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Buccal bone loss after immediate implantation can be reduced by the flapless approach

ABSTRACT

Aim The aim of this study was to evaluate the buccal bone remodeling after immediate implantation with flap or flapless approach.

Material and Methods The mandibular bilateral premolars of 3 dogs were extracted and immediately three implants were placed in both hemi-arches of each dog. Randomly, one hemi-arch was treated with the flapless approach, while in the contra lateral hemi-arch tooth extractions and implant placement were done after mucoperiosteal flap elevation. Non-submerged healing of 12 weeks was provided for both groups. Histomorphometric analysis was done to compare buccal and lingual bone height loss, bone density and bone-to-implant contact in the groups. Fluorescence analysis was performed to investigate the dynamic of bone remodeling in the different groups.

Results There was a significant association between the surgical flap and the extent of bone resorption around immediate implants. The loss of buccal bone height was significantly lower in the flapless group when compared to the flap group (0.98 mm x 2.14 mm, respectively, $p<0.05$). The coronal and apical buccal bone densities of the flap group were significantly higher when compared to the lingual components, showing anatomical differences between the bone plates. Fluorescence analysis showed no major differences in bone healing between the flap and flapless groups, supporting that the higher loss of buccal bone height is linked to the anatomic characteristics of this plate and to the negative influence of the detachment of the periosteum in immediate implant therapy.

Conclusion The flapless approach for immediate post-extraction implants reduces the buccal bone height loss.

KEY WORDS Animal model; Bone resorption; Flapless surgery; Immediate implants.

INTRODUCTION

The immediate implant placement after tooth extraction has been reported to be as predictable as placing implants into healed sites (1); but with advantages such as reduced number of surgical procedures (2-8), reduction of the overall treatment time and also a possible preservation of the morphological contour of the ridges (3, 6). However, in regard to this last consideration, some studies in animals have shown contradictory results, describing pronounced resorption of the buccal, and to some extent, the lingual bone plates after implant placement in fresh extraction sockets (9, 10).

Novaes Jr. (11) showed that the morphological and vascular characteristics of the bone crests may have an important impact on the bone remodeling process that occurs immediately after implantation. Through a histological analysis of specimens sectioned in buccal-lingual direction, they observed that the width of both bone plates increased from the coronal third to the most apical third, being the buccal plates always significantly thinner when compared to the lingual bone plates. More specifically, they described the coronal portion of the buccal bone plate as extremely delicate, which is in agreement with previous observations (10, 12), and this characteristic may explain almost in part why the buccal bone plates are more easily resorbed. They showed some slides where the buccal bone plates appeared without or with very few marrow spaces. In fact, the buccal bone plates were constituted, in a statistically significant way, by a higher cortical bone when compared to the lingual bone plates, in first and second coronal thirds; this bone density analysis was obtained by the subtraction of the bone marrow area from the total bone area. Interestingly, they also evidenced in some histological observations, marrow

spaces in direct contact to the periosteum in the buccal bone plate, sustaining the hypothesis that one of the main functions of the periosteum and periodontal ligament blood vessels is to supply nutrients and cells to the alveolar bone (13).
Conventionally, immediate implantation surgeries give emphasis to some important precautions such as less traumatic tooth extraction and implant primary stability, and are usually done with sulcular incisions and mucoperiosteal flap elevation. However, it is known that the displacement of the periosteum and alveolar bone denudement result in an acute inflammatory response and consequently in bone resorption (14-16). Osteoclasts were observed on surgical exposed alveolar bone areas during the first two weeks of wound healing (17). Besides, a relevant information to consider is that, although a pronounced loss of the buccal bone wall were frequently described after mucoperiosteal surgeries applied in periodontal treatment of dentate areas, the same was not observed on the thicker lingual wall (16, 18, 19).
Kim et al. (20) compared the vascularity of peri-implant mucosa between flap and flapless implant surgeries in a dog model and showed that the soft tissue around implants in flapless sites appeared to be free from signs of inflammation, while approximately half of the implants in the flap sites exhibited a surrounding edematous tissue that bled when gently probed. Additionally, the number of vessels observed was 51.4 ± 9.2 in the flapless group and 38.2 ± 8.1 in the flap group, and this difference was statistically significant. Based on these findings, they suggested that the more richly vascularized peri-implant mucosa provided by the flapless procedure is directly related to an increased blood supply around the implant, which may strengthen the resistance to inflammation.
The purpose of the present study was to evaluate if the flapless approach can interfere in the buccal bone remodeling after immediate implantation in mongrel dogs, analyzing the histomorphometric parameters of bone height loss, bone density and bone-to-implant contact after 12 weeks of healing and also the dynamic of bone remodeling in four different times along this period through fluorescence analysis.

