Alveolar ridge and keratinized gingiva preservation using collagen matrix and inorganic bone substitute in flapless extractions: A case series of exposed biomaterials

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Abstract

Aim: To evaluate the dimensional changes after alveolar ridge preservation (ARP) with flapless and flapped techniques, using demineralized bovine bone mineral (DBBM) and a collagen matrix (CM) intentionally left exposed.

Materials and methods: In this case series, randomly selected patients were divided into one of two surgical approaches, Group 1 (G1): ARP flapless and Group 2 (G2): ARP flapped. Clinical and cone beam computed tomography assessments were performed at 1 week, 4 and 24 months after ARP. Evaluations of postoperative discomfort with visual analogue scale (VAS) were also performed.

Results: Surgical procedures run uneventfully with no healing complications of the treated sockets. There were reductions in the horizontal and vertical dimensions of the socket and in the width of the keratinized gingiva, but they were smaller for G1. The thickness of the keratinized gingiva increased in G1 and reduced in G2. Only the VAS had a statistically significant difference between the groups (P=0.03).

Conclusions: The ARP limited vertical and horizontal socket changes, regardless of the surgical technique used when the biomaterials where left exposed. The flapless approach seems to provide better results regarding dimensional changes and significantly less discomfort.

Keywords: Bone regeneration; Bone substitutes; Cone-beam computed tomography; Tissue grafting; Tooth extraction.

Introduction

After tooth extraction, occur a series of repair processes involved in healing which cause bone resorption and consequent loss of contour and initial volume of the socket in hard and soft tissues (Hämmerle *et al*., 2012; Araújo *et al*., 2015a). The remodelling process begins immediately after tooth extraction and after 2 years 40%-60% of bone resorption is detected (Araújo and Lindhe, 2009; Tan *et al*., 2012). Most of the changes occur in the horizontal dimension reaching up to 50% during the first year following extraction in premolars (Schropp *et al*., 2003). Vertically, bone crest resorption is more pronounced on the buccal aspect (Nevins *et al*., 2006) and more pronounced in the mandible than in the maxilla (Smukler *et al*., 1999).

These events are progressive and irreversible, resulting in major prosthetic, aesthetic and functional challenges during the rehabilitation. The alveolar ridge preservation (ARP) technique has been proposed to reduce the dimensional changes after tooth extraction (MacBeth *et al*., 2017). Current literature has shown that although ARP does not completely prevent bone resorption, the procedure seems to be effective in preserving the volume of the alveolar ridge compared with spontaneous healing (Araújo *et al*., 2015a; Barone *et al*., 2015).

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The latest ARP techniques recommend a combination of soft and hard tissue preservation with a long-term healing period (4-6 months), with a flapless approach. These techniques are known as socket sealing and combine the use of biomaterials placed at the intact bone level and autogenous grafts or soft tissue substitutes at the level of the remaining gingival margin (Jung *et al*., 2013; Meloni *et al*., 2015).

Soft tissue substitutes have been successfully used to seal the socket for ARP, simplify the procedure and reduce postoperative morbidity compared to autogenous grafts (Jung *et al*., 2013; Meloni *et al*., 2015). Collagen matrices (CM), derived from the porcine dermis, consisting of native type I and III collagen fibers plus elastin have been used as a tissue substitute (Barbeck *et al*., 2015).

Considered the gold standard for bone grafting, autogenous bone has morbidity and potential complications associated with the donor site and may not have the desired stability for ridge preservation which provides a reason to consider alternatives. Of all these biomaterials, demineralized bovine bone graft (DBBM) was potentially the most used material for ARP in recent years (Araújo *et al*., 2015b).

In flapless tooth extractions, the periosteum remains in close contact with the cortical bone and thus preserves the blood supply to the underlying cortical bone, leading to a reduction in alveolar bone loss compared to total flap extractions (Engler-Hamm *et al*., 2011; Araújo *et al*., 2015a; Barone *et al*., 2015). A clinical study (Barone *et al*., 2015) showed that the flapless technique can preserve the horizontal dimension of the alveolar bone and increase the keratinized gingiva more successfully than the total flap technique. Furthermore, the flapless approach is known to be a simple, atraumatic and conservative method. Several authors have reported better clinical results with the flapless approach with reduced healing times, discomfort and inflammation (Engler-Hamm *et al*., 2011; Araújo *et al*., 2015b; Barone *et al*., 2015). Total flap elevation was associated with postoperative bone resorption mainly in the initial healing phase, 4 to 8 weeks after tooth extraction (Fickl *et al*., 2008). In addition, marginal recession in adjacent teeth, papilla defects, and loss of keratinized mucosa may be the result of flap manipulation to obtain partial or complete coverage of a post-extraction socket (Landsberg, 1997).

The objective of this study was to evaluate, clinically and tomographically, the dimensional changes of the socket with the ARP technique using DBBM associated with a CM in atraumatic extraction sockets in two clinical situations, flapless and flapped technique. This is the first controlled study aimed at evaluating the clinical performance of mucoderm® left exposed for soft tissue gain in the flapless ARP technique. Single-rooted teeth with clinical and radiographic indications for extraction were selected.

Materials and methods

Design of study

This is a clinical randomized case series carried out at the University of São Paulo in Ribeirão Preto, Brazil, between January 2019 and November 2021. The surgical procedures were performed by a single experienced surgeon (MMS) and were approved by the institutional ethics committee. All selected subjects received detailed information about the study and gave their written consent.

Patient selection

Inclusion criteria

Patients of both sexes who presented:

1. Age: >18 years;
2. Need for extraction of premolars, confirmed by clinical and radiographic examinations;
3. Presence of a tooth adjacent to the extraction site;
4. Systemically healthy, with no contraindication for oral surgical procedures;
5. Socket with 3 walls and 80% of the 4th wall intact (Engler-Hamm *et al*., 2011);
6. Sign the informed consent form.

Exclusion criteria

Patients who had systemic involvement capable of interfering with the periodontium or with the response to treatment, such as diabetes and heart disease; metabolic bone disease; use of drugs that interfere with bone metabolism; prolonged use of anti-inflammatory drugs or corticosteroids; use of bisphosphonates in the last 4 years; known allergy to any biomaterial to be used in the study; smoking; inflammation and/or infection in the tooth that would be extracted; pregnant/lactating women or those who developed this systemic condition during the study; and patients who refused to sign the consent form.

