J Clin Periodontol 2010; 37: 1029–1038 doi: 10.1111/j.1600-051X.2010.01621.x

Journal of Clinical Periodontology

Simultaneous *versus* two-stage implant placement and guided bone regeneration in the canine: histomorphometry at 8 and 16 months

Zvi Artzi[1], Carlos E. Nemcovsky[1], Haim Tal[1], Evgeny Weinberg[1], Miron Weinreb[2], Hari Prasad[3], Michael D. Rohrer[3] and Avital Kozlovsky[1]

[1]Department of Periodontology, School of Dental Medicine, Tel Aviv University, Tel Aviv, Israel; [2]Department of Oral Biology, School of Dental Medicine, Tel Aviv University, Tel Aviv, Israel; [3]Division of Oral Pathoolgy, School of Dentistry, University of Minnesota, Minneapolis, MN, USA

Artzi Z, Nemcovsky CE, Tal H Weinberg E, Weinreb M, Prasad H, Rohrer MD, Kozlovsky A. Simultaneous versus two-stage implant placement and guided bone regeneration in the canine: histomorphometry at 8 and 16 months. J Clin Periodontol 2010; 37: 1029–1038. doi: 10.1111/j.1600-051X.2010.01621.x.

Abstract

Aim: To compare the effect of timing of implant placement and guided bone regeneration (GBR) procedure on osseointegration and newly formed bone at 8 and 16 months.

Material and Methods: In seven dogs, four different sites were bilaterally established: (1) an implant placed in a 6-month healed (6m-GBR) bovine bone mineral (BBM) grafted site; (2) a simultaneously placed implant with the grafted BBM (Si-GBR) followed by a membrane coverage; (3) an implant placed in a membrane-protected non-grafted defect; and (4) an implant placement in a naturally healed site (Cont). Histomorphometry was obtained at 8 and 16 months post-implant placement. Bone–implant contact (BIC), crestal bone resorption (CBR), vertical intra-bony (VIB) defect, bone (BAF) and particle (PAF) area fractions, and osteoconductivity (CON) levels were measured.

Results: In all sites, BIC ranged between 62% and 79% with no significant differences. PAF ranged from 17% to 27%, with no effect of time. At 8 and 16 months, BAF was significantly smaller at the Si-GBR site when compared with all other sites, CON was significantly greater at the 6m-GBR site, and CBR and VIB were significantly smaller at the 6m-GBR when compared with the Si-GBR sites.

Conclusions: The simultaneous and delayed techniques both showed a similar osseointegration level over time. However, the staged approach showed enhanced newly formed bone, higher osteoconduction around the grafted mineral, less CBR, and smaller vertical bone defect over time compared with the combined approach.

Key words: bone augmentation; bone–implant contact; bovine bone; guided bone regeneration; osseointegration; osteoconduction

Accepted for publication 11 August 2010

Conflict of interest and sources of funding statement

The authors declare no conflict of interest at any part of the study.

This study was funded by the Osteology Foundation and by the Gerald A. Niznick Chair of Implant Dentistry and the Alpha Omega Research Fund, Tel Aviv University. The authors are indebted to Biomet 3i for providing the implants; to Geistlich Biomaterials for providing the biomaterials to Dr. Naam Kariv, Mr. Haim Barki, and Mr. Neria Davidov for the animal care; to Mrs. Ilana Gelernter for the statistical analysis; and to Rita Lazar (EASE) for editorial assistance.

Guided bone regeneration (GBR) is a well-established and documented procedure (Dahlin et al. 1988, 1989, Nyman et al. 1990, Buser et al. 1993, 1995, Schenk et al. 1994, Simion 2003). This procedure may be carried out before, concurrent with, and/or after implant placement. Bone augmentation using GBR principles is a reliable technique (McAllister & Haghighat 2007, Donos et al. 2008, Tonetti & Hämmerle 2008). Implants placed in regenerated bone achieved successful osseointegration and proper function over a 5-year period (Buser et al. 1996, Fugazzotto 1997, Corrente et al. 2000, Blanco et al. 2005, Juodzbalys et al. 2007, Dahlin et al. 2009) comparable to implants placed in native bone (Buser et al. 2002). Early implant placement concurrent with GBR technique has shown encouraging results (Donos et al. 2008, Buser et al. 2009). Osseointegration represents a dynamic process, both during its establishment and maintenance (Berglundh et al. 2003). Implants placed in an autogenous onlay bone graft, simultaneously or in a delayed mode, enhance integration and stability in the latter (Lundgren et al. 1999, Rasmusson et al. 1999).

In a systematic review, which focuses on the survival of implants placed in augmented and non-augmented sites, no clear evidence is shown that the simultaneous GBR procedure affects the implant survival rate (Donos et al. 2008). This review also emphasizes that there are no prospective studies that compare the staged approach to pristine sites. The authors conclude that the implant survival rate in augmented (staged or simultaneously) and non-augmented sites is similar. However, there is no study that compares the timing of the augmentation procedure, particularly on the histological level.

When the consensus reports of Donos et al. (2008) and Tonetti and Hämmerle (2008) are considered, there are insufficient data regarding the influence that timing of the augmentation procedure may have on the quality of the regenerated peri-implant osseous tissue in the augmented site, despite the solid data on the outcome of implant survival and long-term function (Fugazzotto 1997, Buser et al. 2002, Donos et al. 2008). Therefore, it is of utmost interest to qualitatively and quantitatively study the degree of regeneration and osseointegration over time in bovine bone mineral (BBM) grafted sites.

The aim of this study was to explore and examine the efficacy of implant placement and GBR procedure whether performed simultaneously or as a two-stage approach, and to compare the influence of timing of the augmentation procedure on the level of osseointegration at 8 and 16 months post-implant placement.

Material and Methods

The study was conducted on seven young male beagle dogs, weighing between 15.6 and 19.3 kg (average 17.6 kg). The Tel Aviv University Institutional Committee for Animal Care and Use approved the study. All surgical procedures were performed under general anaesthesia, achieved by pre-sedation with 1.5 cm^3 (20 mg) 2% xylazine base IM, followed by an i.v. injection of ketamine (Clorketamin® 1000, Vetoquinol, Fort Worth, TX, USA), 5 mg/kg+xylazine base (XYL – M 2, Veterinary), 1 mg/kg. Buccal and lingual local infiltration of 2% lidocaine hydrochloride with norepinephrine (1:100000) on the vestibular mandibular area was administered for haemostasis and to reduce post-operative pain.