MATERIALS AND METHODS

Surgical procedure

The study protocol was approved by the Institution's Animal Research Committee of the School of Dentistry of Ribeirão Preto- University of São Paulo and was performed in three young adult male mongrel dogs that weighed approximately 16 kg. The animals presented intact maxillas, no general occlusal trauma, and no oral viral or fungal lesions and were in good general health, with no systemic involvement as determined by a veterinarian following clinical examination.
Food was withheld in the night preceding surgeries. The animals were pre-anaesthetized with acepromazine 0,2% - 0,05 mg/kg IM. After that an intravenous catheter was placed in the foreleg for induction with thiopental 2,5% - 5 a 8 mg/Kg IV. Animals were then moved to the operating room and maintained on gas anesthesia (1–2% isoflurane/O2 to effect). Conventional dental infiltration anesthesia was used at the surgical sites. The animals received a slow constant rate infusion of lactated Ringer's solution (10–20 ml/kg/h IV) to maintain hydration during surgery. These procedures were made and accompanied by a veterinarian.
The surgical procedures for the mandibular premolar extractions were done in each hemi-arch of each dog. Randomly, one of the sides was treated with the flapless approach (experimental group) (fig. 1A), while the contralateral side was treated with mucoperiosteal flaps (fig. 1B). The teeth were sectioned in a bucco-lingual direction at the bifurcation so that the roots could be individually extracted, without damaging the bony walls, using a periotome. After alveolar debridement, three Ankylos implants measuring 3.3 x 9.5 mm (diameter and length, respectively) were immediately inserted in the mesial socket of the correspondent three pre-molars in both hemi-arches of each dog, totaling 18 implants in the experiment. The implants were placed at the level of bone crest and a gap of 1mm from the buccal cortical wall to the implant was always left (fig. 2) without invading the lingual bone plate with

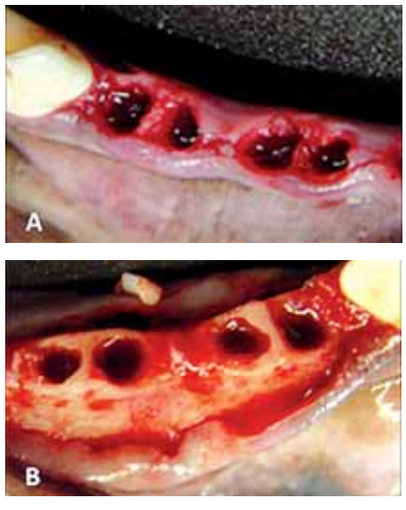

Fig. 1 *One random hemi-arch was treated with the flapless approach (A) and the opposite hemi-arch was treated with a mucoperiosteal flap (B).*

the drill or the implant. Subsequently, healing caps of 1.5 mm of height were adjusted in order to provide a non-submerged healing in both groups. The flaps of the control group were repositioned and sutured with absorbable sutures (Vicryl, Ethicon, Inc., Johnson & Johnson Company, São José dos Campos-SP, Brasil), while the soft tissues were accommodated and then sutured in the experimental group. No grafting materials were used in the gaps between the buccal plates and the implants.

The animals received painkillers and anti-inflammatory agents. A broad spectrum antibiotic (penicillin and streptomycin 20,000 IU; 1.0 g/10 kg IM) was administered immediately post-surgery and re-dosed after 4 days. The animals were maintained on a soft diet for 14 days when the sutures were removed. The healing was evaluated weekly and plaque control was maintained by flushing the oral cavity with chlorhexidine gluconate. The remained teeth were cleaned monthly with ultrasonic points and all implants remained non-submerged during the experimental period.

During the healing period fluorescence bone markers were administered (21) to observe the dynamics of bone formation. One week after implant placement, calcein green (75 mg /Kg body weight-Sigma Chemical Co., St Louis, MO, USA) was intravenously administered; at the second week, it was administered 300 mg of red alizarin S/Kg body weight (Sigma); after 4 weeks it was administered 150 mg oxytetracyclin HCl/Kg body weight (Sigma); and finally after 12 weeks 75 mg calcein blue/Kg body weight (Sigma) were also administered. All dyes were prepared immediately before use with 2% sodium bicarbonate or saline. After preparation, pH was adjusted to 7.4 and the solution was filtered through a 0.45 Ìm filter (Schleider & Schuell GmbH, Dassel, Germany). Each dog received a total dose of 3 ml.

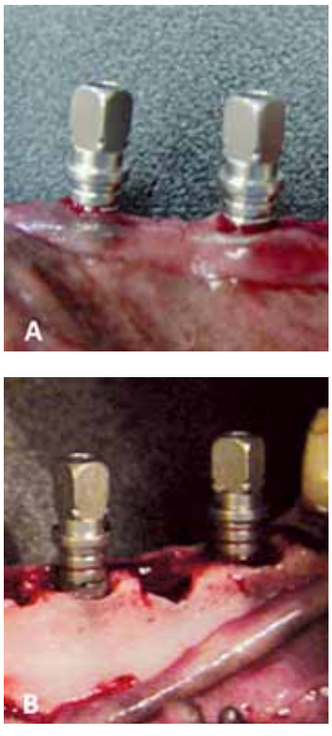

Fig. 2 *The implants were immediately inserted in the mesial alveolus of the correspondent extracted pre-molars in both hemi-arches of each dog. A jumping gap of 1mm from the buccal cortical wall to the implant was always left. (A) Image representative of the flapless group and (B) image representative of the flap group..*