Biomaterials

DBBM (cerabone®, botiss biomaterials, Germany) with 65-80% porosity and 0.5-1.0mm diameter granules containing the synthesized inorganic part of bovine bone (hydroxyapatite 100% pure). The biomaterial was hydrated in sterile saline solution before filling the socket, following the manufacturer's instructions.

CM (mucoderm®, botiss biomaterials, Germany) of native collagen type I and III non cross-linked, with a thickness of 1.2-1.7mm was used as a space maker aiming to conduct the formation of keratinized mucosa and to protect the wound at initial stages. The CM was rehydrated for 20 minutes in sterile saline.

Interventions

T0 (baseline)

Prior to ARP, study participants received oral hygiene instructions, patient-professional plaque control, supra and subgingival scaling and root planing procedures with Gracey periodontal curettes (Hu-friedy®) and ultrasound device, in order to control and eliminate areas of periodontal infections and acute inflammatory processes.

One week before the ARP, an impression was taken to make the radiographic guide in a rigid crystal plate (PET-G 1.0mm thick) with a solid aluminum spherical marker (3.175mm in diameter) inserted in the guide in the center of the tooth that would be extracted and which served as a reference for superimpose cone beam computed tomography (CBCT) and clinical soft tissue assessments.

Surgical procedures were performed after indexes of plaque and bleeding on probing were below 20%.

Patients were randomly distributed between G1, with careful intrasulcular incision made with a 15C surgical blade and flapless technique and G2 with intrasulcular and oblique incisions on the distal surface of the adjacent tooth to be extracted, made with a 15C surgical blade with a total flap.

The atraumatic extractions were performed under local anesthesia of mepivacaine hydrochloride + epinephrine at 36mg+18μg/1.8ml (DFL, Rio de Janeiro, Brazil) with the aid of a periotome and elevators to preserve the buccal cortical bone and the soft and hard tissues surroundings.

In G1, the soft tissue edges of the socket were de-epithelialized using a diamond bur (Figure 1B) and in G2 the flap was raised at a minimum exposure and limited to the visualization of the buccal bone crest, in addition to allowing adequate sealing of the socket by the CM (Figure 1G). After careful curettage and removal of all granulation tissue, the socket was filled with DBBM to the remaining bone level (Figure 1C and H) and sealed with CM positioned slightly below the marginal mucosa in order to overlap the margins of the socket for 2-3 mm (Figure 1D and I). Simple sutures were performed to stabilize the CM in G1 and in G2 whose base of the total flap was not divided, the flap was repositioned and sutured in the original position of the gingival margin (Figure 1E and J). In both groups the CM was left exposed. The sutures were made with 5-0 nylon thread and removed after 15 days.

All patients received systemic antibiotic therapy (875mg of amoxicillin + clavulanic acid 125mg) initiated 24 hours before the surgical procedure, with extension for another 10 days, in a regimen of 12 in 12 hours. An analgesic (750mg paracetamol) was also prescribed, every 8 hours while there was pain until completing a period of 5 days. Patients were instructed to rinse their mouths twice daily with 0.12% chlorhexidine digluconate solution for 15 days and to discontinue toothbrushing in the surgical area during this period. Considering that the CM was left exposed, a protocol was implemented with local applications of 1% chlorhexidine gel and weekly oral hygiene appointments (Slot *et al*., 2010).

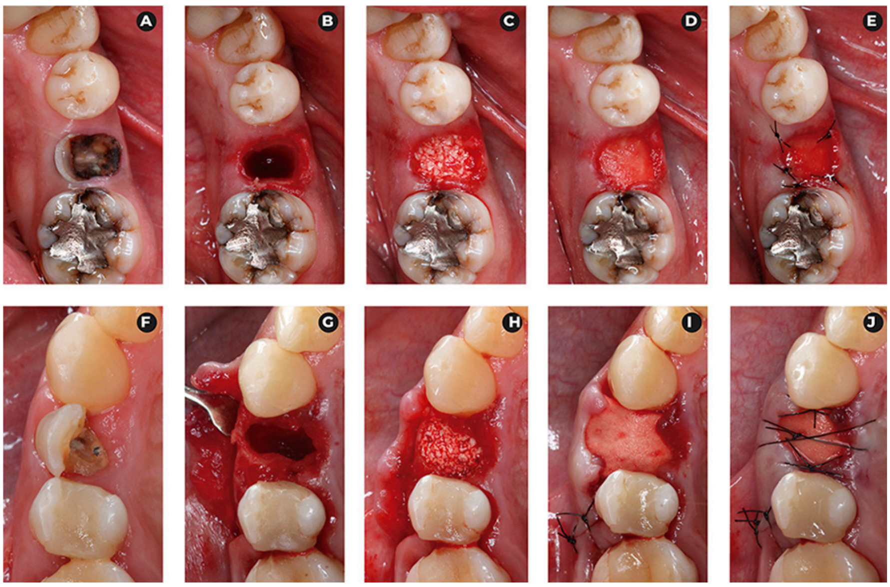

Figure 1. Surgical procedure of G1: Clinical situation before extraction of the residual root of tooth 45 (A). Flapless tooth extraction socket, the soft tissue edges of the socket were de-epithelialized using a diamond bur (B). Socket filled with cerabone® (C). mucoderm® in position, 2-3mm below the gingival margin (D) and single sutures (E); Surgical procedure of G2: Clinical situation before extraction of the residual root of tooth 14 (F). Total flap incisions and visualization of the post-extraction socket (G). Socket filled with cerabone® (H). mucoderm® in position, 2-3mm below the gingival margin (I) and sutures (J).

Follow-up

Immediately after ARP, patients received a Visual Analog Scale diary (VAS 100mm) ranging from 0 (no pain and discomfort) to 100 (worst pain and discomfort imaginable) to be filled in at home. Patients were asked to complete the VAS on the day of surgery every 4 hours and then daily (once a day) until day 7. VAS diaries were collected at the 1 week follow-up visit and a new VAS was collected on this day and at 24-month follow-up. Photographs were taken at the 1 week, 2 weeks, 3 weeks, 4 month and 24 months follow-ups (Figure 2).

The CBCTs were performed by the same operator blinded to the study, on the same tomograph (Eagle X 3D, Dabi Atlante, Ribeirão Preto, Brazil), with the same settings (0.19mm resolution; FOV 6x8; 90KV; 6.3mA) at 1 week (T1), 4 months (T2) and 24 months (T3) after ARP using the custom-made tomographic guide. All examiners blinded to the study were previously calibrated, measurements were performed in duplicate for 2 experienced examiners. Intra- and inter-examiner agreement (kappa) were higher than >0.8.