Study design and time flow chart

The time line is illustrated in Fig. 1. To reduce and minimize animal morbidity, the protocol time line was designed accordingly. Therefore, the post-implant placement/augmentation phases were 8 months apart. At day 0, P4 and M1 were extracted and allowed to heal naturally. At 1 month, the first augmentation procedure occurred at the previous extracted area. At 5 months, P2 and P3 were extracted to establish an edentulous span of approximately 60 mm (including the previous extracted P4 and M1 area). At 6 months, implant placement ($n = 4$) and additional GBR experimental sites were carried out. The exposure phase occurred at 6 months post-implant placement followed by the hygienic phase. An identical protocol was repeated on the contralateral side, however, at a delay of 8 months. Consequently, the hygienic phases lasted for 2 and 10 months in the respective mandibular sides.

Surgical protocol

Phase 1: 6-month augmentation procedure

After soft tissue healing, 1-month post-extraction of P4 and M1, an edentulous area of approximately 33–35 mm mesio-distally was established. A rectangular four-wall intra-bony defect (11 mm $L \times 5$ mm $W \times 7$ mm D) was then created (Berglundh & Lindhe 1997) using a motorized copious irrigated round bur (Fig. 2a). Boundaries were maintained at least 6–7 mm distally from P3 due to future defect location in surgery phase 2. The defect was filled with BBM particles (Bio-Oss®, Geistlich Biomaterials, Wolhusen, Switzerland) (Fig. 2b) and covered by a native collagen membrane (Bio-Gide®, Geistlich Biomaterials). A coronally advanced flap was established to secure primary soft tissue closure using a 4–0 polyamide monofilament non-absorbable suture (Ethilon®, Ethicon®, Johnson & Johnson, Somerville, NJ, USA). Post-operatively, surgical sites were swabbed every 48–72 h with 0.2% chlorhexidine. Antibiotics were administered for the first 10 days post-surgery.

Phase 2: Implant placement phase concurrent with additional GBR procedures

At 6 months, 1-month post-extraction of P2 and P3 (mesio-distal span of approximately 20 mm), two additional rectangular intra-bony defects (11 mm $L \times 5$ mm $W \times 7$ mm D each) were created, partly

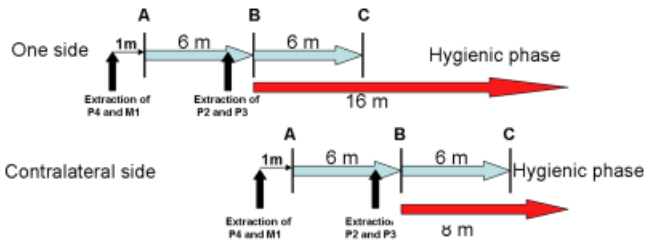

Fig. 1. Time flow chart of the sequence of surgical phases in both sides of the two observation periods: A, first augmentation phase; B, implant placement and second augmentation phase; C, exposure phase (m, months).

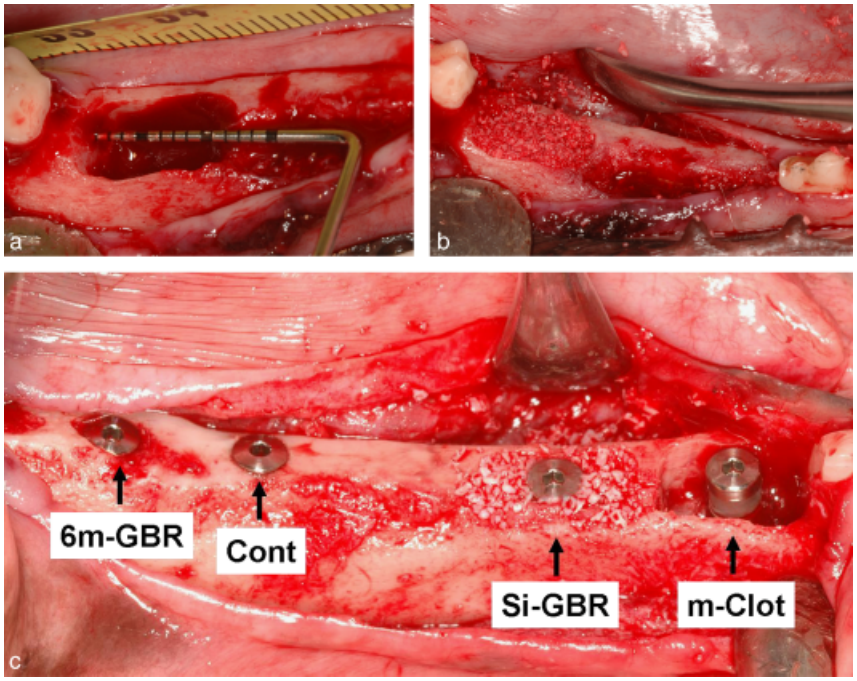

Fig. 2. (a) The first surgical phase comprised of a rectangular four-wall intra-bony defect (11 mm L × 5 mm W × 7 mm D). (b) The intra-bony defect was completely filled with bovine bone mineral (BBM) particles. (c) Four implants were placed: one of the fresh created defects was filled with BBM particles (Si-GBR), one was left to clot, one at the 6-month previously augmented with BBM (6m-GBR) and one at a pristine bone (Cont).

on the previous mesial aspect of P4. Care was taken to preserve at least 2 mm of bone septa between the experimental defects. Then, four dual acid-etched surface implants (Full Osseotite®, Biomet 3i, Palm Beach Gardens, FL, USA) 3.25 mm D × 10 mm L were placed: one in the middle of the 6-month-old augmented site (phase 1 surgery), two in the middle of the currently created surgical defects, and one in a pristine healed site. In the current rectangular defects, the implants were not connected to either the buccal or lingual bony walls and initial stability was obtained in the floor of the defects by their 3 mm apical threads. All implant necks were placed at the level of the osseous crestal rim, whether at the current created defects or flush with the housing bone.

