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Morphometric Changes of the Socket after Site Preservation Using Nanobone and Collagen Membrane or Stypro Versus Extraction Alone

Salahi S^a, Etemadifar R^b, Moosaali F^a

- a. Department of Periodontics, Faculty of Dentistry, Kerman University of Medical Sciences, Kerman, Iran.
- b. Department of Periodontics, Faculty of Dentistry, Bandar Abbas University of Medical Sciences, Hormozgan, Iran.

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 $Corresponding\ Author:$

Fereshteh Moosaali

Department of Periodontics, Faculty of Dentistry, Kerman University of Medical Sciences, Kerman, Iran

Fax:+98-71-36270325 Email: moosaaly@kmu.ac.ir

Tel: +98-71-36280456

Abstract

Statement of Problem: The long-term success of a dental implant relies on implant osseointegration into native and viable bone, implant placement in an ideal position, and optimal hard and soft tissue contour. This requires the presence of sufficient alveolar bone volume, good alveolar ridge (Practically with no sign of atrophy) and good surgical technique.

Objectives: The aim of this randomized controlled clinical study was to evaluate morphometric changes after different alveolar ridge preservation procedures.

Materials and Methods: In this study, 33 patients who had single-rooted premolar, which needed to be extracted, were recruited. Patients were randomly divided into 3 groups and after tooth extraction the following treatments were administered: in group A: NanoBone and a collagen membrane; in group B: NanoBone and Stypro; and in group C: natural healing. The following clinical parameters were evaluated at baseline and 6 months after the extraction: buccolingual width, midbuccal height (with the use of a custom made stent) and width of keratinized gingiva. For data analysis, Paired t-test, one-way ANOVA and Tukey's tests were used.

Results: The average reduction in the buccolingual width, midbuccal height and keratinized gingiva was as follows: group A: 1.18 ± 0.6 , 0.64 ± 0.92 and 3.45 ± 1.75 mm; group B: 2.18 ± 0.75 , 0.73 ± 0.78 and 4.73 ± 0.9 mm; and group C: 1 ± 0.89 , 2.36 ± 1.21 and 5 ± 0.63 mm, respectively. Moreover, a significantly reduced resorption was found in both the buccolingual width and the width of keratinized gingiva in group A as compared to groups B and C (p<0.05).

Conclusions: This study showed that the use of collagen membrane+Nano bone (group A) can significantly reduce the horizontal resorption of the alveolar ridge and keratinized tissue more effectively than stypro+Nano bone (group B) and blood clot alone and natural healing (group C).

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Introduction

Infection, trauma, malpositioned teeth, or mechanical trauma often result in bone loss [BL]. Prior to tooth removal, and during tooth extraction procedure, BL can occur as a result of traumatic extraction. Postextraction alveolar ridge resorption is a common phenomenon that impairs the replacement of dental implant and may cause esthetic and surgical problems in prosthetic dentistry [1,2]. Post-extraction alveolar process resorption in the buccal wall is more pronounced than that in the lingual/palatal wall. Following the atrophy, the center of the ridge will move in lingual/palatal direction [2-4]. The resorption of the buccal/lingual walls of the extraction site occurs in two overlapping phases. During the early phase of remodeling, firstly the bundle bone undergoes resorption and will be replaced with the woven bone. Since the crest of the buccal bone wall is comprised solely of bundle, this modeling will result in substantial vertical reduction of the buccal crest [2,4-6]. Phase 2 included resorption which occurred from the outer surfaces of both bone walls [5-7].

In the horizontal plane, BL occurs largely at the expense of the facial cortical plate, increasing the risk of facial soft tissue recession, especially in the presence of a thin periodontal biotype [1,3]. Interdental BL may lead to the loss of the interdental papilla. It was reported that an alveolar bone shrinkage of 40-60% will occur within the first 2-3 years and then it will continue at a rate of 0.25-0.5% per year until death [5]. Another study found that most of the gain was achieved from 3 to 12 months following extraction, whereas almost the entire loss of height took place during the first 3 months. The reduction in the ridge width is approximately 50%, two thirds of which occur during the first 3 months [2,8]. The percentage of reduction is somewhat larger in the molar regions than in the premolar regions, and in the mandible compared with the maxilla [2].