Linear Measures

The DICOM (Digital Imaging and Communications in Medicine) data were imported into the main database of the OnDemand3D software (Version 1.0.9.3223, CyberMed Inc) and the "Fusion" tool was used to superimpose the CBCTs through the mathematical algorithm method that calculates the best fit and automatically overlays the two volumetric images with voxel-based information. CBCT T1 was chosen for the primary image and CBCT T2 and T3 as secondary images superimposed one by one on top of T1. If the automatic superimposition obtained was not a perfect fit, the secondary image was manually aligned to the primary image using the manual registration tool and the aluminum sphere of custom-made tomographic guide. Finally, to ensure that the axis position corresponded in the two volumetric images, the "Reslicing" function was used. This tool allows the secondary image to be resized based on the primary image so the measurements are performed on the same slice and in the same position (Figure 3). The superimposition between the 2 images was presented as color images to identify areas of bone resorption and dimensional changes (Figure 4).

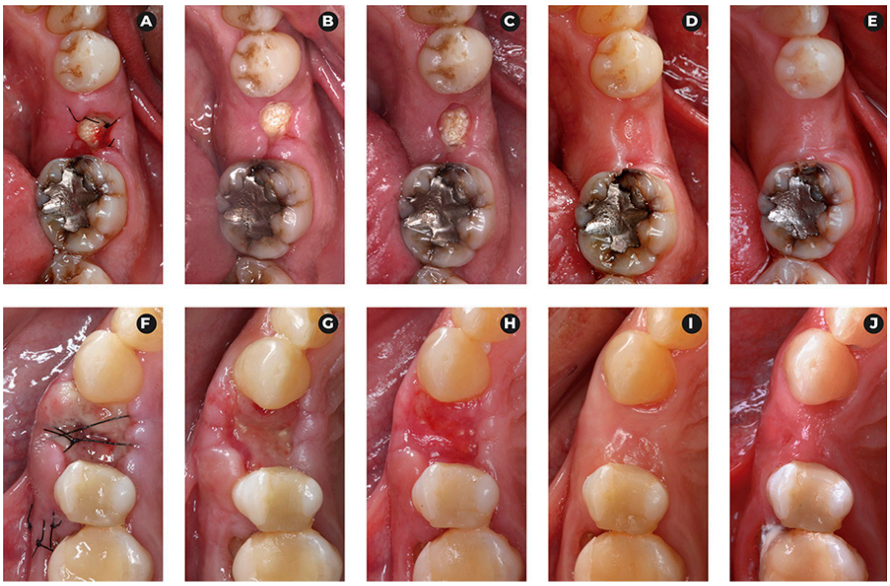

Figure 2. Postoperative follow-up of G1: 1 week (A); 2 weeks (B); 3 weeks (C); 4 months (D) and 24 months (E). Postoperative follow-up of G2: 1 week (F); 2 weeks (G); 3 weeks (H); 4 months (I) and 24 months (J).

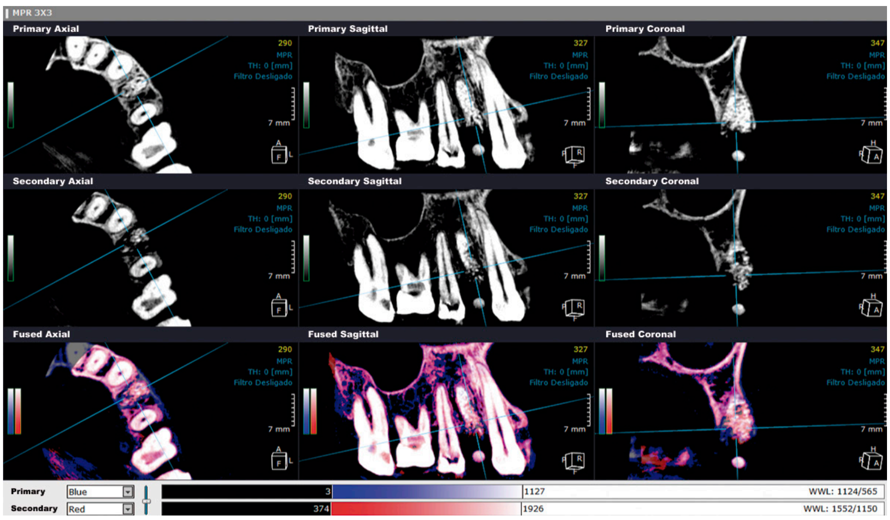

Figure 3. CBCT T1 (blue) and T2 (yellow) superimposed in the OnDemand3D software.

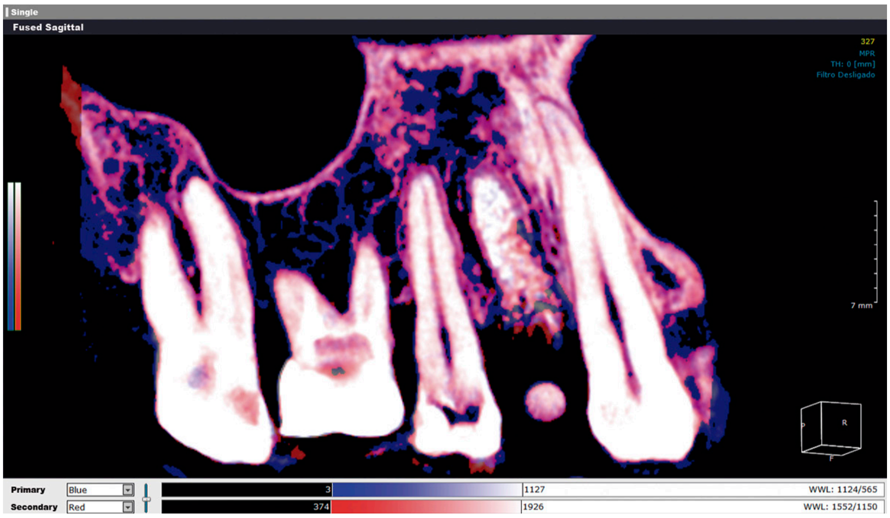

Figure 4. Dimensional changes between T1 (blue) and T2 (yellow) in the sagittal plane.