Inter-implant distance was at least 10 mm apart. One of the current defects was filled with BBM particles while the other remained ungrafted and naturally filled with blood. Both defects were covered by a collagen membrane followed by obtaining primary soft tissue closure as described during the first augmentation procedure.

Consequently, four different sites were established: (1) a combined implant placement and bone augmentation – a simultaneous GBR site (Si-GBR); (2) an immediate implant placement in a non-grafted defect, spontaneously filled with blood and covered by a membrane (m-Clot); (3) a delayed implant placement in a 6-month BBM grafted GBR site (6m-GBR); and (4) an implant placed in a naturally healed site (Cont) (Fig. 2c).

Phase 3: exposure phase

At 6 months post-implant placement (12 months after the first augmentation), implants were exposed using an incisional flap technique. Implant healing screws (4 mm high) were connected followed by soft tissue cuffing using interrupted sutures. Healing screws protruded approximately 2 mm above the surrounding soft tissue.

All phases of the surgical protocol, with identical timing of augmentation and implant placement procedures, were sham-operated on the contralateral side of the mandible, but with a delay of 8 months.

Hygienic phase

Implant superstructure healing screws were maintained under pre-medication only, followed immediately by meticulous cleaning using a toothbrush and 0.2% chlorhexidine digluconate (GlaxoSmithKline, Brentford, Middlesex, UK) antiseptic solution, every 48–72 h.

Autopsy procedure

At 8 and 16 months after implant placement in the right and left sides, respectively, dogs were euthanized by a lethal dose (30 mg/kg) of pentobarbitone sodium i.v. (CTS, Pharmaceutical Industries Inc., Kiryat Mala'achi, Israel). Subsequently, 300 ml of 10% neutral buffered formalin was injected under pressure into the external carotid arteries to achieve optimal tissue fixation (Karnovsky 1965) before specimen block removal. Radiographs of the specimen blocks were taken before histological processing. Mandibular block sections that contained the implants were then prepared for hard tissue non-decalcified preparation.

Histological processing

Blocks were placed in 10% neutral buffered formalin and dehydrated with graded series of alcohols for 9 days. After dehydration, specimens were infiltrated with a light-polymerized embedded resin (Technovit 7200, VLC, Heraeus Kulzer, Hanau, Germany). After 20 days of infiltration with constant shaking at a normal atmospheric pressure, specimens were embedded and polymerized by 450 nm light at 40°C, and prepared using the cutting/grinding method described by Donath and Breuner (1982).

Initially, bucco-lingual cuts at the mid inter-implant distance between fixtures provided tissue blocks that contained a single implant in each to comprise 5.0 mm of hard calcified tissue at the mesial and distal aspects of each implant. Each block was sectioned longitudinally, providing an antero-posterior section of the implant, presenting the mesial and distal aspects of the tissue to the implant interface.

Specimens were cut to a thickness of 150 μm on an EXAKT cutting/grinding system (EXAKT Technologies, Oklahoma City, OK, USA). Slides were polished to a thickness of 40 μ using the EXAKT microgrinding system, followed by alumina polishing paste and stained with Stevenel's blue and Van Gieson's picro fuchsin. Photomicrographs were obtained using a Zeiss Axiolab photomicroscope (Carl Zeiss Microimaging, Thornwood, NJ, USA).

Histomorphometry analysis

All experimental sites were measured by the same investigators (Z. A. and E. W.), without knowing the identification of the sites.

Histomorphometry was conducted on a screen monitor, attached to the microscope (magnification × 35), and performed only in the crestal/implant neck region and not at the total implant length (Fig 3a). The peri-implant area was analysed along a 5.5 mm stretch of the implant, starting from the neck, at a final magnification of × 35. A 1 mm *W* × 5 mm *L* region of interest (ROI) was overlaid on the section adjacent to the implant body, as well as 1 mm away from the implant (Fig. 3b), mesially and distally. The Bioquant Nova Prime System (Bioquant Image Analysis Corp, Nashville, TN, USA) was used to calculate the morphometric measurements for the following parameters: (a) direct bone-to-implant contact (BIC); (b) crestal bone resorption (CBR) – distance between the implant neck and first BIC; (c) vertical intra-bony defect height (VIB) – distance between the crestal marginal bony peak and first BIC; (d) bone area fraction at the proximity (BAF-P) of the implant surface (0-1 mm from the implant surface) and at the distance (BAF-D) – (1–2 mm beyond the implant surface); (e) particle area fraction at the proximity (PAF-P) and at the distance (PAF-D); and (f) osteoconductivity level – which represents the amount of direct contact of the encircling newly formed bone around the grafted particles at the proximity (CON-P) and at the distance (CON-D) zones (Fig. 3b). In each implant, the average of the mesial and distal aspects was calculated for the different parameters. Thus, the inspected area was related to the mesial and distal aspect of the implants, which are augmented zones except for the control group.

Statistical analysis

Each of the four different experimental groups was examined at two different time points, post-implant placement. Means and standard deviations (SD) were calculated for each measured parameter. ANOVA with repeated measures analysed the differences between mean values, two-within time factors (8 and 16 months) and three-within subject factors (treatment modality, proximal, or distant location and time). Statistical significance was recognized at $P \leq 0.05$. A two-tailed Pearson's correlation analysis was applied to test possible associations between the different parameters at each experimental site.