The correct technique for extraction should lead to atraumatic extraction. The fundamental requirements for a good extraction are adequate access and visualization of the field of surgery, an unimpeded pathway for the removal of the tooth and the use of controlled force to luxate and remove the tooth. A 4-month study on dogs showed that tooth extraction after the elevation of the mucosal flaps with or without using socket-preservation techniques during the period of healing caused significantly softer and hard tissue reduction

than a flapless tooth removal. The authors concluded that the exposure of the buccal bone has a detrimental effect on the resorption process occurring after tooth extraction [9].

Arau'jo and Lindhe (2009) in another experiment on five mongrel dogs within a period of 6 months showed that, in contrast to apical/middle portions of the socket site, the dimensional alterations in the coronal portion of the ridge are substantial [10]. The differences between the mentioned study and the previous one may result from different treatment periods, i.e. 6 vs. 4 months. Using the grafting material for preservation and reconstruction of the remaining socket immediately after tooth extraction is recommended [11]. Horowitz et al. (2012) in a review study concluded that there was a benefit in alveolar ridge preservation [ARP]. The researchers did not find any grafting materials, demonstrating a clear benefit over any other material or that a barrier membrane is necessary; they noted that the evidence is too premature about whether socket preservation efforts require primary closure [11,12]. A review (2014) of randomized controlled studies demonstrated that all accepted therapeutic procedures for ridge preservation are more effective than leaving the socket to be filled with blood clot alone [12].

Several techniques have been employed as ridge preservation procedures involving the use of bone grafts, barrier membranes and biologic materials to provide a better restorative outcome. Nanobone and Stypro are a kind of new materials used in this study. NanoBone (ARTOSS GmbH. Friedrich-Barnewitz) is a granular material consisting of nanocrystalline HA embedded in a silica gel matrix. The internal surface of Nanobone is about 84 square meter per gram. The interconnecting pores in the silica gel have sizes ranging from 10 to 20 nm, leading to material porosity of about 60%. Stypro (Pro-tec GmbH, Lindigstr 4, d-63801 Kleinostheim) is a sterile, highly porous, implantable and resorbable sponge used by surgeons and dentists to control bleeding (haemostasis) and improve wound healing [13]. Stypro sponges are made from medicinal gelatine (Pharm. Eur.) and retain the capability, inherent to collagen, to initiate blood clotting [14]. Collagen membrane (BGG, Bioteck Sri, Italy) is made of collagen from the Achille's tendon.

The aim of this study is to compare the two approaches of Stypro+NanoBone and collagen membrane+NanoBone to preserve soft and hard tissue through investigating whether the former which does

Table 1: The variables measured		
Variable	Description	Instrument
BLW^1	The distance between mid-buccal and mid-lingual ridge	Bone caliper
MBH^2	The distance between margin of surgical stent and bone crest	Periodontal probe
KGW ³	The distance between margin of stent and MGJ	Periodontal probe

1-Buccolingual socket width, 2- The height of mid-buccal bone, 3- The width of keratinized gingiva

not uses coronally advanced flap offers more advantages than the later.

Materials and Methods

This study was a randomized controlled clinical trial. The study population consisted of patients whose premolars needed to be extracted because of reconstructive or endodontic reasons. The sample size was calculated to be 33. The selected sampling method was convenience sampling. Given the parameters of the error (α =0.05), test power (1- β =0.9) and effect size=2.9, sample size of 11 patients per group was calculated. Using systematic random sampling, the samples were divided into 3 groups of A, B and C. After tooth extraction, the following treatments were administered: in group A: NanoBone and a collagen membrane; in group B: NanoBone and Stypro; and in group C: natural healing.

The intervention was conducted in two groups: A and B. The control group consisted of those who did not receive any intervention. Clinical evaluation of the extraction site was carried out at baseline (immediately after tooth extraction) and 6 months after tooth extraction.

Tension-free suturing is one of the best techniques for the success of periodontal treatment. In group A, after periosteal dissection, the flap was coronally advanced, while in group B, the flap was sutured without periosteal incision and no primary effort was made for the closure of the flap. Both groups (A and B) were sutured with vertical mattress sutures in combination with a simple interrupted suture (so-called 'Laurell suture') [15]. In the control group, the flap was sutured without periosteal incision with loop suture through the interdental papilla site.

The study was conducted in the Department of Periodontology of Kerman Dental College in Iran. The patients' mean age was 30 years (patients were between 20 to 40 years). They were given oral and written information regarding the study; the recommended treatment was thoroughly explained to each patient and their written informed consent was obtained. The

study protocol was approved by the ethics committee of Kerman University of Medical Sciences.