The T1 and resized T2 and T3 CBCTs were exported and converted to extension (.gipl) by the ITK-SNAP software (version 3.6.0 www.itksnap.org) for correct superimpose in the Slicer software (version 5.0.2 www.slicer.org) used to evaluate measurements by 2 examiners.

Linear measurements were similar to those described by Jung (2013). All measurements were obtained with the following reference lines: a vertical line drawn in the center of the extraction socket and crossing its most apical point, and a horizontal line perpendicular to the extraction socket vertical line and crossing the most apical point of the socket (Figure 5).

The following parameters were evaluated:

1. The buccal (BH) and lingual/palatal (LH) cortical bone height;

2. The horizontal width of the extraction socket measured at 1 mm, 3 mm, and 5 mm below the lingual bone crest (HW-1, HW-3, HW-5);

Clinical analysis

Clinical analyzes were performed by the same examiner blinded to the study at the 3 assessment times.

To assess the height of the keratinized gingiva (WKG) and the thickness of the keratinized gingiva (TKG), the center of the aluminum sphere with the

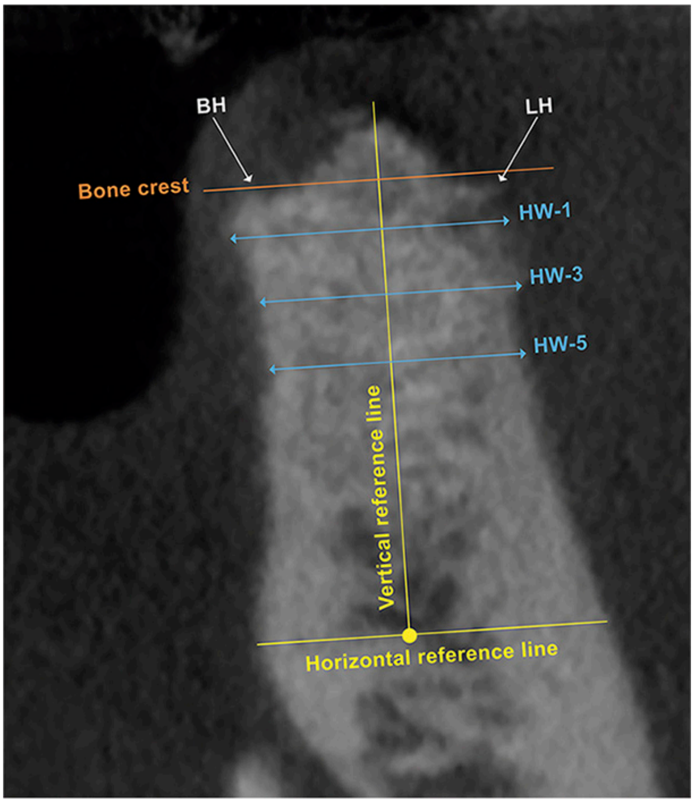

Figure 5. Vertical and horizontal references lines; BH (buccal height); LH (lingual/palatal height); HW (horizontal width) at 1, 3 and 5mm below the bone crest; ST (soft tissue) at 1, 3 and 5mm below the bone crest.

apical margin of the tomographic guide (MTG) was used as a vertical reference. WKG was measured by the distance between the MTG to the mucogingival junction on the reference line, with the aid of a dry tip caliper and recorded by a caliper. The TKG was performed at 1mm from the JMG, at the reference line, using a cursor adapted to a 30G gingival needle that penetrated the keratinized mucosa until it found a hard surface. The measurement obtained was transferred to a caliper. In order for the subsequent TKG assessment to take place at the same location, the distance between a predetermined fixed point on the tomographic guide and the location of this measurement was recorded.

Statistical analysis

Data were grouped and presented as means and standard deviations, were tested for normality and even with normality present, we adopted the Mann-Whitney U-test due to the sample size. The Mann-Whitney U-test was generated using the Real Statistics Resource Pack software (version 7.6 Charles Zaiontz) to identify possible differences between groups. Results with $P<0.05$ were considered statistically significant in this study.

Results

Nine patients underwent surgery phase. Due to the COVID-19 pandemic 1 patient did not undergo the T2 CBCT in the correct period and their data were excluded from this study. There were no cases of infection or necrosis, but one patient reported having removed a fragment of biomaterial from the surgical wound and her data were also excluded. Seven patients attended all appointments, 4 women and 3 men with mean±SD of 51.86±11.49 years (Table 1).

In the intragroup comparison, no statistically significant differences were observed between times T1, T2 and T3 for the means of the evaluated parameters (Table 2).

The postoperative VAS diary had a statistically significant difference (P=0.03) indicating less pain and discomfort for G1 (5.53±9.68) compared to G2 (15.31±21.08) (Table 3). Although there was no statistically significant difference for the other periods evaluated in the VAS, G1 also presented less pain and discomfort in the 1 week (2.33±0.58) and 24-month periods (1.67±1.53) compared to G2. As expected, the VAS scale decreased over time in both groups.

The mean dimensional changes between T1, T2 and T3 are shown in Table 4. Both groups showed reductions in most parameters, but G1 had smaller losses than G2 in the tomographic parameters BH, LH, HW-3, HW -5 in both T2 as in T3. On the other hand, G2 was superior to G1 with gain in HW-1 in T2 and T3. Despite the slight decrease in radiographic parameters between all times evaluated, there were no statistically significant differences. In clinical parameters, there was an increase in TKG G1 in T2 +0.43±0.47mm (+41.7%) and in T3 +0.55±0.66mm (+53.4%) while in G2 there was a reduction of -0.11±0.41mm (-7.1%) and increase of +0.06±0.56mm (+3.9%) respectively, these differences were not statistically significant. In WKG at T2 both groups had reductions of -0.02±1.19mm (-0.8%) in G1 and -0.11±0.41mm (-7.1%) in G2. In T3, G1 increased +0.29±1.23mm (+11.5%) and G2 reduced -1.81±3.48mm (-31.9%). There were also no statistical differences between the groups at the different evaluation times. However, G1 seems to have more favorable results in the WKG and TKG compared to G2 in T2 and T3 changes.

Table 1. Demographic data of patients and sockets included.

Patients parameters	Group 1	Group 2
Age (years)	44.67 ± 7.02	57.25 ± 11.87
Men/women	2/1	1/3
Mandible/Maxilla	3/0	2/2
Thin/thick biotype	2/1	4/0
Reasons for extraction fracture/loss of tooth substance/ prosthetic motif	0/2/1	1/3/0

Values are presented as mean±standard deviation.