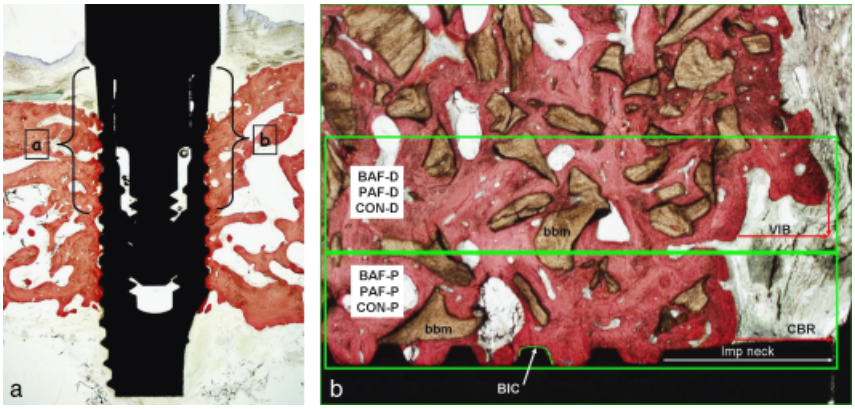

Fig. 3. (a) An osseointegrated implant at 8 months. Histomorphometry was conducted on the surrounding tissues of the coronal 5 mm from the implant neck (a, b). (Stevenel's blue and Van Gieson's picro fuchsin staining × 15 original magnification). (b) Parameters measured in the proximal-P (0–1 mm) and the distance-D (1–2 mm) from the implant surface: bone–implant contact (BIC), crestal bone resorption (CBR), vertical intra-bony (VIB) defect, bone and particle (BBM) area fractions (BAF and PAF, respectively) and the direct particle-bone contact, i.e., conductivity level (CON). (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification).

Results

At the post-surgical phases, soft tissue healing was immaculate and animals showed no signs of mastication distress. One dog developed swelling in the floor of the mouth, which was alleviated by an additional course of antibiotics. The experimental learning curves of previous studies (Artzi et al. 2003a, b, 2004, 2006, Kozlovsky et al. 2007) ensured predictable uncomplicated wound healing. The establishment of buccal and lingual coronally advancing flaps, which obtained a resilient full soft tissue closure with the horizontal internal mattress suturing, ensured no spontaneous exposure of the GBR procedures.

Morphometric evaluations

Tables 1–4 summarize all percentage and metric parameters, as well as the ranges and averages with related SD. Figs 4a–7b represent the mesio-distal non-decalcified section cuts of Si-GBR, 6m-GBR, m-Clot and Cont sites, at 8 and 16 months, respectively.

BIC

At 8 months, the mean direct BIC at the different experimental sites ranged from 62.1% to 77%. At 16 months, the mean BIC ranged from 78% to 79.8%, respectively. At the grafted sites (Si-GBR and 6 m-GBR), BIC significantly increased ($P = 0.003$) from 8 to 16 months. When Si-GBR was compared with Cont sites, BIC was significantly higher at the Cont sites ($P = 0.017$). At 16 months, BIC was comparable between all different sites.

BAF

Remodelled bone was measured in the proximity (BAF-P) of the implant body at 0–1 mm and at the distance (BAF-D) of 1–2 mm and beyond.

At 8 months, the mean BAF-P ranged from 49.5% to 83.7% and BAF-D ranged from 37% to 70.3%, at the different sites. At 16 months, the mean BAF-P ranged from 55% to 77.6% and BAF-D ranged from 43.34% to 67.6%, at the respective sites.

BAF-P was significantly greater ($P = 0.033$) at 6m-GBR compared with the Si-GBR in both periods, but not over time. BAF-P was also greater when Cont was compared with Si-GBR and to m-Clot ($P = 0.019$), and between Cont and 6m-GBR groups at both periods in the latter ($P = 0.003$).

At the distance, BAF-D was significantly greater ($P = 0.022$) at the 6m-GBR over Si-GBR, and timing (8 and 16 months) had a significant effect ($P = 0.003$). BAF-D was also greater at the m-Clot sites compared with Si-GBR ($P = 0.046$), but not when compared with the 6 m-GBR sites.

Table 1. Morphometric measurements (ranges and averages) of bone–implant-contact (BIC) (%), crestal bone resorption (CBR), and vertical intrabony (VIB) defect at 8 months observation period

Sites/parameters	Si-GBR	6m-GBR	m-Clot	Cont
BIC (%)	$62.07^{a} \pm 8.44$ SD (range: 50.74–72.63)	75.97 ± 10.14 (63.20–94.08)	73.94 ± 17.71 (49.30–94.23)	$76.97^{a} \pm 9.35$ (65.98–87.68)
CBR (mm)	$2.34^{bc} \pm 0.42$ (range: 1.79 –2.91)	$1.47^{b} \pm 0.57$ (0.90–2.26)	1.76 ± 1.44 (0.47–3.33)	$1.10^{c} \pm 0.26$ (0.72–1.51)
VIB defect (mm)	$1.89^{cd} \pm 0.72$ (0.91–2.93)	$0.81^{c} \pm 0.34$ (0.38–1.28)	$1.62^{e} \pm 1.39$ (0.47–3.70)	$0.65^{de} \pm 0.35$ (0.22–1.13)

Superscript letters indicate $P < 0.05$.
GBR, guided bone regeneration.

Table 2. Morphometric measurements (ranges and averages) of bone and grafted particle area fractions (%) and the conductivity level (%) at the proximal and distant zones at 8 months observation period

Sites/parameters	Si-GBR	6m-GBR	m-Clot	Cont
Bone area fraction – proximal (BAF-P) (%)	$49.49^{abceh} \pm 15.12$ (20.16–67.41)	$63.52^{adfi} \pm 6.89$ (50.24–70.86)	$62.81^{ejn} \pm 12.91$ (44.00–83.23)	$83.65^{fhijo} \pm 12.21$ (59.10–95.52)
Bone area fraction – distance (BAF-D) (%)	$36.95^{bcgk} \pm 12.51$ (20.64–51.60)	$52.94^{bdl} \pm 9.40$ (38.34–64.7)	$52.52^{gmn} \pm 20.08$ (21.67–86.25)	$70.30^{klmo} \pm 13.23$ (51.45–81.01)
Particle area fraction – proximal (PAF-P) (%)	22.42 ± 5.16 (11.9–27.08)	20.38 ± 4.75 (15.88–29.03)	NA	NA
Particle area fraction – distance (PAF-D) (%)	26.91 ± 8.31 (19.20–43.61)	21.94 ± 10.93 (10.37–35.57)	NA	NA
Conductivity level – proximal (CON-P) (%)	$37.71^{p} \pm 24.31$ (7.95–80.62)	$71.42^{p} \pm 18.29$ (43.79–91.21)	NA	NA
Conductivity level – distance (CON-D) (%)	$45.85^{q} \pm 22.49$ (14.79–77.37)	$53.94^{q} \pm 23.44$ (15.05– 87.65)	NA	NA

Superscript letters indicate $P < 0.05$.
GBR, guided bone regeneration.