Histological processing

The animals were sedated and then sacrificed with an overdose of thiopental twelve weeks after implant placement. The hemi-mandibles were removed, dissected and fixed in 4% phosphate-buffered formalin pH 7, for 10 days, and transferred to a solution of 70% ethanol until processing. The specimens were dehydrated in increasing concentrations of alcohol up to 100%, infiltrated and embedded in LR White resin (London Resin Company, Berkshire, England), and hard-sectioned in bucco-lingual direction using the technique described by Donath & Breuner (22). The most central sections were stained with Stevenel's blue and Alizarin red S for histometric analysis using optic microscopy.

Histomorphometric analysis

Longitudinal buccal-lingual histological sections from each implant were captured through a video camera Leica DC 300F (Leica Microsystems GmbH, Nussloch, Germany) joined to a stereomicroscope Leica MZFL III (Leica Microsystems GmbH, Nussloch, Germany). The images were analyzed through the Image J program (National Institutes of Health, Bethesda,USA). The buccal bone wall resorption was determined in relation to the lingual bone wall as a linear measurement (relative measurement) (Fig. 3, 4). A horizontal imaginary line was drawn in order to evidence the height of the lingual bone plate, and then the measurement of the buccal bone wall resorption was obtained vertically from that line to the peak of the buccal bone plate. The buccal and lingual bone plates were also measured from the shoulder of the implant to the first bone-to-implant contact (absolute measurement). The percentages of bone-to-implant contact (BIC) were calculated throughout the implant perimeter, from the first coronal bone-to-implant contact, considering the mineralized bone in direct contact with the implant surface. The bone density was determined within two rectangles, one of them adjacent to the implant surface (BDA), and the other as mirror image of the first, but distant to the implant surface (BDD). This

analysis was done in two different positions of the implants, one coronal and other apical, permitting an intra-group evaluation. The bone density measurements evaluated the percentages of mineralized bone in relation to the percentages of marrow cavities. A single examiner, with no knowledge of the experimental groups made the measurements.

Fluorescence analysis

Fluorescence microscopic images were longitudinally captured from each implant through a video camera Leica DC 300F (Leica Microsystems GmbH, Nussloch, Germany) joined to a stereomicroscope Leica MZFL III (Leica Microsystems GmbH, Nussloch, Germany), using appropriated barrier filters. The filters of wavelengths used was I3 for calcein green that has an excitation level between 450-490 nm, N2-1 for red alizarin S that has an excitation level between 515-560 nm, D for oxytetracyclin HCl that has an excitation level between 355-425 nm and A for calcein blue that has an excitation level between 340-380 nm. All the images were adjusted and analyzed through the Image J program (National Institutes of Health, Bethesda, USA) to determine the percentages of bone marked.

The bone marked was determined in two different positions along the implants, at coronal and apical levels in both buccal and lingual sides, using the same pre-determined rectangle for all the specimens (fig. 5, 6). The quantity of bone marked represented the percentages of fluorescent bone in relation to the total area. A single examiner, with no knowledge of the experimental groups made the measurements.

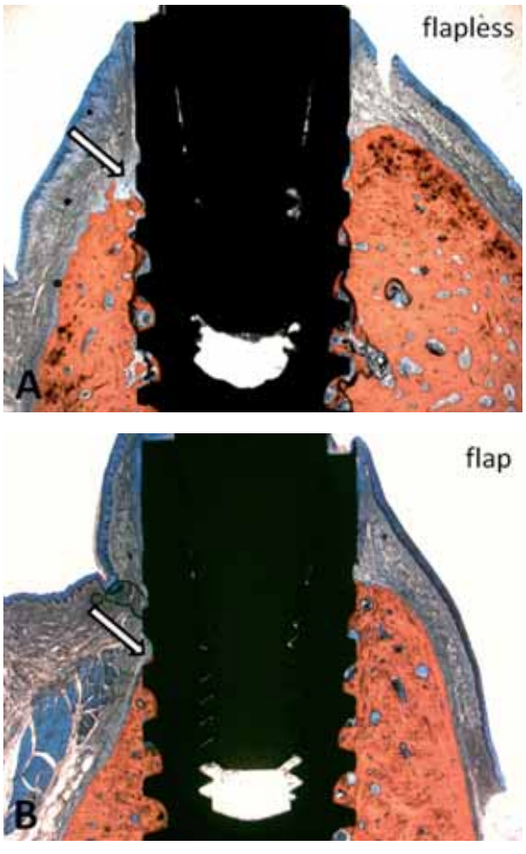

Fig. 3 *After 12 weeks of immediate implantation the specimens were sectioned in bucco-lingual direction to compare buccal and lingual bone plate's dimensions. In (A) a representative image of the flapless group, while in (B) a representative image of the group treated with the elevation of a mucoperiosteal flap. Compare the heights of the buccal bone plates (arrows) between them. Note also the differences of bone density between the buccal bone plates (on the left) and lingual bone plates (on the right) of both images. Stevenel's blue and Alizarin red S stain; magnification x 10.*