Table 2. Intragroup comparisons at 1 week, 4 months and 24 months after ARP.

Parameter Group 1	1 week	4 months	P-value	24 months	P-value
BH (mm)	12.45±1.27	12.11±1.41	0.700	11.89±1.36	0.700
LH (mm)	12.73±0.91	11.81±0.63	0.400	11.77±0.28	0.200
HW-1 (mm)	8.09±0.79	6.58±1.35	0.400	6.09±1.08	0.100
HW-3 (mm)	9.41±1.81	8.74±1.95	0.700	8.70±1.61	0.700
HW-5 (mm)	10.40±3.23	10.16±2.81	0.700	10.15±2.69	0.700
WKG (mm)	2.52±0.70	2.50±0.72	1.00	2.81±0.80	0.700
TKG (mm)	1.03±0.64	1.46±0.30	0.700	1.58±0.42	0.400
Parameter Group 2	**1 week**	**4 months**	**P-value**	**24 months**	**P-value**
BH (mm)	8.04 ± 2.60	7.47 ± 2.50	0.885	7.30 ± 2.55	0.685
LH (mm)	8.82 ± 2.12	7.83 ± 2.50	0.485	7.75 ± 2.38	0.485
HW-1 (mm)	5.89 ± 2.37	6.35 ± 1.37	0.885	6.17 ± 1.45	0.885
HW-3 (mm)	8.83 ± 1.82	7.41 ± 1.42	0.342	7.42 ± 1.34	0.342
HW-5 (mm)	9.81 ± 2.60	9.20 ± 2.72	0.685	9.31 ± 2.64	0.685
WKG (mm)	5.68 ± 2.00	4.76 ± 1.72	0.485	3.87 ± 2.54	0.342
TKG (mm)	1.55 ± 0.55	1.44 ± 0.45	0.885	1.61 ± 0.35	0.685

Values are presented as mean±standard deviation.
BH, buccal cortical height; LH, lingual cortical height; HW, horizontal width at 1mm, 3mm and 5mm below the most coronal aspect of the bone crest; WKG, width of keratinized gingiva; TKG, thickness of keratinized gingiva.

Table 3. Intergroup comparisons of postoperative VAS.

	Group 1	Group 2	P-value
Diary (mm)	5.53 ± 9.68	15.31 ± 21.08	0.038[a)]
1 week (mm)	2.33 ± 0.58	6.5 ± 9.04	0.857
24 months (mm)	1.67 ± 1.53	3.25 ± 2.63	0.628

Values are presented as mean±standard deviation.
Diary, VAS of the first 7 postoperative days; 1 week, VAS at 1 week; 24 months, VAS at 24 months
[a)] Statistically significant difference between the groups.

Table 4. Changes between immediate, 4 months and 24 months after ARP.

		1 week – 4 months			
	Group 1		Group 2		
Parameters	**mm**	**%**	**mm**	**%**	**P-value**
BH (mm)	-0.33 ± 0.15	-2.7	-0.58 ± 0.83	-7.2	0.400
LH (mm)	-0.92 ± 0.29	-7.2	-0.99 ± 0.73	-11.2	0.857
HW-1 (mm)	-1.51 ± 0.58	-18.7	+0.46 ± 1.59	+7.8	0.114
HW-3 (mm)	-0.67 ± 0.15	-7.1	-1.42 ± 0.69	-16.1	0.057
HW-5 (mm)	-0.24 ± 0.54	-2.3	-0.62 ± 0.20	-6.3	0.628
WKG (mm)	-0.02 ± 1.19	-0.8	-0.92 ± 2.48	-16.2	0.857
TKG (mm)	+0.43 ± 0.47	41.7	-0.11 ± 0.41	-7.1	0.228
		1 week – 24 months			
	Group 1		**Group 2**		
Parameters	**mm**	**%**	**mm**	**%**	**P-value**
BH (mm)	-0.56 ± 0.14	-4.5	-0.74 ± 0.65	-9.2	0.400
LH (mm)	-0.96 ± 0.75	-7.5	-1.07 ± 0.63	-12.1	0.857
HW-1 (mm)	-2.00 ± 0.35	-24.7	+0.28 ± 1.45	+4.8	0.057
HW-3 (mm)	-0.71 ± 0.34	-7.5%	-1.40 ± 0.78	-15.9	0.114
HW-5 (mm)	-0.25 ± 0.70	-2.4	-0.50 ± 0.31	-5.1	0.628
WKG (mm)	+0.29 ± 1.23	+11.5	-1.81 ± 3.48	-31.9	0.400
TKG (mm)	+0.55 ± 0.66	+53.4	+0.06 ± 0.56	+3.9	0.400

Values are presented as mean±standard deviation.
- means reduce; + means increase. BH, buccal cortical height; LH, lingual cortical height; HW, horizontal width at 1mm, 3mm and 5mm below the most coronal aspect of the bone crest; WKG, width of keratinized gingiva; TKG, thickness of keratinized gingiva.

Discussion

To our knowledge, this was the first study to evaluate the dimensional changes after ARP using DBBM and CM left intentionally exposed with flapless and flapped approach evaluated at 4 and 24 months after ARP. Comparisons with our study will be generalized according to the available evidence. Therefore, the randomized controlled clinical trials mentioned in this section refer to CM Mucograft® or Mucograft® Seal (Geistlich Pharma AG, Wolhusen, Switzerland).

Regarding biomechanical properties, mucoderm® (MD) should be used after rehydrating while Mucograft® (MG) is used dry. In the dry condition MD showed significantly increased tensile strength (P≤0.036) compared to MG (Kasaj *et al*., 2016). In rehydration in sterile saline, MG showed significantly lower resistance compared to MD (P≤0.028) (Kasaj *et al*., 2016). As for the modulus of elasticity, the highest was found in the MD matrix at 20min of rehydration in sterile saline solution, while MG showed the lowest modulus of elasticity after 10min (P≤0.024) (Kasaj *et al*., 2016). As a stable 3D matrix, MD allows the colonization of fibroblasts which acts as a selective guide for keratinocytes and has a protective function for connective tissue (Barbeck *et al*., 2015).

