CLINICAL ORAL IMPLANTS RESEARCH

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Graft incorporation and implant osseointegration following the use of autologous and fresh-frozen allogeneic block bone grafts for lateral ridge augmentation

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Abstract

Objectives: To compare autogenous bone (AT) and fresh-frozen allogeneic bone (AL) in terms of histomorphometrical graft incorporation and implant osseointegration after grafting for lateral ridge augmentation in humans.

Materials and methods: Thirty-four patients were treated with either AL (20 patients) or AT (14 patients) onlay grafts. During implant installation surgery 6 months after grafting, cylindrical biopsies were harvested perpendicularly to the lateral aspect of the augmented alveolar ridge. Additionally, titanium mini-implants were installed in the grafted regions, also perpendicularly to the ridge; these were biopsied during second-stage surgery. Histological/histomorphometric analysis was performed using decalcified and non-decalcified sections.

Results: Histological analysis revealed areas of necrotic bone (NcB) occasionally in contact with or completely engulfed by newly formed vital bone (VB) in both AT and AL groups (55.9 $\pm$ 27.6 vs. 43.1 $\pm$ 20.3, respectively; $P = 0.19$). Statistically significant larger amounts of VB (27.6 $\pm$ 17.5 vs. 8.4 $\pm$ 4.9, respectively; $P = 0.0002$) and less soft connective tissue (ST) (16.4 $\pm$ 15.6 vs. 48.4 $\pm$ 18.1, respectively; $P \leq 0.0001$) were seen for AT compared with AL. No significant differences were observed between the groups regarding both bone-to-implant contact (BIC) and the bone area between implant threads (BA) on the mini-implant biopsies.

Conclusion: Allogeneic bone block grafts may be an option in cases where a limited amount of augmentation is needed, and the future implant can be expected confined within the inner aspect of the bone block. However, the clinical impact of the relatively poor graft incorporation on the long-term performance of oral implants placed in AL grafts remains obscure.

Presence of adequate local bone volume is among the important factors connected to the high success rates of oral implants (Stanford 2002; Renouard & Nisand 2006). However, several factors such as tooth loss, trauma, periodontitis or other disease often diminish the amount of available bone (Barber & Betts 1993). In absence of adequate bone volume, one of the most predictable ways to restore bone anatomy and enable oral rehabilitation is bone grafting (Chiapasco et al. 2006).

The use of autologous bone (AT) as grafting material is considered the gold standard for bone augmentation procedures in the maxillo-facial region (Misch & Misch 1995; Nowzari & Aalam 2007). Nevertheless, this technique is also associated with some pitfalls and mainly connected to the donor site, such as post-surgical pain, risk of paresthesia, and limitations in the quality and quantity of available bone, leading thus to the necessity of other bone substitute materials (Zerbo et al. 2003). One of the substitute materials for AT that has been largely cited in the literature is fresh-frozen allogeneic bone (AL), which seems to provide a reasonable source for grafting material without the need for a second surgical area (Goldberg & Stevenson 1987; Lee et al. 2010). Other advantages such as unlimited availability and reduced surgical time make this material a plausible clinical alternative (Mankin et al. 1983; Spin-Neto et al. 2011a).

One of the facts that place AL among the range of possible alternatives for bone augmentation

procedures was the establishment of strict guidelines for donor tissue source and processing, including donor screening, bone harvesting, processing and storage, together with the track-record protocols that must be respected (Troyer 2008). Cases of infection, including hepatitis and HIV, due to implantation of contaminated allograft bone have been reported in the past (Simonds et al. 1992; Conrad et al. 1995; Schratt et al. 1996). However, these cases regarded older donor screening procedures and bone allograft processing methods, while newer protocols have significantly increased the safety of AL grafts (Hinsenkamp et al. 2012). Nevertheless, there are still concerns regarding the antigenicity and immunogenicity of AL grafts (Waasdorp & Reynolds 2010).

Allogeneic bone grafts exert primarily osteoconductive action (Mizutani et al. 1990) and seem to show slower incorporation and remodeling (Waasdorp & Reynolds 2010; Spin-Neto et al. 2011a). Recently, lower cumulative success rates of oral implants placed in association with AL grafts have been reported (Carinci et al. 2010). In context, there is a lack of well-conducted, controlled, human histological studies addressing those issues. Therefore, the aim of this study is to histomorphometrically evaluate AL block graft incorporation and remodeling, and implant osseointegration in patients subjected to lateral alveolar ridge augmentation, in comparison with AT grafting.

Materials and methods

The research protocol of this controlled case series was approved by the Araraquara School of Dentistry Ethics Committee (CEP-FO/Car) and by the National Research Ethics Committee (CONEP-MS) under the protocol number 36/08, and it is in accordance with the World Medical Association Declaration of Helsinki (2008).