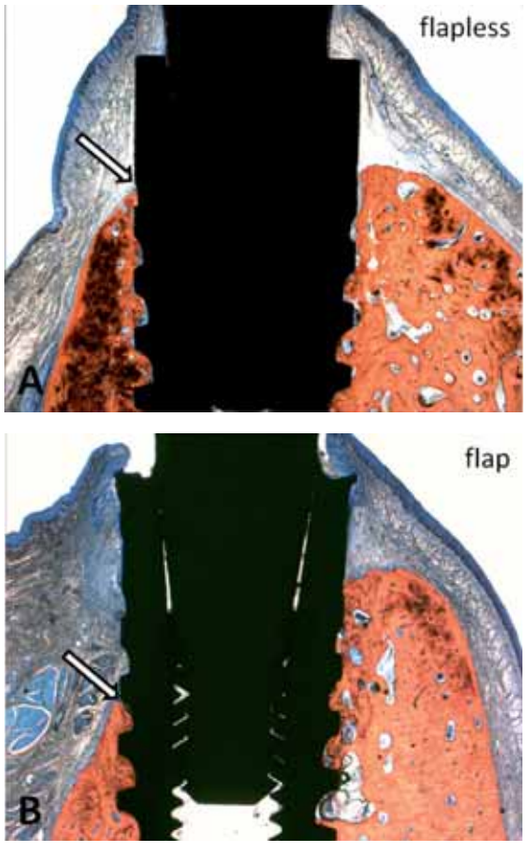

Fig. 4 *As in Figure 3, in (A) there is a representative image of the flapless group, while in (B) a representative image of the flap group. Note again the difference of vertical bone loss between them. In these images is more evident the higher bone density found in the buccal bone plates (on the left) when compared to the lingual bone plates (on the right), which is easily seen by the different number and dimension of marrow spaces found in them.*
Stevenel's blue and Alizarin red S stain; magnification x 10.

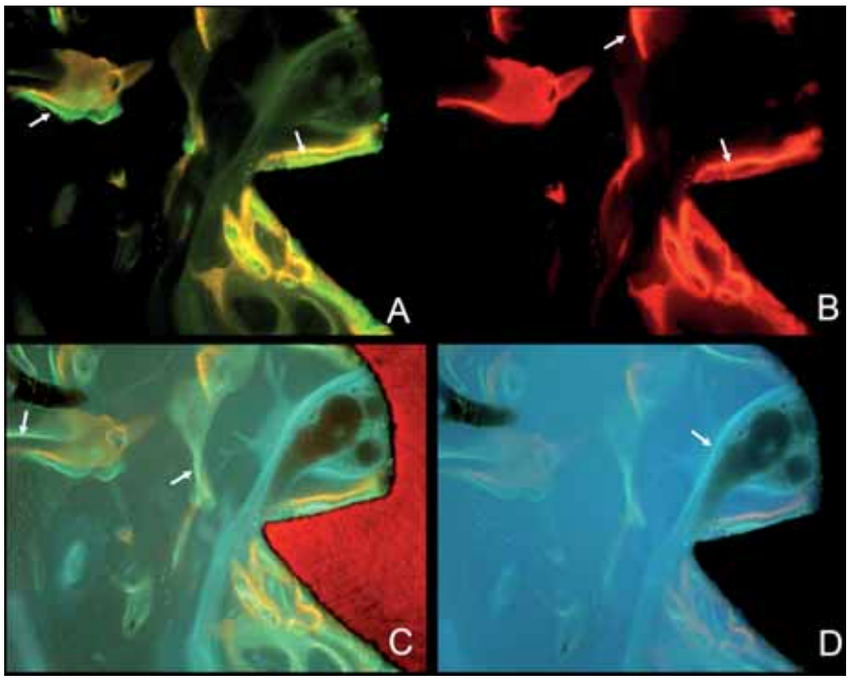

Fig. 5 *Different bone markers at the coronal level of the implant. A: calcein green; B: red alizarin; C: oxytetracyclin; D: calcein blue*