In this study, both G1 and G2 were effective in limiting the vertical and horizontal dimensional changes of the socket in hard and soft tissue. These results are consistent with the literature that show limited dimensional changes by ARP using DBBM and CM compared to spontaneous healing (Jung *et al*., 2013; Parashis *et al*., 2016; Natto *et al*., 2017; Llanos *et al*., 2019; Clementini *et al*., 2019). Furthermore, recently published systematic reviews concluded that even if ARP procedures are performed, it is unlikely that the dimensions of the alveolar ridge will be completely preserved after extraction (Tan *et al*., 2012; MacBeth *et al*., 2017; Bassir *et al*., 2018; Canullo *et al*., 2022).

In the horizontal alterations of the socket, this study found that the HW at 1mm (T2 G1: -1.51±0.58mm;-18.7%; G2: +0.46±1.59mm;+7.8% and T3 G1: -2.00±0.35mm;-24.7%; G2: 0.28±1.45mm;+4.8%), 3mm (T2 G1: -0.67±0.15mm;-7.1%; G2: -1.42±0.69mm;-11.2% and T3 G1: -0.71±0.34mm;-7.5%; G2: -1.40±0.78mm;-15.9%) and 5mm (T2 G1: -0.24±0.54mm;-2.3%; G2: -0.62±0.20mm;-6.3% and T3 G1:-0.25±0.70mm;-2.4%; G2: -0.50±0.31mm;-5.1%) below the marginal bone crest had limited bone resorption when compared to spontaneous healing in other studies and systematic reviews (Jung *et al*., 2013; Natto *et al*., 2017; Majzoub *et al*., 2019; López-Pacheco *et al*., 2021). A recent study (Clementini *et al*., 2019) and a systematic review (Tan *et al*., 2012) revealed that the horizontal alterations means of the socket in spontaneous healing were -3.37±1.55mm; (43.2±25.1%) and 3.79±0.23mm (29%–63%) respectively, which shows greater resorption and dimensional changes of the socket compared to our results.

A study using the flapless technique concluded that the mean HW at 1mm from the bone crest had a reduction of -1.37±0.84mm, at 3mm a reduction of -0.84±0.62mm and at 5mm a reduction of -0.56±0.48mm after 4 months of healing (Llanos *et al*., 2019). These changes compared to the HW of T2 in the present study were slightly reduced at 1mm (G1: -1.51±0.58mm) and slightly greater at 3mm (G1: -0.67±0.15mm) and 5mm (G1: -0.24±0.54mm). Such differences may be associated with the size and composition of the sample since Llanos (2019) evaluated 65 patients with extraction of maxillary canines and incisors and our study evaluated 3 patients with extraction of mandibular premolars.

As for the flapped technique, a study observed a horizontal reduction of the socket at 1mm of -1.56±0.71mm, at 3mm of -1.07±0.69mm and at 5mm of -0.96±0.61mm after 4 months of healing (Clementini *et al*., 2019). Compared to the changes in HW of T2 in G2, they were slightly reduced at 5mm (G2: -0.62±0.20mm; -6.3%) and slightly greater at 3mm (G2: -1.42±0.69mm; -11.2%). The size and composition of the sample may also have influenced these results, Clementini (2019) treated 30 individuals with extraction of anterior teeth and premolars from both arches and G2 treated 4 patients with extraction of premolars from both arches. As for our result of an increase in HW-1mm (G2: +0.46±1.59mm; +7.8%), when superimposing the T1 and T2 CBCTs we observed an expansion of the socket in 2 patients in the T3, probably due to the limited vestibular cortical thickness of 0.3mm in both cases and its consequent more pronounced reabsorption with socket expansion influencing HW measurements. Another characteristic that differentiates these 2 patients from the rest is that the LH was greater than the BH. However, despite the increased volume in hard tissue, the soft tissue showed a reduction in thickness.

Vertical bone changes were also limited in BH (T2 G1: -0.33±0.15mm;-2.7%; G2: -0.58±0.83mm;-7.2% and T3 G1: -0.56±0.14mm;-4.5%; G2: -0.74±0.65mm;-9.2%) and in LH (T2 G1: -0.92±0.29mm;-7.2%; G2 : -0.99±0.73mm;-11.2% and T3 G1: -0.96±0.75mm;-7.5% G2: -1.07±0.63mm;-12.1%) compared to the results of spontaneous healing where BH resorption was -1.94±1.26mm and LH was -1.60±2.05mm (Jung *et al*., 2018). This agrees with other studies, where ARP resulted in less vertical resorption 4-6 months after tooth extraction when compared to spontaneous healing (Jung *et al*., 2013; Araújo *et al*., 2015a; Natto *et al*., 2017; Jonker *et al*., 2021).

In a recent study, vertical bone resorption in the flapless technique showed similar results to G1, where the BH suffered a reduction of -0.30±1.09mm (G1: -0.33±0.15mm; -2.7%) but for LH the reduction was a slight less -0.27±2.30mm compared to G1 -0.92±0.29mm; -7.2% (Natto *et al*., 2017). On the other hand, Llanos (2019) reported a reduction in BH of -6.95±3.82 and LH of -1.03±0.95mm, different from the findings of the present study, this is probably due to the teeth that were extracted in each study and the sample sizes. While Llanos (2019) included 65 individuals with extraction only of maxillary canines and incisors with the presence of up to 50% of the buccal bone, G1 included 3 mandibular premolars with the presence of up to 80% of the buccal bone.

In the surgical technique with flap, other studies had reductions in dimensions similar to G2, in which BH had resorption of -0.31±0.33mm (Lim *et al*., 2017; Clementini *et al*., 2019) and LH -0.82±0.61mm (Nart *et al*., 2017) while G2 had -0.58±0.83mm; -7.2% and -0.99±0.73mm; -11.2% respectively. The small differences found between these studies and our results can be explained by the size and composition of the samples.

The position of the extracted tooth also influences the dimensional differences. Januario (2011), showed that approximately 50% of the coronal portion of the buccal cortical bone in maxillary incisors and canines has a mean thickness of 0.6 mm. This explains the discrepancy regarding the amount of resorption between the results of this study (BH G1: -0.33mm (-2.7%), G2: -0.58mm (-7.2%); LH G1: -0.92mm (-7.2%), G2: -0.99mm (-11.2%)) and the results of Araújo (2015a) (BH -3.6mm (35.8%) and LH -1.4mm (13.4%)). While our study evaluated premolars, their study was composed mainly of anterior teeth that had greater buccal bone plate resorption. It can be explained by alveolar bone that has high rates of resorption, while regions with greater thickness are composed of alveolar and lamellar bone with lower rates of resorption (Araujo *et al*., 2005; Discepoli *et al*., 2013;).