Table 3. Morphometric measurements (ranges and averages) of bone–implant-contact (BIC) (%), crestal bone resorption (CBR), and vertical intrabony (VIB) defect at 16 months observation period

Sites/parameters	Si-GBR	6m-GBR	m-Clot	Cont
BIC (%)	77.95 ± 11.24 SD (range: 61.40–94.80)	79.82 ± 7.54 (66.12–86.68)	75.93 ± 20.77 (52.45–100)	79.47 ± 6.87 (70.98–88.42)
CBR (mm)	$1.11^{a} \pm 0.26$ (0.80–1.47)	$0.92^{a} \pm 0.33$ (0.65–1.05)	1.24 ± 0.45 (0.62–1.78)	1.20 ± 0.35 (0.75–1.64)
VIB defect (mm)	$0.88^{bc} \pm 0.43$ (0.33–1.34)	$0.50^{b} \pm 0.37$ (0.12–1.05)	0.76 ± 0.18 (0–1.10)	$0.66^{c} \pm 0.33$ (0.34–1.19)

Superscript letters indicate $P < 0.05$.
GBR, guided bone regeneration.

As well, Cont sites were significantly greater when compared with Si-GBR ($P = 0.009$) and to 6m-GBR sites ($P = 0.004$). However, timing of observation had no effect.

The grafted sites showed a significant difference between BAF-P and BAF-D ($P = 0.009$) within the three subject factors (site, time, and different areas). This difference was also shown when Si-GBR was compared with m-Clot ($P = 0.003$), 6 m-GBR to Cont ($P = 0.003$), and when m-Clot was compared with Cont ($P < 0.001$). However, this difference was neither site nor time-dependent.

PAF

Similar to BAF, the PAF was divided and measured at the 0–1 mm proximity (PAF-P) and beyond at a 1–2 mm distance (PAF-D). At the grafted sites, the mean PAF-P ranged from 20.4% to 22.4% and from 17.3% to 21.9% at 8 and 16 months, respectively. Mean PAF-D ranged from 21.9% to 26.9% and from 22.8% to 23.2%, at the respective periods. No difference was found in relation to the site type or timing of observation. However, when the examined areas were compared within each site, there was a significant difference between PAF-P and PAF-D ($P = 0.013$), which was independent of site and timing of observation.

Conductivity level (CON)

The CON level was estimated by the amount of direct contact of the particles to the surrounding bone at the proximal (CON-P) and distant (CON-D) areas. Mean CON-P ranged from 37.7% to 71.4% and from 80% to 85% at 8 and 16 months, respectively. Mean CON-D ranged from 45.9% to 53.9% and from 76.4% to 80.9%, at the respective periods. At the proximal area, a greater level of conductivity was shown at the 6m-GBR sites ($P = 0.024$), as well as a significant increase at 16 months ($P = 0.024$). This difference was closely dependent but not statistically significant ($P = 0.06$) on the applied type of site.

Beyond the proximal area, at 1–2 mm distance, CON-D increased significantly at the 16-month period ($P = 0.01$). However, no difference was found between the Si-GBR and 6m-GBR sites.

Table 4. Morphometric measurements (ranges and averages) of bone and grafted particle area fractions (%) and the conductivity level (%) at the proximal and distant zones at 16 months observation period

Sites/parameters	Si-GBR	6m-GBR	m-Clot	Cont
Bone area fraction – proximal (BAF-P) (%)	$55.04^{aceh} \pm 5.60$ (46.74–63.99)	$63.42^{adf} \pm 9.41$ (52.80–78.86)	$70.01^{e} \pm 18.32$ (47.46–94.69)	$77.62^{fh} \pm 12.28$ (58.62–89.12)
Bone area fraction – distance (BAF-D) (%)	$43.35^{bcij} \pm 14.16$ (23.33–63.96),	$58.73^{bdg} \pm 12.98$ (33.09–76.19)	$66.81^{i} \pm 14.14$ (49.33–85.97)	$67.55^{gj} \pm 14.85$ (37.84–82.25)
Particle area fraction – proximal (PAF-P) (%)	17.29 ± 11.24 (3.85–33.83)	21.93 ± 5.40 (14.25–29.81)	NA	NA
Particle area fraction – distance (PAF-D) (%)	23.16 ± 13.71 (2.59–39.92)	22.84 ± 4.52 (17.40–29.14)	NA	NA
Conductivity level – proximal (CON-P) (%)	80.01 ± 17.40 (50.97–99.09)	84.97 ± 13.56 (65.95–97.02)	NA	NA
Conductivity level – distance (CON-D) (%)	76.44 ± 20.57 (38.85–99.56)	80.93 ± 12.70 (63.62–95.43)	NA	NA

Superscript letters indicate $P < 0.05$.
GBR, guided bone regeneration.

CBR and VIB defect height

Mean CBR ranged from 1.10 to 2.34 mm and from 0.92 to 1.24 mm at 8 and 16 months, respectively. CBR was statistically significantly smaller ($P = 0.032$) at the 6m-GBR, compared with the Si-GBR sites in both observation periods. This difference was also shown between the Si-GBR and Cont sites ($P = 0.023$). A significant difference in favour of the extended time observation (16 months) was found in all sites ($P < 0.001$) except for the Cont sites.

The mean VIB defect height ranged from 0.65 to 1.89 mm at 8 months and from 0.50 to 0.88 mm at 16 months. There was a significant difference between 6m-GBR and Si-GBR sites ($P = 0.014$), as well as a significant improvement (smaller VIB) with time ($P = 0.003$). VIB was significantly greater at the Si-GBR group when compared with the Cont group within each observation period ($P = 0.023$), as well as during time ($P = 0.01$). In addition, VIB at the Cont group was smaller compared with the m-Clot group ($P = 0.034$), but not in time.

The two-tailed Pearson's analysis (Table 5) shows that all sites were correlated between CBR and VIB at the 8-month observation period. Also, a negative correlation ($R = -0.77$; $P = 0.04$) between the increasing CBR and the decreasing BIC at the 6m-GBR sites at 16-month observation, is noteworthy.