Patient selection

The patients included herein are from those presenting for treatment in the Department of Periodontology, Araraquara Dental School (UNESP – Univ. Estadual Paulista), Araraquara, São Paulo, Brazil, and desiring oral rehabilitation with titanium implants. The patients had at least one site with severe bone deficiency (i.e., <4 mm alveolar ridge width) precluding placement of a regular size implant. None of the patients presented with systemic diseases affecting bone turnover, or were pregnant or lactating, or had habits that could interfere with treatment (for example, smoking, alcoholism, and drug use). Alveolar ridge width was determined on the cross-sectional view of CBCT (i-CAT Classic, Imaging Sciences International, Hatfield, PA, USA)-generated images (DICOM-based data sets) with a resolution of 96 dpi, 14-bit gray scale, and 0.25 mm voxel size. The CBCT unit was set at 120 kVp, 5 mA, with a 20-sec. exposure time.

Patients judged as not having adequate amounts of donor intraoral bone were treated with AL grafts, that is, group allocation was performed in a non-randomized prospective manner and was based on the treatment needs of each individual patient. This decision was taken based on the clinical screening examination and the CBCT examination and depended on a subjective judgment of the amount of bone resorption and/or number of sites requiring reconstruction in each patient. Thus, 20 patients were treated with AL and 14 with AT (12 men/22 women; average age: 47 years; range: 27–69 years) from May to December 2009. To fulfill Brazilian regulations, documents regarding allogeneic biomaterial request were filled out and sent to the registered bone bank that supplied the allografts (UniOss, Marília, Brazil) prior to surgery. The fresh-frozen allogeneic bone blocks were collected from the femoral head and processed according to the American Association of Tissue Banking guidelines – AATB (Troyer 2008).

Ridge augmentation procedure

Immediately prior to surgery, all patients rinsed their mouth with 15 ml 0.12% chlorhexidine digluconate for 1 minute, then povidone-iodine 10% solution was applied to the peri-oral skin, and the patients were covered with sterile drapes. Under local anesthesia, a full thickness flap was raised to provide a full visualization of the alveolar ridge. In the AT group, cortical block grafts of adequate size according to defect dimensions were retrieved from the mandibular ramus. In the AL group, standard size cortico-cancellous bone blocks (15 × 10 × 6 mm) were used. The AL block was removed from the freezer and put into sterile saline solution for 10 min prior to use, allowing them to hydrate and obtain room temperature gradually. Both types of blocks were trimmed in length and height to fit the defects, while the AL block grafts were also compressed using surgical pliers to reduce the cancellous portion as much as possible. After compression, AL grafts showed width (thickness) comparable to AT grafts, ranging from 3.5 to 4.5 mm (evaluated with a caliper). Under copious saline solution irrigation, the resident cortical bone was penetrated at the recipient sites with small size round burs to enhance vascularization toward the base of the block grafts. The blocks were then fixed with their cancellous bone side facing resident bone by means of 1.5 mm in Ø × 10 or 12 mm long titanium screws (Neodent, Curitiba, Brazil). The grafts were covered with a collagen membrane (Genius Baumer, São Paulo, Brazil), and the flaps were repositioned and sutured with interrupted nylon 4–0 single sutures for primary intention healing.

Post-surgical infection control included systemic antibiotics (Amoxicillin 500 mg × 3 daily × 7 days) and chlorhexidine digluconate 0.12% mouth rinses for the following 7 days. In addition, non-steroidal anti-inflammatory treatment (Nimesulide 100 mg × 2 daily × 5 days) and analgesics (Acetaminophen 750 mg, according to individual needs) were prescribed. Sutures were removed 14 days after surgery.

Bone biopsies: clinical procedures and evaluation

Implant installation surgery was performed 6 months after the grafting procedure. During this session, one cylindrical biopsy – including the graft and portion of resident bone – was retrieved perpendicularly to the lateral aspect of the augmented ridge by means of a trephine bur (2-mm internal Ø) from each patient (Fig. 1a).

The biopsies were routinely fixed, decalcified, dehydrated, embedded in paraffin, and sectioned. Three 6-μm-thick hematoxylin–eosin-stained sections, representing central aspects of the cylindrical biopsy, were used for histological and histomorphometrical analysis using a DIASTAR light microscope (Leica Reichert & Jung, Wetzlar, Germany) connected to a Leica Microsystems DFC-300-FX digital camera (Leica Microsystems, Wetzlar, Germany). A standardized area of interest (AOI; 2 mm × 6 mm) including the most external (buccal) portion of the biopsy was digitally traced with the computer mouse; thus, the AOI represented 60–75% of grafted bone and 25–40% of resident bone for both groups. The relative amounts (%) of viable bone (VB), necrotic bone (NcB), and soft tissues (ST) within grafted and resident bone in the AOI were planimetrically estimated using Image J (NIH, Bethesda, MD, USA).