Inclusion criteria were single-rooted tooth indicated for extraction and inability to correct the problem of the tooth by periodontal or endodontic treatment.

Exclusion criteria were presence of underlying systemic disease with detrimental effects on wound healing, smoking, pregnancy, severe periodontal disease, fracture of the buccal or lingual/palatal wall during tooth extraction, surgical wound dehiscence, and the infection of the membrane or extrusion of the bone graft material.

*Preparing surgical stent

In the first meeting, the patient's jaw impression body was made from alginate impression material and the east was made from the mold. By using the prepared cast, a surgical stent was made for each sample of the corresponding site. In this study, acetate stent was used.

*Pre-extraction measurements

In the second meeting, with respect to the stent, a caliper was used to measure the buccolingual ridge width (BLW), which is the distance between the mid-buccal point and mid-palatal/lingual around the tooth that is exactly under the stent edge (Table 1). To measure the height of the mid-buccal bone (MBH), the prepared surgical stent was placed on the ridge of the patient. To avoid bias, primary and secondary measurements were performed by a resident who was blind to the groups; furthermore scaling and surgery were performed by another resident.

To preserve the integrity of the alveolar septum and buccal wall, surgical planning was based on atraumatic extraction of the compromised tooth with periotome and forceps. To achieve this goal, the extraction had to be conducted as gently as possible to minimize the possible post-extraction trauma. Therefore, after local anesthesia had been administered, sulcular incisions were given around the premolars with a sharp instrument such as a No.15 scalpel. Periotome was used to dissect the attached fibers. The blade had to be angled at 20 degrees to ensure that the tip of the periotome was within the crest of the alveo-

lar bone and not out of the ridge. The instrument was inserted first in the gingival sulcus and then in the periodontal ligament space. The periotome was moved repeatedly in a mesio-distal direction, along the circumference of the root. It was possible to reach up to two thirds of the root length by repeating this movement many times. After finishing this procedure, the tooth was attached to the alveolus only by the most apical part of the periodontal ligament.

Finally, the extraction of the tooth was performed with forceps without distorting or damaging the alveolar bone. Following tooth extraction, the extraction socket was thoroughly debrided to remove any remnants of tooth fragments. After atraumatic extraction and debridement of the socket, clinical measurements were performed from the extraction site for vertical and horizontal dimensions.

Following the tooth extraction, Ibuprofen (400 mg) was administered every 6 hours to relieve pain in patients experiencing severe ache. All subjects were instructed to abstain from oral hygiene procedures or any hard or hot food materials while performing a twice-daily rinse with a 0.2 percent chlorhexidine solution for 2 weeks. All these recommendations were asked to be visited after 2 weeks to evaluate the healing process and remove the sutures, and then 6 months after tooth extraction to measure the alterations that had occurred during the treatment. To analyze the data, descriptive statistics (tables, graphs and etc.) were used. To evaluate the changes in the parameters, paired t-test was applied to compare the baseline and 6-month measurements. The significance was set at p<0.05. One-way ANOVA /Tukey's tests ware used to compare the mean changes in the parameters between groups.

Results

In this study, a total of 33 patients were recruited. The patients were referred for extraction with due to root fracture, endodontic failure, and periodontal disease. Post-treatment period was uneventful in all patients and, accordingly, no local complications and infections were reported during the 6-month period. Vertical bone alterations as well as the width of keratinized gingival [KGW] were evaluated at baseline and 6 months post-extraction, with the use of a custom made stent at the bucco-lingual and mid-buccal site, respectively.

According to the study, the BLW of the alveolar

ridge in test groups A and B versus the control group C was in the range of 0-2, 1-3 and 0-4, respectively. Paired sample t-tests of width measurements of buccolingual ridge and keratinized gingiva in groups A, B and C all showed statistically significant differences, between baseline and 6 months after tooth extraction (p-value <0.05). BLW change of group A was 1.18±0.6, which was significantly lower than those of group B (2.18±0.75, p-value=0.034) and group C (2.36±1.21, p-value=0.011).

Regarding keratinized gingiva variations, the measurements 6 months later indicated that an average resorption of 3.45 ± 1.75 mm (range: 2 to 7 mm), 4.73 ± 0.9 mm (range: 3 to 6 mm) and 5 ± 0.63 mm (range: 4 to 6 mm) had occurred, respectively, in the groups A, B and C. By evaluating the differences in betweengroup, there were statistical differences between group A and the other two groups, and there was no statistical difference between group B and C (p > 0.05). Moreover, In terms of KGW, group A revealed statistically significantly lower resorption rates when compared with B and C. It seems that the membrane can limit the recession of KGW while Stypro or using no material cannot do so.