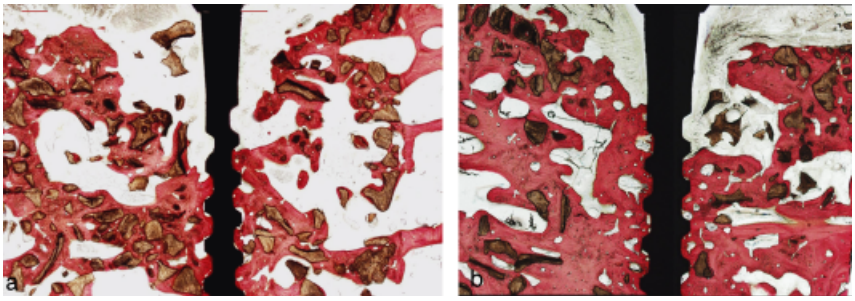

Fig. 4. (a) At 8 months, a coronal part of a simultaneous implant placement and bone augmentation procedure using bovine bone mineral particles. Note the crestal bone level in reference to the implant neck. (The implant core was trimmed due to lack of interest and to allow an expanded view at the periphery). (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification). (b) At 16 months, a coronal part of a simultaneous implant placement and bone augmentation procedure. Note the improved crestal bone level (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification).

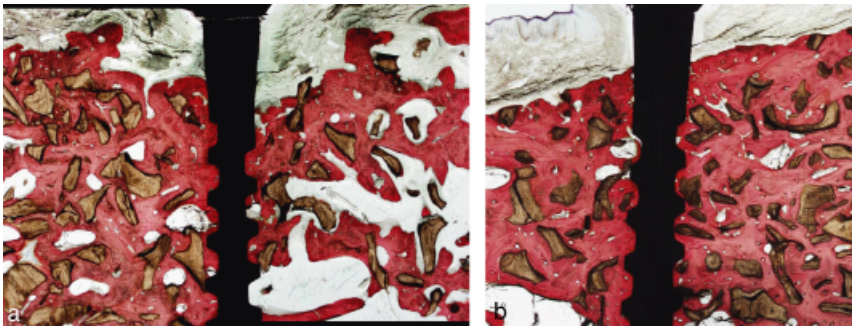

Fig. 5. (a) A coronal part of an implant placed at a 6-month regenerated grafted bovine bone mineral site (6m-GBR) at 8 months (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification). (b) A coronal part of a 6m-GBR site at 16 months (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification).

Discussion

Numerous case series (Buser et al. 1996, 2009, Fugazzotto 1997, Blanco et al. 2005, Juodzbalys et al. 2007, Dahlin et al. 2009) and systematic data (Donos et al. 2008) report the clinical efficacy on the survival/success rate of implants placed in augmented and non-augmented sites. In this study, an interpretation of a possible distinct observation between combined implant placement and bone augmentation, or in a two-staged approach event, was explored histologically. Implant placement and alveolar GBR procedure, whether performed simultaneously or in a delayed approach showed comparable osseointegration as measured histologically by direct BIC. However, timing of the augmentation

procedure showed an impact on several peri-implant parameters.

Similar observations were made whether a simultaneous implant placement or a delayed two-stage approach occurred. The degree of osseointegration as measured by the amount of BIC along the implant surface at the augmented sites (Si-GBR and 6m-GBR) was comparable to the non-grafted sites (m-Clot and Cont) particularly at 16 months.

The resemblance between the outcome of the membrane-protected grafted and ungrafted (m-Clot) sites may also be attributed to the high regenerative potential of the 4-wall intra-bony experimental surgical site. The fact that the surgical sites were well protected by a membrane and the clot was stabilized by the surrounding walls may have a role in enhancing healing in both surgical models (Haney et al. 1993).

At 8 months, BIC at the delayed approach was higher than at the combined mode – 76% *versus* 62%. However, significant validity could not be established because of the relatively small sample size. The pristine bone (Control) group was the only group to present a statistically significant higher BIC over the Si-GBR group. In a short-term study (Veis et al. 2007), a lower BIC was shown in a membrane-protected defect compared with an implant placed in a native bone when experimental defects were placed in the iliac bone of three dogs. In the current study, all sites exhibited an equal amount of osseointegration at 16 months. The increase of BIC over time indicates that there is a continual remodelling and osseous adaptation around the implant body, regardless of the type of surgical modality performed. Both, the simultaneous and delayed augmentation techniques demonstrated a high grade of osseointegration.

Furthermore, although BIC at the Si-GBR sites was inferior at 8 months, it increased to an equal level compared with other sites at 16 months. This may indicate that the prolonged presence of the grafted particles with osteoconductive property could present a certain influence on the increasing BIC level.

At all surgical sites, grafted or non-grafted, BAF-P showed a greater percentage of mineralized tissue over BAF-D. It can be assumed that the properties of the rough implant surface (Klokkevold et al. 1997, 2001) might play a role as an osteoconductor vehicle to attract surrounding cells to enhance bone apposition. In the current study, the contribution of the implant surface characteristics was not explored. Regardless of the grafting augmentation timing, the mineralized BAF occupied more than one-half of the total area at the augmented sites. However, the delayed approach showed additive superiority. The amount of bone formation was significantly greater at the 6m-GBR sites, both at 1 and 2 mm beyond the implant surface at both observation periods. When simultaneous implant placement and augmentation were compared with the delayed

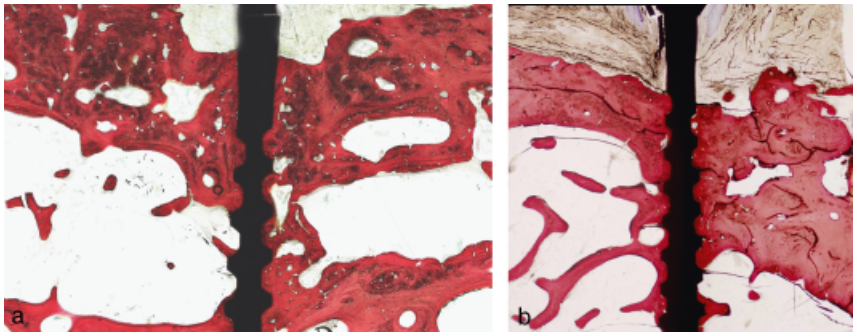

Fig. 6. (a) A coronal part of an implant placed at a clotted non-grafted membrane-protected site (m-Clot) at 8 months (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification). (b) A coronal part of an implant placed at the m-Clot site at 16 months observation (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification).