Mini-implants: clinical procedures and evaluation

During implant installation surgery, apart from biopsy sampling, a mini-implant (2.0 mm in Ø × 5.5 mm; acid-etched surface – Neodent, Curitiba, Brazil) was also installed in each patient, perpendicularly to the lateral

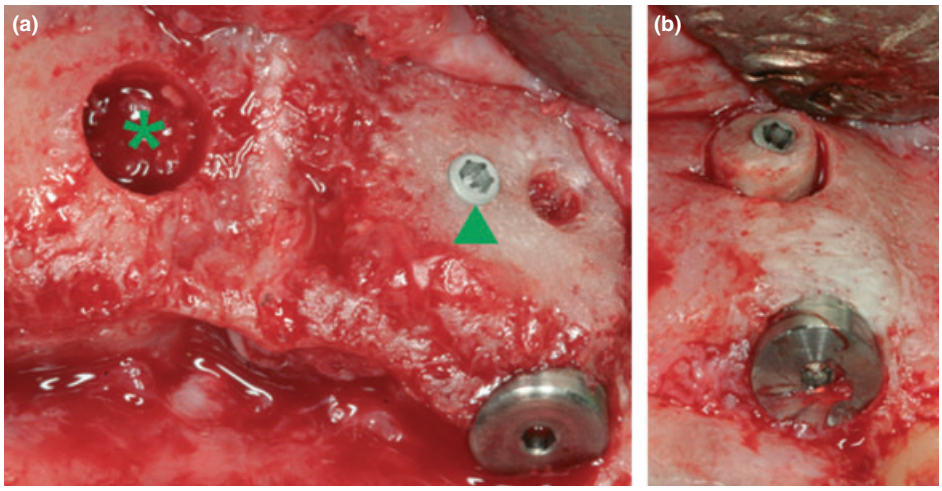

Fig. 1. Case treated with allogeneic bone (AL) grafting. (a) Six months after grafting, a bone biopsy was retrieved from the prepared implant site (*); in addition, a mini-implant was placed perpendicularly to the augmented alveolar ridge (▲). (b) The mini-implant was retrieved 12 months after grafting.

aspect of the augmented alveolar ridge (Figure 1a), unless it was considered they might interfere with the regular implants. The mini-implants, including a small amount of surrounding bone, were retrieved by means of a trephine bur (3 mm internal Ø), during second-stage surgery for healing abutment placement (i.e., 6 months after implant placement for the maxilla and 4 months for the mandible) (Fig. 1b).

The biopsies were routinely processed for undecalcified histology using the cutting-grinding technique (Donath & Breuner 1982), and one 40-μm-thick, central longitudinal section was obtained from each mini-implant and stained with Toluidine blue. In each side of the mini-implant, three equally sized regions (length: 650 μm; area: 3500 μm^2) were traced, representing the "coronal" (R1), "middle" (R2), and "appical" (R3) portions of the mini-implant (corresponding to threads 1–2, 7–8, and 13–14, from the head of the screw, respectively). The amounts (%) of bone area between implant threads (BA) and bone-to-implant contact (BIC) were then planimetrically estimated. The average from the left- and right-side values was used to represent each region; in addition, the average of the three regions was used to represent the entire mini-implant.

Statistical evaluation

The data were described as means, standard deviations, and medians. Normality of the data was confirmed with the Kolgomorov–Smirnov test, and comparisons were made using the *t*-test for non-paired data and ANOVA followed by Tukey post hoc test. Statistical significance was set at $P < 0.05$. GraphPad Prism 5.0 for Windows (GraphPad Software Inc., La Jolla, CA, USA) was used for the statistical evaluation and to draw the graphs.

Results

A flowchart describing the number of included patients, grafted, lost and analyzed blocks, as well as analyzed biopsies and mini-implants, is uploaded as supplementary material (Chart S1).

Clinical observations

In four patients treated with AL grafts, some blocks were lost as a result of intrasurgical complications, that is, improper fixation during placement, which apparently lead to block mobility and lack of incorporation. These block grafts were found loose during the implant surgery session and were removed, and the treatment plan of the patients was modified to fit that. In addition, one of these patients presented with an exposed graft at the 30-day post-op control. The patient was instructed to apply chlorhexidine 1% gel over the exposed area twice daily for 14 days; after this period, the graft was again covered by soft tissue, and no further clinical signs of inflammation were observed. In all other cases/sites, the alveolar ridge was adequately augmented allowing implant installation. All subsequently inserted implants were osseointegrated and prosthetically restored.