Clinical analysis of soft tissues showed an increase in the TKG of G1 (+0.43±0.47; +41.7%) very similar to the results of a recent study that had a gain of +0.47±1.16mm in the experimental group that used DBBM and CM in the flapless technique and 4 months of healing (Natto *et al*., 2017). Although this increase in TKG occurred in G1, it was not statistically significant when compared to G2. The results of G2 (-0.11±0.41mm; -7.1%) were very similar to those found by Hong (2019) −0.15±0.23mm in a flap approach and 6 months of healing.

The reduction in WKG in G1 (-0.02±1.19mm; -0.8%) was also similar to this same study, where the DBBM and CM experimental group in the flapless surgical technique had a WKG reduction of -0.08±0.54mm (Natto *et al*., 2017) at 4 months of healing. G2, on the other hand, showed a reduction in WKG of -0.92±2.48mm; -16.2%, which was slightly lower when compared to the findings of Hong (2019) −1.57±0.51mm. Again, the different sample sizes and composition may explain these small differences.

The differences between G1 and G2 in the dimensional changes of soft tissues may be due to the different surgical techniques and manipulation of these tissues, as verified by several studies (Engler-Hamm *et al*., 2011; Fickl *et al*., 2017; Avila-Ortiz *et al*., 2019; Hong *et al*., 2019). One study also reported that total flap elevation and healing by first intention resulted in reduced soft tissue dimensions when compared to flapless surgical technique and healing by second intention (Hong *et al*., 2019).

VAS was used to compare postoperative pain and discomfort at different times: first 7 days (daily), 1 week and 24 months. There was a statistically significant difference only in the VAS diary, which showed that G1 resulted in less postoperative pain and discomfort. This is in agreement with a recent systematic review with meta-analysis that identified significantly lower VAS pain scores in flapless ARP and no heterogeneity was identified (Atieh *et al*., 2022). Other authors who performed ARP and compared pain and discomfort in flapless and total flap surgical techniques also identified significant differences for the flapless groups (Engler-Hamm *et al*., 2011; Aladmawy *et al*., 2019).

According to a systematic review with meta-analysis, flapless ARP promoted smaller reduction in the horizontal width of the socket when compared to the flapped, this difference was not statistically significant (Atieh *et al*., 2022). Furthermore, the vertical changes were very similar with no significant differences between the surgical techniques (Atieh *et al*., 2022). Changes in WKG and TKG favor the use of flapless ARP technique (Atieh *et al*., 2022).

The biomaterials used in this study were intentionally exposed. No signs of infection or necrosis of the DBBM or the CM was observed and several recent studies also have shown that biomaterial exposure does not appear to adversely affect ARP outcomes (Jung *et al*., 2013; Parashis *et al*., 2016; Natto *et al*., 2017).

Even with interesting results, some limitations must be addressed. First, the reduced sample size for each group may not represent results that reflect reality or be immediately extrapolated. Second, maxillary and mandibular teeth were included in this study and there are differences in alveolar bone anatomy, as discussed above, that may influence ARP results. In addition, the only two maxillary teeth were randomized to G2. Third, the non-inclusion of a spontaneous healing control group.

The present study demonstrated that ARP limited vertical and horizontal socket changes, regardless of the surgical technique used and the exposure of biomaterials. The flapless approach seems to provide better results regarding dimensional changes and significantly less discomfort.

Acknowledgements and conflict of interest

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References

Aladmawy MA, Natto ZS, Steffensen B et al. A Comparison between Primary and Secondary Flap Coverage in Ridge Preservation Procedures: A Pilot Randomized Controlled Clinical Trial. *BioMed Research International* 2019; **2019**:1-7.

Araújo MG, da Silva JCC, de Mendonça AF and Lindhe J. Ridge alterations following grafting of fresh extraction sockets in man. A randomized clinical trial. *Clinical Oral Implants Research* 2015a; **26**:407–412.

Araújo MG, Silva CO, Misawa M and Sukekava F. Alveolar socket healing: what can we learn? *Periodontology 2000* 2015b; **68**:122–134.

Araujo MG and Lindhe J. Dimensional ridge alterations following tooth extraction. An experimental study in the dog. *Journal of Clinical Periodontology* 2005; **32**:212–8.

Araújo MG and Lindhe J. Ridge alterations following tooth extraction with and without flap elevation: an experimental study in the dog. *Clinical Oral Implants Research* 2009; **20**:545–9.

Atieh MA, Alfardan L and Alsabeeha NHM. Flapped versus flapless alveolar ridge preservation: a systematic review and meta-analysis. *International Journal of Oral and Maxillofacial Surgery* 2022; **51**:133–142.

Avila-Ortiz G, Chambrone L and Vignoletti F. Effect of alveolar ridge preservation interventions following tooth extraction: A systematic review and meta-analysis. *Journal of Clinical Periodontology* 2019; **46**:195–223.

Barbeck M, Lorenz J, Kubesch A et al. Porcine Dermis-Derived Collagen Membranes Induce Implantation Bed Vascularization Via Multinucleated Giant Cells: A Physiological Reaction? *Journal of Oral Implantology* 2015; **41**:e238–e251.

Barone A, Borgia V, Covani U, Ricci M, Piattelli A and Iezzi G. Flap versus flapless procedure for ridge preservation in alveolar extraction sockets: a histological evaluation in a randomized clinical trial. *Clinical Oral Implants Research* 2015; **26**:806–13.

Bassir S, Alhareky M, Wangsrimongkol B, Jia Y and Karimbux N. Systematic Review and Meta-Analysis of Hard Tissue Outcomes of Alveolar Ridge Preservation. *The International Journal of Oral & Maxillofacial Implants* 2018; **33**:979–994.

Canullo L, Pesce P, Antonacci D et al. Soft tissue dimensional changes after alveolar ridge preservation using different sealing materials: a systematic review and network meta-analysis. Clinical Oral Investigations 2022; 26:13–39.

Clementini M, Agostinelli A, Castelluzzo W, Cugnata F, Vignoletti F and de Sanctis M. The effect of immediate implant placement on alveolar ridge preservation compared to spontaneous healing after tooth extraction: Radiographic results of a randomized controlled clinical trial. *Journal of Clinical Periodontology* 2019; **46**:776–786.