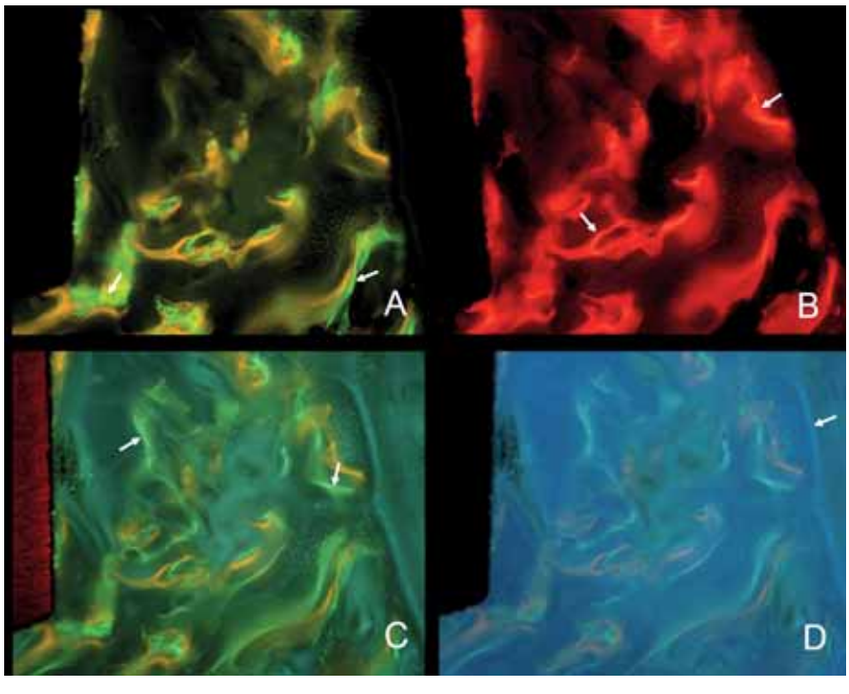

Fig. 6 *Different bone markers at the apical level of the implant. A: calcein green; B: alizarin red; C: oxytetracyclin; D: calcein blue.*

Statistical analysis

Mean values and standard deviations were calculated. The data were grouped using the dogs as units for analysis. The mean differences between the groups for each histomorphometric parameter were analyzed through the Mann-Whitney nonparametric test with a confidence level of 95%. Besides for the fluorescence analysis, all measurements were statistically evaluated using the non-parametric analysis of variance, Kruskal-Wallis, and Dunn test was used for multiple comparisons among the means. The confidence level was 95%.

RESULTS

Clinical and histological observations

Healing was uneventful for all animals and no implant was lost. All implants became osseointegrated after the 12-week postoperative

		FLAPLESS		FLAP	
		coronal	apical	coronal	apical
Buccal	BDA	90.37 ± 6.12	85.80 ± 13.87 * / ?	93.42 ± 4.43 #	94.39 ± 5.29 &
	BDD	91.95 ± 8.84	95.52 ± 2.56 * / ?	97.08 ± 2.19 ?	95.91 ± 2.96 ¥
	BIC	77.39 ± 9.07		77.75 ± 12.58 **	
Lingual	BDA	87.13 ± 7.99 ¨	59.88 ± 13.19 ? / ¨	84.55 ± 4.97 # / »	50.69 ± 9.90 & /»
	BDD	86.80 ± 7.01 :	56.82 ± 14.19 ? / :	89.57 ± 5.83 ? / º	46.70 ± 9.00 ¥ / º
	BIC	70.50 ± 12.17		66.00 ± 7.69 **	

¨ p=0.0023
: p=0.0006
* p=0.0262
? p=0.0070
? p=0.0006
p=0.0070
? p= 0.0041
» p=0.0006
º p=0.0006
& p=0.0006
¥ p=0.0006
** p=0.0262

Table 1 *Percentages of bone density adjacent (BDA) and distant (BDD) and bone-to-implant contact (BIC) described as mean ± SD.*

period. The marginal gaps between the buccal walls and the implants disappeared without the migration of connective tissue in both groups.

Histomorphometric analysis

The loss of buccal bone height was statistically lower in the flapless group when compared to the flap group (0.98 ± 0.45 mm x 2.14 ± 0.34 mm) (p<0.0001). Additionally the comparisons of the absolute values of bone loss around the implants for the flapless and flap groups showed statistically significant differences between the buccal bone resorption of the experimental groups (2.46 ± 0.42 mm x 3.83 ± 0.21 mm, flapless and flap, respectively) (p<0.0001), but not between the lingual remaining bone heights (1.48 ± 0.27 mm x 1.70 ± 0.31 mm, flapless and flap, respectively). The comparisons within the groups showed statistically significant differences between the buccal and lingual bone resorption in the flapless (2.46 ± 0.42 mm x 1.48 ± 0.27 mm, buccal and lingual, respectively) (p<0.0001) and flap groups (3.83 ± 0.21 mm x 1.70 ± 0.31 mm, buccal and lingual, respectively) (p<0.0001). The loss of the buccal bone in the flap group was more than 100% greater than the lingual bone.

The buccal bone density was numerically higher in all the parameters evaluated when compared to the lingual bone density (Fig. 3. 4). These differences were statistically significant for all the comparisons tested, except for the flapless coronal buccal bone density (table 1).

Although the buccal bone density was numerically higher for the flap group compared to the flapless group, these differences were not statistically significant for both coronal and apical parameters (table 1).

The comparisons between coronal and apical bone density were statistically significant only for the lingual bone for both flapless and flap groups, with the apical bone having a lower density (table 1).