The control group lost ≥1 mm of the midbuccal ridge height, whereas the case group lost <1 mm of the midbuccal ridge height. The average alteration of MBH in group A was 0.64±0.92 mm (range 0-3) and in groups B and C it was 0.73±0.78 mm and 1±0.89 mm, respectively with the range of 0-2 in both groups. In the all three groups, a significant decrease in the MBH was detected at 6 months when compared with baseline. No significant difference was found with regard to the MBH when comparing different groups two by two.

Discussion

Post-extraction progressive three-dimensional resorption in the alveolar ridge is an inevitable process and the premolar area is not an exception. Consequently, socket grafting techniques have been readily adopted by dentists throughout the world. A great amount of research has been conducted to examine the effectiveness of several materials or membranes. The use of invasive techniques is hardly recommended at this treatment time point, since any procedure requiring primary healing intention with the advancement of flaps may result in increased inflammatory response, a decrease in vestibular depth, and creation of unaes-

thetic scars [6]. The greater part of this combined hard and soft tissue atrophy takes place in the first 3 months after extraction [6,16]. Accordingly, bony and connective tissue augmentations are necessary to allow prosthetically correct positioning of the implants at such sites [17].

Various concepts for the treatment of extraction socket have been presented previously. These methods range from covering the extraction socket with resorbable and non-resorbable membranes to filling them with resorbable and nonresorbable bone replacement materials [6,18]. A reasonable choice is to establish a surgery with osteo-conductive bone graft with low resorption rate which is covered with a resorbable membrane because this method can sufficiently maintain the volume and contour of the ridge as well as providing aesthetically acceptable result [18].

The application of the NanoBone offers an alternative to autogenous bone blocks. NanoBone is a highly porous material. Proteins such as fibrins adsorb on the nanostructures, forming a proteinaceous matrix, which invaded by small vessels and cells. This makes them more attractive for the cells which initiate bone formation. The body accepts the NanoBone block as if it is endogenous, so the material produces no foreign body reactions or inflammation [13].

Stypro is a sterile, resorbable and implantable gelatine sponge of porcine origin. It has a rapid haemostatic effect and excellent tolerance and biocompatibility, without thrombin or other additives. Furthermore, this material is shown to be chemotactic for monocytes and has a mitogenic effect on both mesenchymal and fibroblast cells. Overall, these effects hasten epithelial tissue regeneration [14].

Flap elevation is a factor that may have an effect on bone remodeling process after ARP, often resulting in poor blood supply, more bone loss, delayed wound healing, and compromised soft tissue appearance [10]. However, related studies have demonstrated controversial results. Therefore, in this study, in order to create the same conditions for all patients, tooth extraction was done by full thickness flap in all cases.

Comparison of clinical variables among the three groups after 6 months showed that the width of alveolar ridge and keratinized gingiva were better preserved in the test group A compared to the control group. It should be taken into consideration that some studies showed less bone reabsorption in ARP sites after 6 months compared to the control sites [19-23]. Lekovic *et al.* (1997) conducted a study on 10 patients who

needed two or more anterior teeth extractions to evaluate the effect of ARP on the alveolar ridge preservation. Extraction procedures were done with a full thickness surgical flap approach and minimum of trauma to the surrounding bone. They showed that 6 months after the extraction, clinical and model measurements have shown a statistically significant better ridge dimensions at the experimental sites (covered with an expanded polytetrafluoroethylene (ePTFE) barrier membrane) than those at the control group (p ≤0.05). In 3 patients with exposed membranes, dimensional changes were similar to those of the control group [18]. The results of our study showed that, on average, the alterations of the alveolar ridge were more predominant in groups C and B compared to group A. These data support the results reported by Lekovic *et al.* [19].

Iasella et al. (2003) investigated the effects of FDBA and a collagen membrane versus extraction alone during a 6-month period in 24 patients (10 males and 14 females) with the mean age of 51.5 who required extraction of their single rooted tooth. The changes of the horizontal ridge width for the test and control groups were 1.2±0.9 mm and 2.6±2.3 mm as well as vertical changes of 1.3±2 mm and 0.9±1.6 mm, respectively [20]. This is consistent with the results of the present study in which the vertical ridge height exhibited no significant difference between extraction and ARP groups although the vertical changes in each group were significant between baseline and 6 months post-extraction. It seems that one of the reasons for the lack of access to optimal treatment results in group B could be lack of Stypro's ability to keep NanoBone quite inside the socket during the early stages of healing.

