 2/3 days and rinses with Chlorhexidine 2/3 times a day for 15/20 days.

After fourth months, patients were scheduled for implant insertion. Before fixture insertion, sampling was performed using a core drill with a diameter of 2 mm for a depth of 2 mm. The bone samples were placed in sterile and



Fig. 1. *Pre-surgical radiograph*

labelled blisters, immersed in formalin and subsequently sent to the laboratory for histological examination.

Histological analysis

Specimens were treated with Osteosoft® to decalcify the bone samples fixed in formalin. Samples were then embedded in paraffin. A microtome (RM2025 Leica Instruments, Nussloch, Germany) was used to obtain a 5 µm thick section. These paraffin sections, collected on a microscope slide, were deparaffinated, rehydrated, and stained with Haematoxylin and Eosin.

After staining, the sections were dehydrated in alcohol, cleared in xylene, and then preserved using a suitable mounting medium for morphological observations. All reagents were obtained from Sigma-Aldrich.

Histological slides were scanned using an APERIO ScanScope slide scanner (Leica Biosystems, Buccinasco-Milano, Italy), obtaining an image file with .svs (ScanScope Virtual Slide) format for every sample. The .svs files were analyzed using a free software program called ImageScope. The total area of the histological section was measured, as well as the areas occupied by bone and connective tissues.

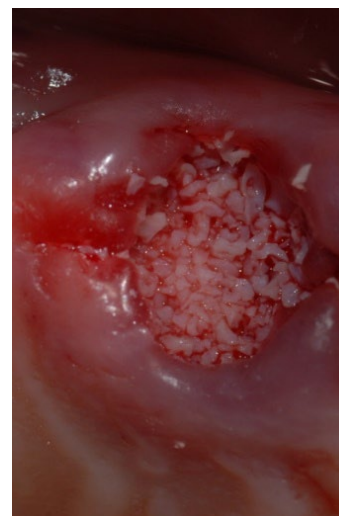


Fig. 2. *Socket preservation surgery*

RESULTS

Fig. 5 shows histological pictures of test and control specimens. On the left are the areas limited by red and green lines, which correspond to the total sample area and connective tissue area. The bone area is derived from the difference between total and connective tissue areas. The length of the sample, the total area, and the percentage of bone and connective tissues in the scanned histology slides were quantified by ImageScope software.

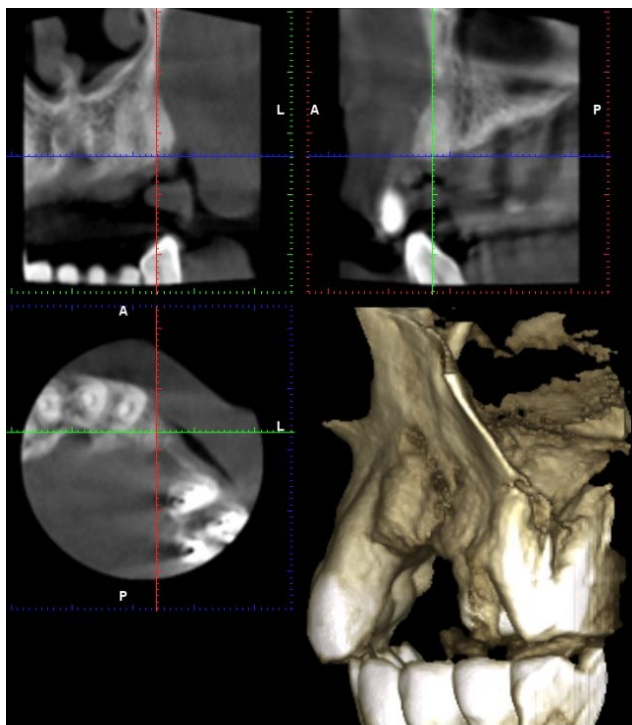


Fig. 3. *Cone beam performed after 4 months of socket preservation*

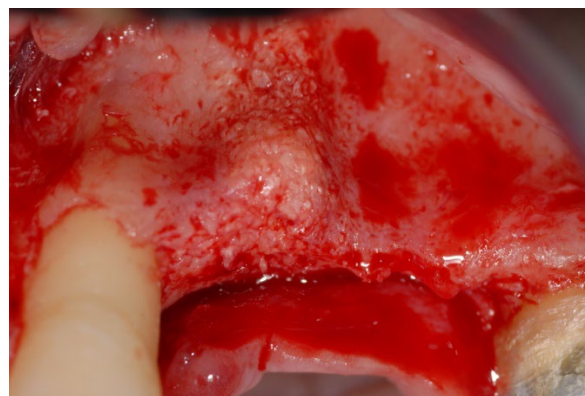


Fig. 4. *Surgical field just before implant insertion showing the healed alveolus*

Table I reports the average values obtained by all specimens. In treated alveoli, there is an average increase of 6.3% in bone area. Since the standard deviation is similar to the bone gain in the test sample, no statistically significant difference was obtained.