There were no statistically significant differences between adjacent and distant bone densities for all the possible comparisons (table 1).

All the implants presented considerable good

		BUCCAL		LINGUAL	
		coronal	apical	coronal	apical
Calcein green	flap	0	8.66	5.41	6.6
	flapless	1.76	9.83	9.02	6.54
	p value	p>0.05	p>0.05	p>0.05	p>0.05
Alizarin red	flap	1.05	21.1	21.71	15.83
	flapless	9.83	26.07	13.36	12.76
	p value	p>0.05	p>0.05	p>0.05	p>0.05
Oxytetracyclin	flap	0	5.71	3.62	2.16
	flapless	1.26	5.31	4.36	1.5
	p value	p>0.05	p>0.05	p>0.05	p>0.05
Calcein blue	flap	0	2.4	1.12	2.27
	flapless	1.49	2.89	1.79	1.45
	p value	p>0.05	p>0.05	p>0.05	p>0.05

Table 2 *Fluorescence analysis. Comparisons between flapless and flap groups considering the percentage of each bone marker administered during different time periods of bone healing.*

GROUP	EVALUATED AREA		BONE MARKER				P VALUE	
			Calcein green	Alizarin red	Oxytetracyc	Calcein blue		
Flapless	Buccal	Apical	9.83	26.07*	5.31	2.89*	*p<0.05	
		Coronal	1.76	9.83	1.26	1.49	p>0.05	
	Lingual	Apical	6.54	12.76* / *	1.50*	1.45*	*p<0.01	*p<0.01
		Coronal	9.02	13.35*	4.36	1.79*	*p<0.01	
Flap	Buccal	Apical	8.66	21.1*	5.71	2.40*	*p<0.01	
		Coronal	0	1.05	0	0	p>0.05	
	Lingual	Apical	6.6	15.83	2.16	2.27	p>0.05	
		Coronal	5.41	21.71*	3.62	1.12*	*p<0.01	

Table 3 *Fluorescent analysis. Intra-group evaluation of the percentage of each different bone marker found in the apical and coronal areas of the experimental groups.*

GROUP	EVALUATED AREA		BONE MARKER			
			Calcein green	Alizarin red	Oxytetracyc	Calcein blue
Flapless	Buccal	Apical	9.83	26.07	5.31	2.89
	Lingual	Apical	6.54	12.76	1.5	1.45
		p value	p>0.05	p>0.05	p>0.05	p>0.05
	Buccal	Coronal	1.76	9.83	1.26	1.49
	Lingual	Coronal	9.02	13.35	4.36	1.79
		p value	p>0.05	p>0.05	p>0.05	p>0.05
Flap	Buccal	Apical	8.66	21.1	5.71	2.4
	Lingual	Apical	6.6	15.83	2.16	2.27
		p value	p>0.05	p>0.05	p>0.05	p>0.05
	Buccal	Coronal	0.00*	1.05*	0	0
	Lingual	Coronal	5.41*	21.71*	3.62	1.12
		p value	*p<0.05	*p<0.01	p>0.05	p>0.05

Table 4 *Fluorescent analysis. Intra-group evaluation of the percentage of each different bone marker found in the buccal and lingual areas of the experimental groups.*

indications of bone to implant contact and the results were remarkably similar between the groups. The buccal BIC in both groups is numerically higher when compared to the lingual BIC results, and statistically significant in the flap group (table 1).

The analysis under fluorescent microscopy showed bone remodeling in the groups evaluated. The old bone always appeared darker and without labeling. Calcein green appeared in very well delineated green bands (fig. 5, 6A) as did in red the alizarin red marker, which in some specimens also showed a smeared diffuse pattern (fig. 5, 6B); oxytetracyclin showed thin yellow-green lines (fig. 5, 6C) and finally calcein blue was characterized by a soft blue color in a very diffuse pattern (fig. 5 and 6D). In many specimens the secondary osteons were demonstrated by the deposition of the labels in a concentric arrangement. The bone marker quantifications sequentially represented the healing pattern of each different group. The percentages of newly formed bone in the different parts are described in tables 2, 3 and 4. Table 2 represents the analysis between the flap and flapless groups considering the different parts, while tables 3 and 4 show the results of the intra-group analysis, separately.

A pattern of bone remodeling between the experimental groups (flap and flapless) and also between the different evaluated areas (buccal and lingual; apical and coronal) was detected (fig. 7, 8; tables 3, 4). No statistically significant differences were found between the flap and flapless groups (table 2), however numerically different values of bone formation were observed at the buccal coronal area of the groups, especially at the red alizarin period of application. Generally, the initial phases of bone remodeling that were represented by the calcein green and red alizarin, one week and two weeks after implant placement respectively, showed higher values of bone formation when compared to the other periods evaluated, after 4 and 12 weeks of implant placement. The alizarin red bone marker comprised the peak of bone formation for all groups. Administered after 2 weeks of implant placement, it exhibited the highest levels of marked bone (fig. 7, 8).