A clinical and histomorphometric study on forty subjects requiring tooth extraction and implant placement evaluated Xenograft versus extraction alone for ridge preservation [21]. The researchers assessed the desired parameters immediately after extraction and 7 months prior to implant placement. They concluded that ridge-preservation approach using the porcine bone in combination with the collagen membrane significantly limited the resorption of hard tissue ridge after tooth extraction compared to extraction alone. It was consistent with the result of Iasella's study in using collagen membrane. Similar results were seen in the present work, the least loss was seen in group A that was based on Nanobone and collagen membrane for ARP. Likewise, a significantly greater horizontal

resorption was observed in group C (extraction alone sites) (2.36 ± 1.21) and group B (2.18 ± 0.75) compared to group A (1.18 ± 0.6) ; this is in accordance with Baron's study: $(4.3\pm0.8 \text{ mm})$ vs. $(2.5\pm1.2 \text{ mm})$ for the test and control groups, respectively. Although both groups showed a significant vertical reduction from baseline to the final examination, the Tukey's test showed no significant vertical recession between the groups. Moreover, the ridge changes were statistically significant for the buccal and lingual sites.

Cardaropoli (2008) in a study with a 4-month period on 10 patients (10 single extraction sites in the posterior area received a bone substitute in which the osteoconductive material was covered by a collagen membrane in all cases) showed a reduction of 1.85±1.65 in the horizontal dimension of the alveolar ridge; this is in agreement with our study. However, they noted that the statistical difference between the initial and final bone width was not significant [4]. This could be due to more time in the current study for repair of the soft and hard tissues following tooth extraction, i.e. 2 months more in comparison with Cardaropoli's study. So during this time period more resorption has occurred.

One important difference between the current study and the experiment done by Blanco *et al.* (2008) is the length of the healing period, i.e. 6 vs. 3 months. For example, it has been shown by Schropp *et al.* (2003) that dimensional changes following tooth extraction are not completed after 3 months but between 3 and 12 months, additional resorption and reduction will occur. The data from the present experiment, therefore, suggest that some differences between groups B and C would probably occur after longer healing periods.

Our results clearly indicate that the use of Nano-Bone and collagen membrane succeeded in reducing the alveolar ridge from remodeling when compared to other groups. This means that group A outperformed all others. Recently, Thalmair *et al.* (2013) [22] performed a study on 30 patients randomly assigned to a treatment. Their results showed the best outcome which was related to the Tx1 group (xenogenic bone substitute-prehydratedcollagenatedcortico-cancellous porcine bone); this supports our study more as they noted that horizontal contour shrinkage at the buccal aspect during a 4-month healing period in all groups was observed and it ranged from 0.8±0.5 mm (Tx1) to 2.3±1.1 mm (Tx4(no further treatment)/Control). The comparison of the groups by unpaired t-tests (one-way

ANOVA) showed significant differences in dimensional change between the test groups Tx1 and Tx2 (free gingival graft alone) compared with the control group Tx4. Their results, as well as those of Pelegrine's study [23], are completely in the same line with those of the present study in terms of horizontal ridge changes. Finally, despite the use of NanoBone in the socket and Stypro to cover the socket, in the present study, the results were not significant compared with the control group and this could be due to the inability of Stypro in improving the soft tissue, namely Stypro as a main influencing factor makes the results more similar to the natural healing.

Conclusions

In contrast to the use of Stypro+NanoBone or natural healing, the utilization of collagen membrane+ NanoBone is superior to the other therapies. Given the limitations of this study (the limited follow-up time), the following conclusions can be drawn from the comparison of Group A with Groups B and C:

- 1) Nanobone and collagen membrane can be used to limit the dimensional changes of the alveolar ridge.
- 2) During a 6-month observation period, vertical changes of the alveolar ridge were minimum in comparison to the other parameters. Moreover, the mean differences were not significant in all three groups.
- 3) No significant differences were observed in treatment outcomes of group B versus C; therefore, the use of Nanobone+Stypro is not recommended because its usage not only doesn't improve the treatment outcome but also increases the cost.

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