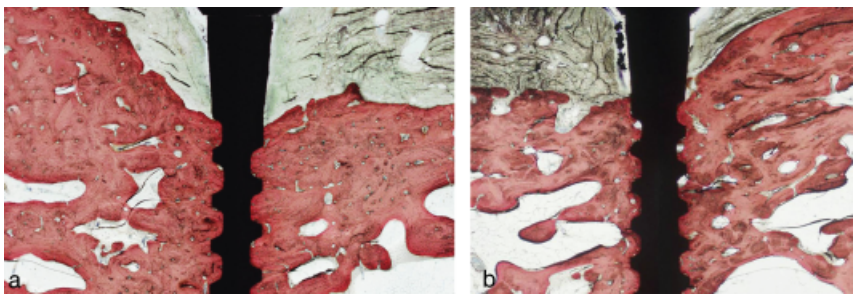

Fig. 7. (a) A coronal part of an implant placed at a healed pristine (Cont) site at 8 months observation (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification). (b) A coronal part of an implant of the Cont group at 16 months observation (Stevenel's blue and Van Gieson's picro fuchsin staining × 35 original magnification).

Table 5. Significant correlations between the different parameters at the different sites (two-tailed Pearson's analysis)

Parameters/period	Si-GBR	6m-GBR	m-Clot	Cont
BIC/BAF-P/8 months			$R = 0.84$; $P = 0.03$	
BIC/CBR/16 months		$R = 0.77$; $P = 0.04$		
BAF-P/BAF-D/16 months			$R = 0.88$; $P = 0.009$	
BAF-P/PAF-P/8 months		$R = -0.98$; $P < 0.001$		
BAF-P/PAF-P/16 months	$R = 0.85$; $P = 0.015$			
CON-P/CON-D/16 months	$R = 0.92$; $P = 0.004$	$R = 0.77$; $P = 0.04$		
CBR/VIB/8 months	$R = 0.95$; $P = 0.03$		$R = 0.99$; $P < 0.001$	$R = 0.82$; $P = 0.02$

BIC, bone–implant-contact; BAF-D, bone area fraction – distance; BAF-P, bone area fraction – proximal; CBR, crestal bone resorption; VIB, vertical intra-bony defect; PAF-P, particle area fraction at the proximity; CON-D, conductivity level – proximal; CON-P, conductivity level – distance; GBR, guided bone regeneration.

technique, it was suggested that timing had a significant effect on the degree of ossification in the distant zone (BAF-D). Apparently, there is continuous remodelling and bone apposition. Therefore, implants placed in 6-month old augmented sites (6m-GBR group) present advantages over fresh ones (Si-GBR), which are expressed by higher BAF values. However, at the 6m-GBR group, BAF-P was similar at both observation periods, probably due to the extended remodelling time of the grafting procedure, which was examined in an extended matured stage at 14 and 22 months, respectively.

Significant differences were also shown on the CON level where the grafted particle osteoconduction was significantly higher at the 6m-GBR when compared with the Si-GBR sites. This difference was particularly noticed at 8 months and reduced at 16 months. The implication is that the delayed approach (6m-GBR) showed a higher osteconductivity at least at the phase of bone remodelling and during the establishment of osseointegration properties. Significant improvement at both the Si-GBR and 6m-GBR sites was evident with time. However, despite the differences, time and the applied timing of the grafting procedure were independent. The fact that bone apposition increased over time, regardless of the type of site, indicated that the grafted particles continuously attracted osteogenic activity whether grafted before or at the implant placement phase. This material promotes osteoblastic differentiation and matrix synthesis (Tapety et al. 2004). The resemblance of the particle configuration of BBM (Rosen et al. 2002) and its slow resorption pattern (Artzi et al. 2004) probably contribute to its higher conductivity in the long term. It can be assumed that the macro- and microporous configuration of BBM particles result in better osteoconductive properties (Spector 1994, 1999, Jensen et al. 1996, Hämmerle et al. 1997, 1998, Skoglund et al. 1997, Artzi et al. 2000, 2001, Norton et al. 2003), as well as in experimental defects around implants (Hockers et al. 1999, Polyzois et al. 2007), and in a recent comparative human study in socket sites (Lee et al. 2009).

The percentage of PAF that occupied the augmented site, whether grafting occurred before or simultaneously with implant placement, ranged from 20% to 25% in both observation periods at the proximal body and away from the implant body. This has been well supported in previous studies (Artzi et al. 2004, 2005, Meijndert et al. 2005) when BBM maintained its volume fraction after a limited early phase of resorption, independent from continual increase of newlyformed bone. Consequently, it is self-evident that except for the early observation at 6m-GBR, no correlation was found between BAF and PAF.

One of the most striking findings revealed at the implant neck level was the CBR and VIB defect, which were distinguishable among the different surgical sites. When implant placement and ridge augmentation were performed separately, both CBR and VIB were significantly smaller than the resorption of the combined technique. This observation was evident at both 8 and 16 months.

Furthermore, at 8 months, a significant correlation was shown between these two measured parameters at all surgical types except for 6m-GBR. It is noteworthy that in the delayed technique (6m-GBR), a lower CBR level statistically correlated with the increasing percentage of osseointegration (BIC).

Apparently, this critical site of the implant neck/grafted particles and overlay membrane results differently in the remodelled osseous level, embracing the neck when the implant was placed in a remodelled matured ridge compared with the level of crestal bone when implant placement and augmentation occurred concurrently. However, it is clear that under proper maintenance, CBR and VIB improved and reduced with time, as shown at 16 months, regardless of the type of the surgical site.

When BBM was used as the grafting biomaterial in human studies (Hämmerle et al. 2008, Iezzi et al. 2008, Meijndert et al. 2008, Dahlin et al. 2009), bone formation was supported, there was excellent osseointegration, and the crestal bone level was maintained.