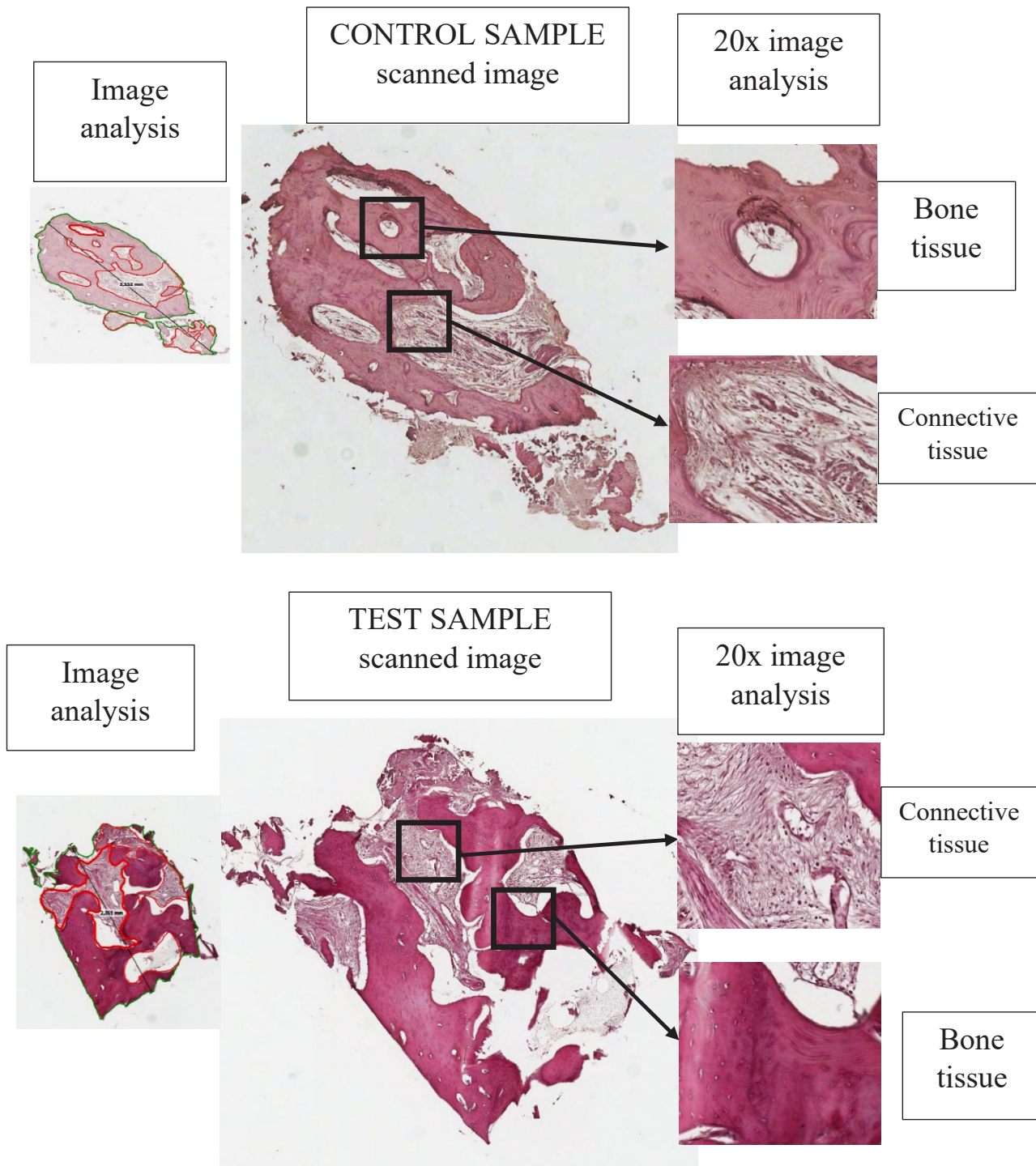


Fig. 5. Histological and histomorphometric image of a control sample and a test sample.

Table I. Average values obtained in bone histomorphometry.

	Average values				
	Age	Length of samples	Analyzed area	% bone	% connective
Control samples	71.0	3.4 (± 0.4)	4.2 (± 2.3)	59.9 (± 5)	40.1 (± 5)
Test samples	49.0	3.8 (± 1.94)	5.2 (± 0.8)	66.2 (± 4.96)	33.8 (± 4.96)

DISCUSSION

ASPT is any procedure that takes place immediately after dental extraction to preserve the volume of the alveoli (13). One of the major problems after the extraction of multi-rooted elements in the upper molar area is the loss and/or fracture of the buccal cortex, which makes subsequent management of the implant/prosthetic case much more difficult (14).