Bone biopsies

In several instances, a portion of the cylindrical biopsies broke off either during harvesting or during taking them out from the trephine (approximately 30% of the cases). Care was taken to "reassemble" the cylinder and keep the two pieces together throughout the histotechnical processing, including embedding. In the cylindrical bone biopsies retrieved from the AL group, areas of necrotic bone (NcB), that is, empty osteocyte lacunae and absence of capillaries in the harversian channels, of variable size were consistently observed. In general, the interface between resident bone (i.e., buccal aspect of the alveolar ridge during surgery) and AL block graft was distinct. Nevertheless, within the biopsy, newly formed bone and NcB portions of the graft were clearly distinguishable; newly formed bone rich in osteoblasts was in direct contact with and/or completely surrounded NcB (Figs 2 and 3). There was a tendency to higher amounts of NcB at the aspects of the biopsies distant from resident bed (Fig. 4) and newly formed bone near resident bed. Distinction between resident bone and AT block graft was rather difficult and in some instances impossible. In contrast to the AL group biopsies, it was not easy to distinguish between newly formed bone and graft within the AT group biopsies. Nevertheless, areas of NcB were also observed in bone biopsies retrieved from the AT group. No conspicuous inflammatory cell infiltrate was observed, and only a few osteoclasts could be seen in the sections, from both groups.

The results of the histomorphometric evaluation regarding the cylindrical biopsies are presented in Table 1. Although somehow larger amounts of NcB were found in the AT compared with the AL grafts (55.9 vs. 43.1, respectively), the difference was not statistically significant ($P = 0.19$). In contrast, statistically significant larger amounts of VB (27.6 vs. 8.4; $P = 0.0002$) and less ST (16.4 vs. 48.4; $P \leq 0.0001$) were seen in the sections from the AT group compared to those from the AL group, respectively.

Mini-implants

Twelve patients in the AL and 8 in AT bone block group received a mini-implant. No complications were observed in regard with this part of the study, and all mini-implants were found clinically osseointegrated (i.e., immobile) during the harvesting session (Fig. 1b). A larger variation in terms of BA and BIC was observed in the AL group compared to the AT group (Fig. 5a,b), and quantitative histomorphometric analysis showed larger amounts of BIC regarding the mini-implants placed in AT compared to those in AL, especially at the middle and apical aspects of the mini-implants. However, the differences between AL and AT groups were not statistically significant regarding BIC and BA in the various regions (R1, R2, and R3), or when the entire mini-implant was considered (Table 2).

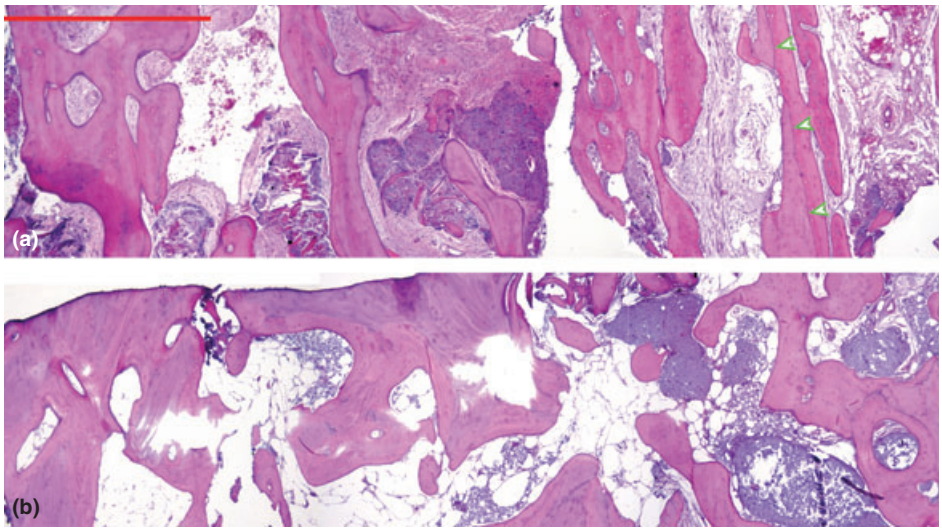

Fig. 2. (a) Histomicrograph of the entire biopsy retrieved from an allogeneic bone (AL)-treated ridge. Green arrowheads indicate the interface between the host (to the right of the image) and the grafted bone. (b) Histomicrograph of the entire biopsy retrieved from an autogenous bone (AT)-treated ridge. The interface between the host (to the right in the image) and the grafted bone is impossible to discern. Hematoxylin–eosin stain; bar = 1 mm.