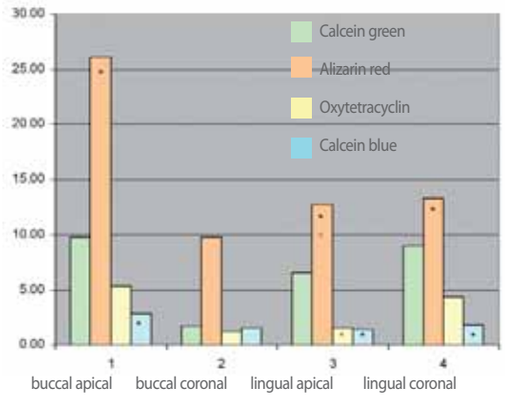

Fig. 7 *Dynamic of bone formation at the different evaluated areas in the flapless group.*

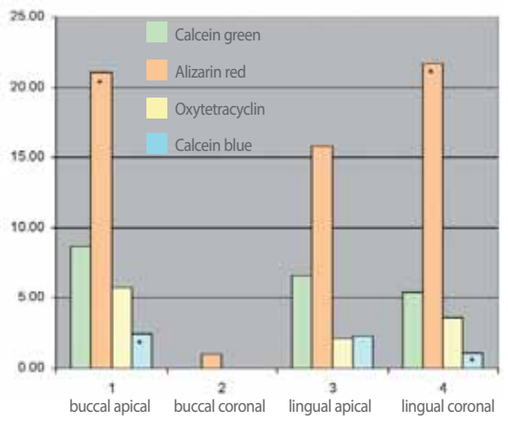

Fig. 8 *Dynamic of bone formation at the different evaluated areas in the flap group.*

Statistically significant differences were observed between red alizarin and calcein blue for bone remodeling evaluations at the flapless buccal apical areas and also at the flapless lingual apical and coronal areas (table 3). Still considering the intra-group analysis, the flap buccal apical and the flap lingual coronal areas also showed statistically significant differences between the alizarin red and calcein blue marked bone (table 3).
When comparing the buccal and the lingual areas of the flap and flapless groups, statistically significant differences were found only between buccal coronal and lingual coronal areas of the flap group at the calcein green and red alizarin periods of application (table 4).

DISCUSSION

The flapless surgical approach significantly favored the preservation of the alveolar buccal plate height after immediate implant placement and a reasonable explanation could be the non-detachment of the periosteum and its vascular network. In this study the only difference between the groups was the flap elevation in the control group, which exhibited at least twice as more buccal bone loss when compared to the flapless group. Even better results were demonstrated by another study with a similar methodology, where the buccal bone loss of 2.11 mm for the flap sites was confronted by the 0.6 mm found at the test immediate implants treated with flapless surgery (23).
Many years ago, Wilderman et al. (24) have primarily demonstrated that "although the exposure of bone by surgery allows its observation, some bone resorption is the penalty for this type of examination". More recently, the evaluation of the microvascular responses after mucoperiosteal flap surgery in dogs confirmed that the elevation of the periosteum may cause circulatory insufficiency and then bone resorption (25). In general, the bone surface that is temporarily exposed usually undergoes a necrotic process that finishes in bone resorption, with exception of the broad bone plate that contains a significant number of marrow spaces and could have less bone height loss at the end of the healing period.
Considering that one of the main functions of the periodontal ligament (PDL) blood vessels is to supply nutrients to the osteoblasts in the alveolar bone (13), it is easy to understand that after tooth extractions, only the vascularization provided by the periosteum remains. However, the elevation of mucoperiosteal flaps also compromises the blood supply from the periosteum. Fickl et al. (26) evaluated the hypothesis that tooth extraction without the elevation of a mucoperiosteal flap may decrease the post-surgery resorption level, and demonstrated that the act of leaving the periosteum in place decreased the resorption index of the extraction socket. They highlighted that the great impact of this finding might be when dealing with thin periodontal biotypes, where the osteoclastic activities of the internal and external sides could merge together and cause a more pronounced buccal bone plate loss.
The results of the present study were consistent with these statements, especially for the flap approach group in which the loss of the buccal bone was more than 100% greater than the lingual bone as shown by the absolute measurements of bone loss around the implants. The statistically significant difference between flapless and flap groups when considering the buccal bone loss confirmed the importance of periosteum preservation in this type of implant therapy. On the other hand, there were no significant differences between the flap and flapless on the lingual bone plate resorption, indicating that the

morphology of the buccal and lingual plates might represent another crucial factor in determining the final bone resorption.

Based on these facts it could be speculated that the immediate implant therapy was not the only factor that influenced the high level of buccal bone height loss of 2,5 mm in relation to the lingual bone plate described by Araujo et al. (9) after flap surgery.