Discepoli N, Vignoletti F, Laino L, de Sanctis M, Muñoz F and Sanz M. Early healing of the alveolar process after tooth extraction: an experimental study in the beagle dog. *Journal of Clinical Periodontology* 2013; **40**:638–644.

Engler-Hamm D, Cheung WS, Yen A, Stark PC and 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**:377–387.

Fickl S, Zuhr O, Wachtel H, Stappert CFJ, Stein JM and Hürzeler MB. Dimensional changes of the alveolar ridge contour after different socket preservation techniques. *Journal of Clinical Periodontology* 2008; **35**:906–913.

Fickl S, Fischer K, Petersen N et al. Dimensional Evaluation of Different Ridge Preservation Techniques: A Randomized Clinical Study. *The International Journal of Periodontics & Restorative Dentistry* 2017; **37**:403–410.

Hämmerle CHF, Araújo MG and Simion M. Evidence-based knowledge on the biology and treatment of extraction sockets. *Clinical Oral Implants Research* 2012; **23**:80–82.

Hong HR, Chen CY, Kim DM and Machtei EE. Ridge preservation procedures revisited: A randomized controlled trial to evaluate dimensional changes with two different surgical protocols. *Journal of Periodontology* 2019; **90**:331–338.

Januario AL, Duarte WR, Barriviera M, Mesti JC, Araújo MG and Lindhe J. Dimension of the facial bone wall in the anterior maxilla: a cone-beam computed tomography study. *Clinical Oral Implants Research* 2011; **22**:1168–1171.

Jonker BP, Gil A, Naenni N, Jung RE, Wolvius EB and Pijpe J. Soft tissue contour and radiographic evaluation of ridge preservation in early implant placement: A randomized controlled clinical trial. *Clinical Oral Implants Research* 2021; **32**:123–133.

Jung RE, Philipp A, Annen BM et al. Radiographic evaluation of different techniques for ridge preservation after tooth extraction: a randomized controlled clinical trial. *Journal of Clinical Periodontology* 2013; **40**:90–98.

Jung RE, Sapata VM, Hämmerle CHF, Wu H, Hu XL and Lin Y. Combined use of xenogeneic bone substitute material covered with a native bilayer collagen membrane for alveolar ridge preservation: A randomized controlled clinical trial. *Clinical Oral Implants Research* 2018; **29**:522-529.

Kasaj A, Levin L, Stratul SI et al. The influence of various rehydration protocols on biomechanical properties of different acellular tissue matrices. *Clinical Oral Investigations* 2016; **20**:1303–1315.

Landsberg CJ. Socket seal surgery combined with immediate implant placement: a novel approach for single-tooth replacement. *The International Journal of Periodontics & Restorative Dentistry* 1997; **17**:140–9.

Lim HC, Jung UW, You H and Lee JS. Randomized clinical trial of ridge preservation using porcine bone/cross-linked collagen vs. bovine bone/non-cross-linked collagen: cone beam computed tomographic analysis. *Clinical Oral Implants Research* 2017; **28**:1492–1500.

Llanos AH, Sapata VM, Jung RE et al. Comparison between two bone substitutes for alveolar ridge preservation after tooth extraction: Cone-beam computed tomography results of a non-inferiority randomized controlled trial. *Journal of Clinical Periodontology* 2019; **46**:373–381.

López-Pacheco A, Soto-Peñaloza D, Gómez M, Peñarrocha-Oltra D and Alarcón MA. Socket seal surgery techniques in the esthetic zone: a systematic review with meta-analysis and trial sequential analysis of randomized clinical trials. *International Journal of Implant Dentistry* 2021; **7**:1-14.

MacBeth N, Trullenque-Eriksson A, Donos N and Mardas N. Hard and soft tissue changes following alveolar ridge preservation: a systematic review. *Clinical Oral Implants Research* 2017; **28**:982–1004.

Majzoub J, Ravida A, Starch-Jensen T, Tattan M and Suárez-López del Amo F. The Influence of Different Grafting Materials on Alveolar Ridge Preservation: a Systematic Review. *Journal of Oral & Maxillofacial Research* 2019; **10**:1-13.

Meloni SM, Tallarico M, Lolli FM, Deledda A, Pisano M and Jovanovic SA. Postextraction socket preservation using epithelial connective tissue graft vs porcine collagen matrix. 1-year results of a randomised controlled trial. *European Journal of Oral Implantology* 2015; **8**:39–48.

Nart J, Barallat L, Jimenez D et al. Radiographic and histological evaluation of deproteinized bovine bone mineral vs. deproteinized bovine bone mineral with 10% collagen in ridge preservation. A randomized controlled clinical trial. *Clinical Oral Implants Research* 2017; **28**:840–848.

Natto ZS, Parashis A, Steffensen B, Ganguly R, Finkelman MD and Jeong YN. Efficacy of collagen matrix seal and collagen sponge on ridge preservation in combination with bone allograft: A randomized controlled clinical trial. *Journal of Clinical Periodontology* 2017; **44**:649–659.

Nevins M, Camelo M, de Paoli S et al. A study of the fate of the buccal wall of extraction sockets of teeth with prominent roots. *The International Journal of Periodontics & Restorative Dentistry* 2006; **26**:19–29.

Parashis AO, Hawley CE, Stark PC, Ganguly R, Hanley JB and Steffensen B. Prospective Clinical and Radiographic Study of Alveolar Ridge Preservation Combining Freeze-Dried Bone Allograft with Two Xenogeneic Collagen Matrices. *Journal of Periodontology* 2016; **87**:416–425.

Schropp L, Wenzel A, Kostopoulos L and 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**:313–23.

Slot D, Rosema N, Hennequin-Hoenderdos N, Versteeg P, van der Velden U and van der Weijden G. The effect of 1% chlorhexidine gel and 0.12% dentifrice gel on plaque accumulation: a 3-day non-brushing model. *International Journal of Dental Hygiene* 2010; **8**:294–300.

Smukler H, Landi L and Setayesh R. Histomorphometric evaluation of extraction sockets and deficient alveolar ridges treated with allograft and barrier membrane: a pilot study. *The International Journal of Oral & Maxillofacial Implants* 1999; **14**:407–16.

Tan WL, Wong TLT, Wong MCM and Lang NP. A systematic review of post-extractional alveolar hard and soft tissue dimensional changes in humans. *Clinical Oral Implants Research* 2012; **23**:1–21.