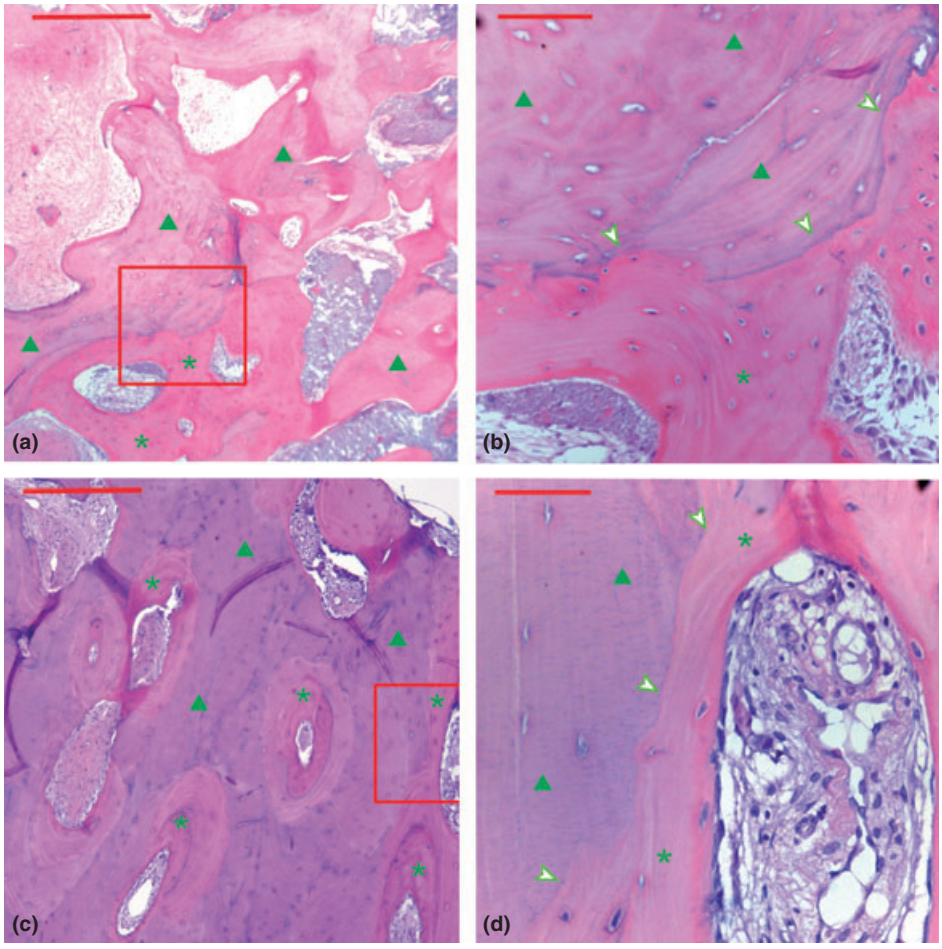

Fig. 3. (a) Histomicrograph from a biopsy retrieved from an allogeneic bone (AL)-treated ridge and (b) higher magnification of the area within the red frame in (a); (c) Histomicrograph from a biopsy retrieved from an autogenous bone (AT)-treated ridge and (b) higher magnification of the area within the red frame corresponding to the rectangle in c. Necrotic bone (▲) in direct contact with viable bone (•), presenting a large numbers of osteocytes is observed. In the majority of cases, the junction between block graft and newly formed bone was distinct (arrowheads). (hematoxylin–eosin stain; bar = 500 μm in a & c and = 100 μm in b & d).

Discussion

The results of this study revealed that 6 months after lateral bone augmentation using fresh-frozen allogenic bone blocks (AL), only limited amounts (8%) of vital bone were present within the augmented tissues, which consisted basically of soft connective tissue and non-vital necrotic bone (NcB). In contrast, sites augmented with autogenous bone blocks (AT) presented approximately three times more vital bone (VB) (28%). In this context, the histomorphometric evaluation herein was performed within a standardized AOI, including the most buccal portion of the biopsy and representing approximately 60–75% of grafted bone and 25–40% of resident bone for both groups. This small variation in grafted/resident bone representation was impossible to avoid due to variation in preoperative block graft dimensions and also due to differences in block graft resorption rates. In the present material, using a CBCT-based analysis, AL resorption rate was on average three times larger than that in the AT group (9.0% vs. 3.0%, respectively) (Spin-Neto et al. 2011b). Thus, it is reasonable to suggest that a relatively larger portion of the AOI in the AL group represented resident bone – compared with the AT group – and thus, the true amount of VB within the AL grafts was in fact less than what estimated. On the other hand, the larger resorption rates in the AL group should be partially attributed to the different microarchitecture in AT and AL blocks, the former being largely cortical while the latter were mostly cortico-cancellous.

The patients in both groups in the present study were similar in regard with the inclusion criteria for treatment, both on the systemic- and site-level, except that patients in the AL group showed inadequate amounts of intraoral bone available for grafting. Treatment allocation in the present study was thus not random. Nevertheless, the only possibility of a confounder between groups would have been if the lack of adequate amounts of intraoral bone available for grafting in the AL group was related to systemic disorders, which as mentioned this was not the case herein.