Sisti et al. (15) reported that ASPT minimized resorption of the alveolar ridge and provided better regeneration results in sites with buccal bone defects greater than 5 mm compared to the traditional regeneration procedure performed after healing of the socket.

The detachment and careful removal of the granular tissue, combined with a minimally invasive extraction to reduce trauma to the alveolar bone, is of paramount importance to obtain good results with ASPT (16).

The problem of the loss of the vestibular cortex can be attributed to various factors such as trauma, alveolar dehiscence, fracture during avulsion manoeuvres and endodontic infections; to minimize the number of variables, patients without periodontal disease were enrolled in the present study.

Until now, most human studies on ASPT focus on molars extracted for severe periodontitis (17, 18). For example, a study by Rasperini et al. (19), conducted in the posterior area, reported only data for preserved alveoli with four intact walls, while Zhao et al. (20) evaluated only sites in anterior areas.

Carmagnola and coll. (21) performed a study using Bio-Gide® and Bio-Oss® (Geistlich Pharma AG, Switzerland). They divided patients into 3 groups and did histological examinations at 4-7-12 months. At 4 months, connective tissue was present in the membrane graft group, and 40% of the newly formed bone was around the biomaterial.

Cardaropoli et al. (22), in a randomized study, showed that the ASPT group has a significant minor reduction in width and height of the buccal bone crest with respect to the control group with the following values: 1.04 ± 1.08 mm vs 4.48 ± 0.65 mm in width, and 0.46 ± 0.46 mm vs 1.54 ± 0.33 mm in height.

Lee et al. (23) confirmed the potential of ASPT in areas where the vestibular cortex had been compromised. Tomasi et al. (24) stated that if the buccal cortex maintained a thickness greater than 1 mm after extraction, ASPT is unnecessary, while it is indispensable when the thickness of bone is lower.

Since a new biomaterial has been recently introduced in the market and partially investigated (11, 12), we decided to perform a study to evaluate histological healing of post-extractive sites in the upper molar region. Our results show that in treated alveoli, there is an average increase of 6.3% in bone tissue. However, no statistically significant difference is obtained since the standard deviation is as great as to the bone gain in the test sample.

CONCLUSIONS

Alveolar volume preservation is of paramount importance to have a subsequent proper implant rehabilitation. ASPT is a well-known technique. In addition, various biomaterials are available on the market. Here we investigated a new xenograft inserted in the upper molar area.

From a histological point of view, the healing process has a favourable course with slight increases in bone deposition compared to untreated sites. However, no statistically significant difference was obtained due to the great standard deviation. Furthermore, it is due to the small sample size. Therefore, we believe that additional studies with greater sample sizes are needed to obtain definitive results.

