

Original Article

EFFECT OF A NEW XENOGRAFT MATERIAL IN MANDIBULAR POST-EXTRACTION SITES: A CASE SERIES

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ABSTRACT

The alveolus bone is a tooth-dependent tissue. The extraction of the dental element determines the resorption of the alveolar crest, which trophism is linked to the presence of the periodontal ligament. Several materials have been used to improve alveolar bone healing and maintain alveolar ridge. The aim of the study is to evaluate the effects of a new matrix of bovine bone processed at low temperature in association with a membrane of the bovine pericardium in post-extraction sites using histological analysis comparing treated and untreated alveoli. Five patients with non-recoverable teeth were enrolled in the present study for teeth extraction. In treated sites, the alveolus was packed with Decellularized and Antigen-free Bovine Bone (RE-BONE® Ubgen, Padova, Italy) and subsequently covered with a bovine-derived pericardium membrane (SHELTER® FAST Ubgen, Padova, Italy). Four alveoli of two patients were left to heal spontaneously as control sites. The tissue sampling was performed during the implant site preparation four months after extraction. Specimens were decalcified, and sections were stained with hematoxylin and eosin. Bone histomorphometry of regeneration tissues from treated sites showed an average increase of 2.9% in bone tissue. However, no statistically significant differences can be detected since standard deviations are very high. Generally, the alveolar preservation technique is a valuable method to guarantee alveolar volume stability. The material studied here showed a slight increase in bone production after 4 months from a tooth extraction in treated sites, which is an expression of a good healing process. However, since the limited number of cases analyzed, additional studies are needed to verify the bone gain in alveolar bone healing.

KEYWORDS: bone, graft, alveolus, mandible, lower jaw

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INTRODUCTION

The alveolus bone is a tooth-dependent tissue. The extraction of the dental element determines the resorption of the alveolar crest, which trophism is linked to the presence of the periodontal ligament. Tooth extraction leads to a reduction of alveolar ridge (1-4).

Bone resorption is most evident in sites where the thickness of the cortical bone is thin (5) or where the root anatomy of the teeth is more prominent in the vestibular sense (6, 7). Furthermore, human and animal studies showed that most of the tissue lost in the initial phase occurred in the coronal part, while the apical part was less affected. In untreated sites, continuous remodelling occurs over time, and a significant variability in ridge resorption between subject/site exists (8, 9). The studies of Pietrokovski & Massler (2) and Schropp et al. (3) performed on plaster models in which one tooth was extracted on one side while the contralateral tooth maintained a more than double resorption in the vestibular compared to the lingual part. Authors reported a bone loss of 30% at 3 months and 50% after 12 months (with a mean > 6mm), while the mesial and distal parts underwent reduced resorbing.

Animal models were used to study graft materials to counteract ridge remodelling following extraction. In these studies, biomaterial was inserted in post-extractive sockets (10).

These studies failed to demonstrate that the biomaterials entirely prevent the resorption of the buccal wall and the remodelling of the ridge in a general sense but showed that their use, under certain conditions, significantly reduced alveolar crest resorption (8-11). Alveolar Socket Preservation (ASP) is defined as "any procedure undertaken at the time or following the extraction, aimed at minimizing the external resorption of the ridge and maximizing the formation of bone within the alveolus" (12).

Since a new biomaterial has been recently introduced in the marker (13-18), we decided to evaluate the healing process of the post-extractive socket after 4 months by inserting a new matrix of bovine bone processed at low temperature and covered with a membrane in the bovine pericardium.

MATERIAL AND METHODS

Patients were enrolled based on the following criteria: non-recoverable teeth due to destructive caries, traumatic events (i.e., vertical root fracture), endodontic treatments (i.e., teeth no longer retractable). Five test sites and four control sites were evaluated for the study. Three female patients and two males, mean age of 61 years, were enrolled for test sites. The five test alveoli were one premolar and four molars. Two male patients, mean age of 52.5 years, were enrolled as controls. One premolar and three molars were investigated.

Exclusion criteria were as follows: patients smoking more than 10 cigarettes/day, pregnant women, patients with chronic diseases (diabetes, orofacial neoplasms, etc.), bisphosphonate therapy, or with an acute infection in progress, untreated periodontitis, autoimmune diseases, allergies to one or more materials, drugs used during treatment, alcohol and/or drugs intake.

Surgical procedure

During the extraction, an attempt was made to lift the flaps in the least invasive way possible to preserve the alveolus from further resorption due to surgical exposure. The alveolus was packed with Decellularized and Antigenfree Bovine Bone (RE-BONE® Ubgen, Padova, Italy) and subsequently covered with a bovine-derived pericardium membrane (SHELTER® FAST Ubgen, Padova, Italy). The same surgical procedure was used for the control sites, but no biomaterial was grafted, and the alveoli were left to heal spontaneously.

Compression sutures were performed in monofilament in e-PTFE (Gore-Tex®), removed after 10 days, anti-inflammatory therapy with Nimesulide was prescribed as well as soft and cold diet for at least 2/3 days, ice packs



Fig. 1. Pre-surgical image

for few hours were delivered as well as rinses with Chlorhexidine 2/3 times a day for 15/20 days. Monthly checks were carried out until the fourth month, when bone sampling was scheduled.