The findings of the present study are in accordance with the results from older studies showing faster/better incorporation and remodeling of AT compared to various types of allograft bone. Studies using animal and human models have shown inadequate revascularization, little creeping substitution, decreased mineral accretion, and a small

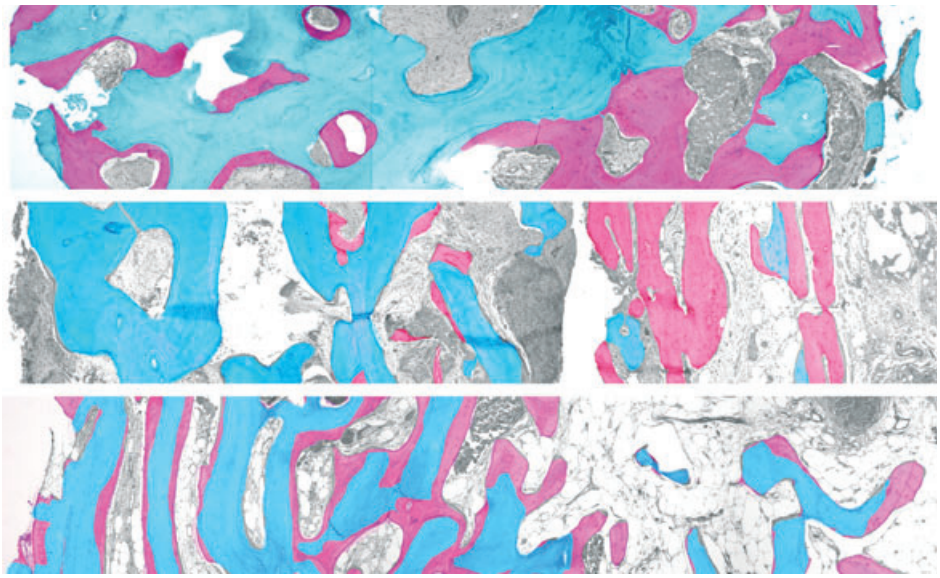

Fig. 4. Histomicrographs of biopsies from three different allogeneic bone (AL)-treated ridges, illustrating the typical variation in healing patterns observed in the present study. The color of the images was digitally replaced to visualize viable bone (in pink) and necrotic bone (in blue). Despite the obvious differences in healing patterns in terms of vital to necrotic bone ratio among the biopsies, there was always a tendency for higher amounts of necrotic bone at the lateral aspects of the biopsies, that is, distant to resident bone (left side of the figure). (hematoxylin–eosin stain).

Table 1. **Means ± standard deviations (medians) of necrotic bone, soft tissues, and viable bone (%) expressed in for both treated groups**

Group	Soft tissues	Necrotic bone	Viable bone
Allogeneic bone (n = 20)	48.4 ± 18.1 (52.8)*	43.1 ± 20.3 (37.1)***	8.4 ± 4.9 (9.1)**
Autogenous bone (n = 14)	16.4 ± 15.6 (11.4)	55.9 ± 27.6 (64.0)	27.6 ± 17.5 (24.5)

*P < 0.001; **P = 0.0002, ***P = 0.19; t-test for non-paired data, reg. the differences between groups.

number of cells involved in the remodeling process of AL grafts, indicating that AL grafts performed histologically and clinically were worse compared with AT grafts (Oklund et al. 1986; Goldberg & Stevenson 1987; Schwarz et al. 1991; Delloye et al. 1992; Spin-Neto et al. 2011a). The reduced incorporation/remodeling of AL grafts observed in the older studies were attributed partially to their antigenicity and microcracks (Stevenson et al. 1991; Kirkeby et al. 1992). The establishment from AATB, however, of strict guidelines for donor bone processing during recent years has minimized, and antigenicity of AL grafts, and in fact AL grafting in the present group of patients, did not seem to challenge relevant components of the immune system significantly, that is, IL-10, IL-1β, IFN- γ, and TNF-α serum values were within physiological levels up to 6 months post-grafting (Spin-Neto et al. 2012). Besides that, there is evidence of osteoblast-related vital cells escaping the freezing process during AL preparation (Simpson et al. 2007); these cells could survive the suggested AATB protocol and might be involved in specific immune responses due to the presence of anti-HLA-specific antibodies against AL grafts, which could have an adverse effect on the graft's incorporation and increase the incidence of rejection (VandeVord et al. 2005). Thus, the potential for risk of disease transmission from bone allografts seems existent, and the absence of any reports on cross-contamination during recent years should be largely attributed to the improved precision in donor screening and to the advances in tissue processing, disinfection, and sterilization methods (Hinsenkamp et al. 2012). This in turn underscores the necessity of using accredited sources for bone allografts.