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REFERENCES

1. Pietrokovski J, Massler M. Alveolar ridge resorption following tooth extraction. *The Journal of Prosthetic Dentistry*. 1967;17(1):21-27. doi:10.1016/0022-3913(67)90046-7
2. Trombelli L, Farina R, Marzola A, Bozzi L, Liljenberg B, Lindhe J. Modeling and remodeling of human extraction sockets. *Journal of Clinical Periodontology*. 2008;35(7):630-639. doi:10.1111/j.1600-051x.2008.01246.x
3. Cardaropoli G, Araujo M, Lindhe J. Dynamics of bone tissue formation in tooth extraction sites. An experimental study in dogs. *Journal of Clinical Periodontology*. 2003;30(9):809-818. doi:10.1034/j.1600-051x.2003.00366.x
4. Araújo MG, Lindhe J. Dimensional ridge alterations following tooth extraction. An experimental study in the dog. *Journal of Clinical Periodontology*. 2005;32(2):212-218. doi:10.1111/j.1600-051X.2005.00642.x
5. Lekovic V, Kenney EB, Weinlaender M, et al. A bone regenerative approach to alveolar ridge maintenance following tooth extraction. Report of 10 cases. *Journal of Periodontology*. 1997;68(6):563-570. doi:10.1902/jop.1997.68.6.563
6. Schropp L, Wenzel A, Kostopoulos L, Karring T. Bone healing and soft tissue contour changes following single-tooth extraction: a clinical and radiographic 12-month prospective study. *The International Journal of Periodontics & Restorative Dentistry*. 2003;23(4):313-323.
7. Engler-Hamm D, Cheung WS, Yen A, Stark PC, Griffin T. Ridge Preservation Using a Composite Bone Graft and a Bioabsorbable Membrane With and Without Primary Wound Closure: A Comparative Clinical Trial. *Journal of Periodontology*. 2011;82(3):377-387. doi:10.1902/jop.2010.090342
8. Dimova C. Socket Preservation Procedure after Tooth Extraction. *Key Engineering Materials*. 2013;587:325-330. doi:10.4028/www.scientific.net/KEM.587.325
9. Cardaropoli D, Cardaropoli G. Preservation of the postextraction alveolar ridge: a clinical and histologic study. *The International Journal of Periodontics & Restorative Dentistry*. 2008;28(5):469-477.
10. Barone A, Aldini NN, Fini M, Giardino R, Calvo Guirado JL, Covani U. Xenograft Versus Extraction Alone for Ridge Preservation After Tooth Removal: A Clinical and Histomorphometric Study. *Journal of Periodontology*. 2008;79(8):1370-1377. doi:10.1902/jop.2008.070628
11. Gardin C, Ricci S, Ferroni L, et al. Decellularization and Delipidation Protocols of Bovine Bone and Pericardium for Bone Grafting and Guided Bone Regeneration Procedures. *PLOS ONE*. 2015;10(7):e0132344. doi:10.1371/journal.pone.0132344
12. Ludovichetti F, Ludovichetti M. Utilizzo di osso bovino decellularizzato e delipidato e di una membrana in pericardio per la rigenerazione di alveoli post-estrattivi e successiva riabilitazione implanto-protetica: risultati clinici e istologici. *Dental Tribune Italy*. Published online April 14, 2020.
13. Tomasi C, Tessarolo F, Caola I, et al. Early healing of peri-implant mucosa in man. *Journal of Clinical Periodontology*. 2016;43(10):816-824. doi:10.1111/jcpe.12591
14. Tan WL, Wong TLT, Wong MCM, Lang NP. A systematic review of post-extraction alveolar hard and soft tissue dimensional changes in humans. *Clinical Oral Implants Research*. 2011;23(Suppl 5):1-21. doi:10.1111/j.1600-0501.2011.02375.x
15. Sisti A, Canullo L, Mottola MP, Covani U, Barone A, Botticelli D. Clinical evaluation of a ridge augmentation procedure for the severely resorbed alveolar socket: multicenter randomized controlled trial, preliminary results. *Clinical Oral Implants Research*. 2011;23(5):526-535. doi:10.1111/j.1600-0501.2011.02386.x
16. Hong B, Bulsara Y, Gorecki P, Dietrich T. Minimally invasive vertical versus conventional tooth extraction. *The Journal of the American Dental Association*. 2018;149(8):688-695. doi:10.1016/j.adaj.2018.03.022
17. Yang J, Lee HM, Vernino A. Ridge preservation of dentition with severe periodontitis. *Compendium of Continuing Education in Dentistry*. 2000;21(7):579-583; quiz 584.
18. Koo T, Song YW, Cha J, Jung U, Kim C, Lee J. Histologic analysis following grafting of damaged extraction sockets using deproteinized bovine or porcine bone mineral: A randomized clinical trial. *Clinical Oral Implants Research*. 2019;31(1):93-102. doi:10.1111/clr.13557
19. Rasperini G, Canullo L, Dellavia C, Pellegrini G, Simion M. Socket grafting in the posterior maxilla reduces the need for sinus augmentation. *The International Journal of Periodontics & Restorative Dentistry*. 2010;30(3):265-273.

20. Zhao L, Xu T, Hu W, Chung KH. Preservation and augmentation of molar extraction sites affected by severe bone defect due to advanced periodontitis: A prospective clinical trial. *Clinical Implant Dentistry and Related Research*. 2018;20(3):333-344. doi:10.1111/cid.12585
21. Carmagnola D, Adriaens P, Berglundh T. Healing of human extraction sockets filled with Bio-OssR. *Clinical Oral Implants Research*. 2003;14(2):137-143. doi:10.1034/j.1600-0501.2003.140201.x
22. Cardaropoli D, Tamagnone L, Roffredo A, Gaveglio L, Cardaropoli G. Socket preservation using bovine bone mineral and collagen membrane: a randomized controlled clinical trial with histologic analysis. *The International Journal of Periodontics & Restorative Dentistry*. 2012;32(4):421-430.
23. Lee JS, Choe SH, Cha JK, Seo GY, Kim CS. Radiographic and histologic observations of sequential healing processes following ridge augmentation after tooth extraction in buccal-bone-deficient extraction sockets in beagle dogs. *Journal of Clinical Periodontology*. 2018;45(11):1388-1397. doi:10.1111/jcpe.13014
24. Tomasi C, Donati M, Cecchinato D, Szathvary I, Corrà E, Lindhe J. Effect of socket grafting with deproteinized bone mineral: An RCT on dimensional alterations after 6 months. *Clinical Oral Implants Research*. 2018;29(5):435-442. doi:10.1111/clr.13141