Fig. 2. Pre-surgical radiograph



Fig. 3. Socket preservation I



Fig. 4. Socket preservation II

Fig. 7. Prosthetic restoration



Fig. 5. *Re-entry after 4 months for implant insertion*



Fig. 6. Abutment

The tissue sampling was performed during the preparation of the implant site, using a 2 mm core drill for a depth between 2 to 3 mm. The bone samples were placed in sterile and labelled blisters and immersed in formalin 10% (Merck, Darmstadt, Germany) and subsequently sent to the laboratory for histological evaluations (Fig. 1-8).

Histological analysis

The bone samples were decalcified with Osteosoft[®] and subsequently embedded in paraffin. A microtome (RM2025 Leica Instruments, Nussloch, Germany) was used to obtain a 5 μ m thick section. These paraffin sections,



Fig. 8. Radiographic check

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 Merck (Darmstadt, Germany).

For the subsequent analysis, 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. Finally, the .svs files were viewed and analyzed using a free software program called ImageScope.

RESULTS

The tissue samples (obtained by a core drill during implant site preparation) were decalcified, and sections were stained with hematoxylin and eosin. The analysis by ImageScope software of the scanned histology slides quantified the length of the sample, the total area, the percentage of bone and fibrous tissue, and, when present, the area with residues of bovine-derived pericardium membranes.

Fig. 9 shows control and test samples with the respective magnifications of connective and bone tissues. 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 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.

Table I summarized histomorphometric results showing an average increase of 2.9% in the bone area in the treated samples compared to the controls. However, no statistically significant differences can be detected since standard deviations are very high.

DISCUSSION

Healing in a post-extraction socket occurs through a series of events, including clot formation and maturation, matrix



Fig. 9. Images of a control sample and a test sample and the respective magnifications

	Average values							
	Age	Length of	Analyzed area	% bone	% connective	% shelter®		
		samples				residues		
Control samples	52.5	2.2 (±1.1)	3.1 (± 1,1)	59.6 (± 5.8)	35 (± 3.4)	5.4 (± 2.8)		
Test samples	60.8	2.3 (±0.3)	$2.7 (\pm 0.8)$	62.5 (± 18.1)	31.8 (± 31,8)	5.7 (± 9.9)		

Table I. Average values obtained from the analysis.

deposition and mineralization. Usually, the residual ridge decreases by 15% at six months, both vertically and horizontally (19). This dimensional change can lead to aesthetic and functional disadvantages for the subsequent placement of the implant since an adequate residual ridge width is one of the main prerogatives for the long-term success of prosthetically guided implant therapy (19).

Biomaterials and/or biological agents such as autologous bone, bioactive glasses, hydroxyapatite, human-derived bone (allografts), and especially animal-derived bone (xenografts) were used and analyzed to counteract the alveolar ridge resorption and make the site available for the insertion of an implant (20, 21). It has been shown that these biomaterials could be embedded in a newly formed bone, kept as inactive fillers, or reabsorbed by the host tissue during its natural remodelling course (22).

Although the ability of biomaterials to decrease resorption and preserve adequate edentulous ridge volumes has been extensively documented in the literature, the quality of grafted tissue has not yet been widely understood.

A De Risi et al. (21) meta-analysis showed that no histological differences exist between the different procedures compared to spontaneous healing. The highest percentage values of regenerated bone at 3 months came from procedures using allografts (54.4%), while the lowest at 5 months were those using xenografts (23.6%). Regarding the presence of connective tissue in the grafted sites, the highest value at 7 months was referred to allografts (67%), and the lowest to the alveoli treated with alloplast (27%). As for the residual biomaterial, the lowest percentages were attributable to sites with allografts (12.4 - 21.11%), while those with xenografts and alloplast showed better results at 7 months (37.14 - 37.23%) (21).

In our report, although a slight increase in bone formation was detected in treated alveoli (2.9%), no statistically significant differences were obtained due to the great standard deviation value. This fact is probably related to the small sample size.

In the literature, no differences were highlighted regarding the superiority of an alveolar preservation technique over others (i.e., GBR, site filling, site sealing) regarding the three-dimensional preservation of the site, bone formation, amount of keratinized tissue and complications (20-23).

In a systematic review, Chan et al. (24) analyzed the proportion between bone and connective tissue in grafted and untreated alveoli. They found that in ungrafted sites, the percentage of vital bone and connective tissue was $38.5\% \pm 13.4\%$ and $58.3\% \pm 10.6\%$, respectively.

Chan et al. (24), reported that four studies investigating the effect of xenografts gave the most contrasting results: the presence of vital bone ranged from - 22% (decreased) to + 9.8% (increased), instead alloplastic grafts increased the amount of vital bone from 6.2% to 23.5%. Furthermore, many residual biomaterials were noted when hydroxyapatite and xenografts were used, ranging from 15% to 36% of the healed alveolus.

Using grafting materials for ASP might change the proportion of vital bone compared to sockets allowed to heal without grafting. In 2020 Koo et al. (25) compared two xenografts, one of bovine and one of porcine derivation. Histology was comparable in the percentages of newly formed bone, residual connective tissue and residual graft particles at 4 months.

CONCLUSION

In conclusion, the English literature shows that alveolar preservation techniques provide acceptable tissue volumes for implant therapy, but bone quality does not show significant differences between the various biomaterials with respect to spontaneous healing. In our report, although a slight increase in bone formation was detected in treated alveoli (2.9%), no statistically significant differences were obtained due to the great standard deviation value. This fact is probably related to the small sample size, so additional studies are needed.

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