It may be suggested that if a longer healing time was used herein, larger amounts of VB might have been observed in the AL group. Indeed, in a recent case series report of similarly performed maxillary alveolar ridge augmentations, a gradual decrease in the amount of necrotic bone (NcB) from 6 to 9 months post-operatively was observed (61% to 41%, respectively) (Acocella et al. 2012). Nevertheless, complete incorporation/remodeling throughout an AL graft has yet to be reported, and low long-term success rates of oral implants inserted in AL augmented jaws have been reported (Carinci et al. 2010). Those authors observed increased marginal peri-implant bone loss after 4 years post-op, leading to a success rate of only 40%, in contrast to the success rates reported for implant placed in AT augmented ridges (in general >90%) (Chiapasco et al. 2009). This observation could be explained by the results of the histomorphometric analysis in the cylindrical biopsies herein, showing that the major portion of VB was confined relatively close to the resident bone, and VB was only sporadically seen toward the buccal aspects of the AL augmented sites. Thus, some portion of the marginal peri-implant bone in this study might have consisted by necrotic bone. It is reasonable to expect that necrotic bone would more readily develop microcracks due to occlusal loading compared to living bone and therefore would also resorb more readily than living bone, because no potential for microcrack repair can be expected in necrotic bone. Thus, the high survival rates of oral implants placed in AL grafts reported in recent publications in short-term follow-up (Accetturi et al. 2002; Barone et al. 2009; Carinci et al. 2009a,b), which seem similar to those observed with AT grafts (Chiapasco et al. 2009), should be considered with caution. In this context, it may be suggested that if a shorter healing time has been used, a larger failure rate in block incorporation and/or implant osseointegration might have been observed in the AL group; however, 6 months is an average healing time often used in bone augmentation procedures (Chiapasco et al. 2009).

In this study, the amount of NcB in the AT group was 56% of the AOI. Previously published studies (Zerbo et al. 2003; Burger et al. 2011) have reported smaller amounts of NcB in AT biopsies compared with herein. For example, Zerbo et al. (2003) found 11% (range: 1–34%) and Burger et al. (2011) only 1.5% (range: 0.5–8.9%) of NcB in trephine biopsies taken after similarly performed lateral ridge augmentation. In contrast, in another recent study where biopsies were harvested in a similar manner as in the present study, large amounts of NcB (average 58%; range 34–81%) were also observed (Acocella et al. 2010). The differences among the studies can be due to different methods for estimating the amount of NcB, but might also be due to variations regarding the site of biopsy harvesting and the cortico-cancelous block graft architecture. As already mentioned, the revascularization and incorporation of