In our histological specimens the buccal bone crest appeared significantly thinner when compared to the lingual component. This pattern was also observed in different studies (10,12,17,19,27). Furthermore, the bone densities of buccal and lingual plates were very different in both groups. In general, while the buccal plates were constituted by a cortical bone type with sparse and decreased number of marrow areas, the lingual bone plates exhibited numerous and large marrow areas. This difference between the buccal and lingual bone densities was statistically significant in the apical portion of test and control groups, and was also statistically significant in the coronal portion of the flap group. This last finding could mean that this portion exhibited insufficient bone marrow spaces and source of blood vessels, and consequentially compromised angiogenesis that is usually related to bone loss (11, 25). There were no statistically significant differences between the bone densities adjacent and distant to the implants in both groups, but there was for the buccal bone densities of the apical portion of the flapless group. The significant lower density adjacent to the implant of the buccal bone observed in the intra-group evaluation (85.80% adjacent and 95.52% distant), and also the numerical difference between the groups considering this parameter (85.80% for flapless and 94.39% for flap) could be understood as another advantage of the non-detachment of the periosteum, providing vessels and consequently nutrients to the cortical bone plates.

All the implants presented good BIC levels and the results were very similar between flapless and flap groups. The buccal BIC is numerically higher in both groups when compared to the lingual BIC and this could be related to the higher number of marrow areas found in the lingual bone plate.

To sum up, the current study supports the existence of a close relationship between angiogenesis and bone resorption/formation (25), in which the remodeling process is strongly dependent on the interaction between new blood vessels and bone.

Qahash et al. (28) demonstrated a significant association between the width of the buccal alveolar ridge and extent of bone resorption evaluated by incandescent and fluorescent light microscopy. They suggested that the width of the buccal alveolar ridge should be at least 2 mm to maintain the alveolar bone level. These observations have general implications for implant placement with most surgical protocols, and even more for immediate implantation. Studies about the alveolar bone healing potential in peri-implant critical-size defects, showed that the thicker lingual bone plate provided a large wound space that was correlated with enhanced bone regeneration, while the implants placed closer to the buccal plate were associated with increased crestal bone loss (29,30).

Another comparative study between flapless and flap surgeries for immediate post-extraction implants, also found a minor reduction of the buccal bone plate with the flapless approach, but emphasized the importance of the location of the implants in the confines of the alveolus (31). Based on this, it could be discussed that one reason for the higher buccal bone plate resorption of Araujo et al. (19) study could be due to the use of a 4.1 mm diameter implants in alveoli that are smaller (3.5 mm is the diameter of third premolars and of 3.9 mm of fourth premolars in dogs); in other words, the diameter of the implant was greater than the alveoli themselves.

In the present study the implants were placed 1 mm away from the buccal marginal bone wall without invading the lingual bone plate with the drill or the implant. No residual defect was observed on the histological specimens after 12 weeks of healing and the formation of new bone could be a possible explanation as well as bone loss to some extent. This jumping gap distance has already been studied and it was shown that this defect may heal with new bone and a high degree of osseointegration without the use of barrier membranes (32). It was described that this kind of defect “allowed the formation of a coagulum that, even in the absence of a barrier membrane, it was properly protected by the periosteum of the soft tissue flap. In other words, during the healing of a ‘self-contained’ bone defect and in the presence of a proper periosteum, the use of a barrier membrane may not be required”, but this is dependent on the implant surface and time of healing allowed after implant installation and gap distance.

From the fluorescence analysis of the present study no statistically significant differences were obtained between the flap and flapless groups, but the evaluation of the buccal coronal areas showed numerically higher new bone formation for the flapless group and the lack of statistical significance could be explained by the size of the sample.

It was also observed that bone remodeling followed a pattern not only in the two experimental groups, but also in the different evaluated sections of the implants in an intra-group analysis – coronal and apical, buccal and lingual.

It is well-known that the bone formative and resorptive phases are systematically intercalated (33).

In the present study the peak of bone mineralization for the groups and subgroups studied comprised the period of 2 weeks after implant placement as marked by the red alizarin dye. This is in accordance to Abrahamsson et al. (33) that had already characterized this time period as a very active phase in the process of mineralization.
The statistically significant differences found within the groups, for example between the red alizarin and calcein blue marked bone in the buccal apical area confirmed that bone remodeling is an ongoing process that also involves a decrease in the mineralization levels along time. This could explain the replacement of woven bone by lamellar bone as a physiological process.
Finally, the evaluation within the experimental groups comparing buccal and lingual halves showed statistically significant differences for the flap coronal area at the first two periods of evaluation. This result could be explained by the very low values of bone thickness found for the buccal plate in this area, reinforcing the histomorphometric findings that described the lingual plates as thicker.

CONCLUSION

In summary no major differences in the dynamics of bone healing, evidenced by the fluorescence analysis, has been detected between the flap and flapless groups that supports the hypothesis that the higher loss of buccal bone height is linked to the anatomic characteristics of the buccal bone, the negative influence of the detachment of the periosteum during the flap procedure in immediate implant therapy and the presence of a gap between the implant and the buccal bone plate.
Within the limitations of this study, it can be concluded that the flapless approach for immediate post-extraction implants reduces the buccal bone plate resorption

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