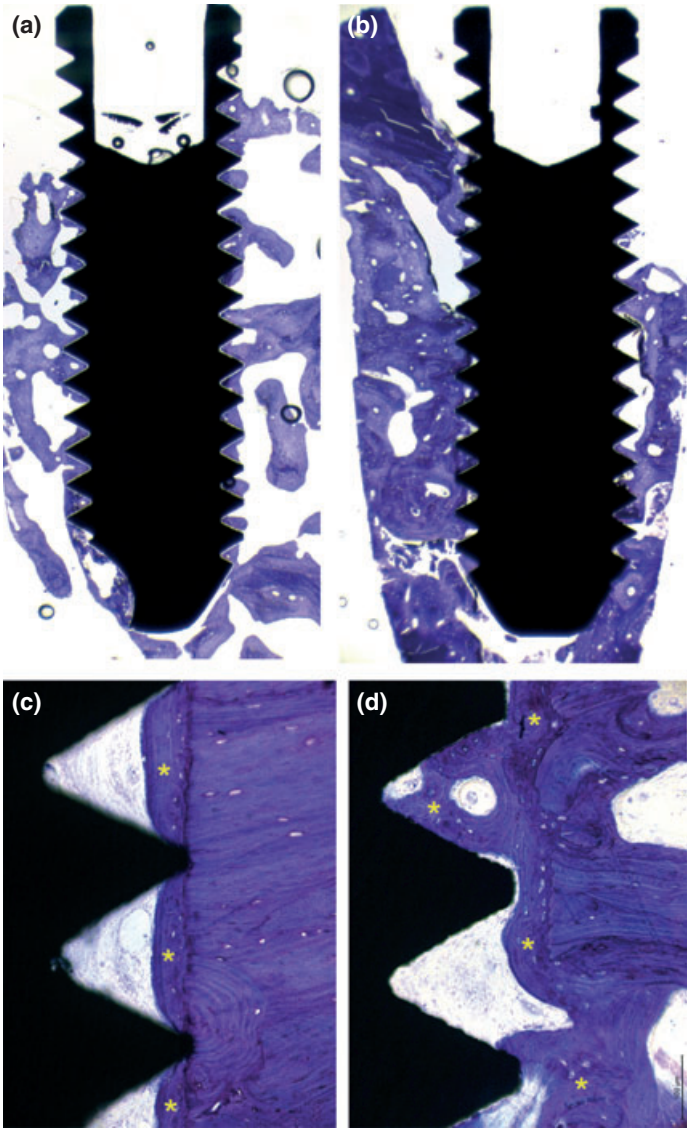

Fig. 5. Histomicrograph of entire mini-implant biopsies retrieved from an allogeneic bone (AL)-treated (a) and an autogenous bone (AT)-treated (b) ridge. New bone formation (bone tissue between the treads) and extensive bone-to-implant contact (BIC) is observed irrespective treatment group. (c, d). Histomicrographs of the middle region (R2) of two mini-implant biopsies retrieved from the AL-treated group. In (c), newly formed vital bone (*) has formed as a relatively thin layer on top of the bone block, and BIC can only be seen at the pitches of the mini-implants, while in (d) bone formation (*) and BIC extends at the thread area of the implant (Toluidine blue stain).

cancellous AT bone blocks is faster compared with cortical AT bone blocks (Goldberg & Stevenson 1993; Chen et al. 1994). In the present study, similarly to the Acocella et al. (2010) study, the AT blocks consisted primarily or exclusively of cortical bone and the biopsies were intentionally harvested from the external aspect of the augmented alveolar ridge, that is, the major portion of the biopsies consisted of the block graft itself. In contrast, in the other evaluated studies (Zerbo et al. 2003; Burger et al. 2011), no information is given regarding the relative cortico-cancellous composition of the blocks, and the biopsies were harvested through implant site preparation; it is possible that part of the entire length of the biopsy in those studies consisted from resident (vital) bone.

So far, no information regarding osseointegration of implants in areas previously grafted with fresh-frozen bone allografts is present in the literature. In the present study, mini-implants placed in AL or AT blocks showed no statistical significant differences in BA, that is, similar amounts of osseointegration. The mini-implants used herein had a size (length) similar to the bone block thickness. Apparently the titanium implant has exerted an osteoconductive effect allowing new bone formation to occur along the entire length of this relatively short implant. Obviously, no extrapolations can be made on whether the same would occur if clinical-size implants were placed entirely surrounded by AL bone. In this context, based on the observations in the cylindrical biopsies and the biopsies including the mini-implants from the AL group in the present study, where the larger amounts of VB (i.e., graft incorporation) were found close to the resident bone, it seems reasonable to suggest that the appropriateness of AL bone blocks as a graft material may depend on the amount of lateral augmentation required.

Thus, AL bone block grafts may be an option in cases where a limited amount of augmentation is needed, and the future implant can be expected confined within the inner (medial) aspect of the bone block, which shows large amounts of VB. The clinical impact of the poorer graft incorporation observed for the AL grafts compared with AT on the long-term performance of oral implants placed in AL grafts remains obscure, and no firm assumptions can be made on the basis of the available literature in terms of survival or rate of biological complications (i.e., peri-implantitis). The results, however, from a recent preclinical *in vivo* study suggest that peri-implantitis may progress faster when the peri-implant tissues contain non-vital bone biomaterials comparing with cases where implants are installed in pristine sites (Stavropoulos et al. 2012).

Table 2. **Means ± standard deviations of bone-to-implant contact (BIC) and bone area between the threads (BA) in three distinct regions ("coronal" – R1, "middle" – R2 and "appical" – R3) and for the whole mini-implant, in the allogeneic bone (AL) (*n* = 12) and autogenous bone (AT) (*n* = 8) groups**

	R1		R2		R3		Mini-implant	
Parameter	AL	AT	AL	AT	AL	AT	AL	AT
BIC (%)	33.6 ± 20.8 (34.1)	37.8 ± 28.9 (44.3)	41.9 ± 32.2 (50.0)	59.3 ± 24.4 (57.6)	42.0 ± 31.6 (48.0)	57.7 ± 36.1 (66.4)	39.2 ± 14.3 (38.1)	44.4 ± 24.4 (47.1)
BA (%)	39.0 ± 27.3 (37.3)	46.5 ± 26.0 (58.2)	39.2 ± 32.1 (46.6)	58.7 ± 22.4 (57.2)	45.9 ± 33.3 (53.0)	51.9 ± 36.2 (58.5)	41.4 ± 15.6 (39.7)	43.5 ± 24.3 (42.0)

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. CONSORT 2010 checklist of information to include when reporting a randomised trial.

Chart S1. Flow-chart describing the number of included patients, grafted, lost and analyzed blocks, as well as analyzed biopsies and mini-implants is uploaded as supplementary material.