Clinical implications

(1) Within the limitations of this study, it is noteworthy that over time, no clinical difference has been shown between implant placement simultaneously with augmentation or via a two-stage approach. Both showed a resembling and maintainable osseointegration.
(2) Using the present experimental model, bone formation was greater at the proximity of the implant surface. The delayed surgical approach was superior compared with the combined technique and also over time.
(3) In accordance to this model, the level of osteoconduction associated with the grafted particles was higher at the staged approach.
(4) The staged approach shows less CBR and smaller vertical bone defect over time when compared with the combined approach; both models show, however, significant improvement at 16 months.

References

Artzi, Z., Givol, N., Rohrer, M. D., Nemcovsky, C. E., Prasad, H. S. & Tal, H. (2003a) Qualitative and quantitative expression of bovine bone mineral in experimental bone defects. Part 1: description of a dog model and histological observations. *Journal of Periodontology* **74**, 1143–1152.

Artzi, Z., Givol, N., Rohrer, M. D., Nemcovsky, C. E., Prasad, H. S. & Tal, H. (2003b) Qualitative and quantitative expression of bovine bone mineral in experimental bone defects. Part 2: morphometric analysis. *Journal of Periodontology* **74**, 1153–1160.

Artzi, Z., Kozlovsky, A., Nemcovsky, C. E. & Weinreb, M. (2005) The amount of newly formed bone in sinus grafting procedures depends on tissue depth as well as the type and residual amount of the grafted material. *Journal of Clinical Periodontology* **32**, 193–199.

Artzi, Z., Tal, H. & Dayan, D. (2000) Porous bovine bone mineral in healing of human extraction sockets. Part 1: histomorphometric evaluations at 9 months. *Journal of Periodontology* **71**, 1015–1023.

Artzi, Z., Tal, H. & Dayan, D. (2001) Porous bovine bone mineral in healing of human extraction sockets. Part 2: histochemical observations at 9 months. *Journal of Periodontology* **72**, 152–159.

Artzi, Z., Weinreb, M., Givol, N., Rohrer, M. D., Nemcovsky, C. E., Prasad, H. S. & Tal, H. (2004) Biomaterial resorption rate and healing site morphology of inorganic bovine bone and beta-tricalcium phosphate in the canine: a 24-month longitudinal histologic study and morphometric analysis. *International Journal of Oral & Maxillofacial Implants* **19**, 357–368.

Artzi, Z., Weinreb, M., Tal, H., Nemcovsky, C. E., Rohrer, M. D., Prasad, H. S. & Kozlovsky, A. (2006) Experimental intrabony and periodontal defects treated with natural mineral combined with a synthetic cell-binding Peptide in the canine: morphometric evaluations. *Journal of Periodontology* **77**, 1658–1664.

Berglundh, T., Abrahamsson, I., Lang, N. P. & Lindhe, J. (2003) De novo alveolar bone formation adjacent to endosseous implants. *Clinical Oral Implants Research* **14**, 251–262.

Berglundh, T. & Lindhe, J. (1997) Healing around implants placed in bone defects treated with Bio-Oss. An experimental study in the dog. *Clinical Oral Implants Research* **8**, 117–124.

Blanco, J., Alonso, A. & Sanz, M. (2005) Long-term results and survival rate of implants treated with

guided bone regeneration: a 5-year case series prospective study. *Clinical Oral Implants Research* **16**, 294–301.

Buser, D., Dula, K., Belser, U., Hirt, H. P. & Berthold, H. (1993) Localized ridge augmentation using guided bone regeneration. 1. Surgical procedure in the maxilla. *International Journal of Periodontics and Restorative Dentistry* **13**, 29–45.

Buser, D., Dula, K., Belser, U. C., Hirt, H. P. & Berthold, H. (1995) Localized ridge augmentation using guided bone regeneration. II. Surgical procedure in the mandible. *International Journal of Periodontics and Restorative Dentistry* **15**, 10–29.

Buser, D., Dula, K., Lang, N. P. & Nyman, S. (1996) Long-term stability of osseointegrated implants in bone regenerated with the membrane technique. 5-year results of a prospective study with 12 implants. *Clinical Oral Implants Research* **7**, 175–183.

Buser, D., Halbritter, S., Hart, C., Bornstein, M. M., Grütter, L., Chappuis, V. & Belser, U. C. (2009) Early implant placement with simultaneous guided bone regeneration following single-tooth extraction in the esthetic zone: 12-month results of a prospective study with 20 consecutive patients. *Journal of Periodontology* **80**, 152–162.

Buser, D., Ingimarsson, S., Dula, K., Lussi, A., Hirt, H. P. & Belser, U. C. (2002) Long-term stability of osseointegrated implants in augmented bone: a 5-year prospective study in partially edentulous patients. *International Journal of Periodontics and Restorative Dentistry* **22**, 109–117.

Corrente, G., Abundo, R., Cardaropoli, D., Cardaropoli, G. & Martuscelli, G. (2000) Long-term evaluation of osseointegrated implants in regenerated and non-regenerated bone. *International Journal of Periodontics and Restorative Dentistry* **20**, 390–397.

Dahlin, C., Linde, A., Gottlow, J. & Nyman, S. (1988) Healing of bone defects by guided tissue regeneration. *Plastic and Reconstructive Surgery* **81**, 672–676.

Dahlin, C., Sennerby, L., Lekholm, U., Linde, A. & Nyman, S. (1989) Generation of new bone around titanium implants using a membrane technique: an experimental study in rabbits. *International Journal of Oral & Maxillofacial Implants* **4**, 19–25.

Dahlin, C., Simion, M. & Hatano, N. (2009) Long-term follow-up on soft and hard tissue levels following guided bone regeneration treatment in combination with a xenogeneic filling material: a 5-year prospective clinical study. *Clinical Implant Dentistry and Related Research* doi: 10.1111/j.1708-8208.2009.00163.x.

Donath, K. & Breuner, G. (1982) A method for the study of undecalcified bone and teeth with attached soft tissues. The Sage-Schliff (sawing and grinding) technique. *Journal Oral Pathology* **11**, 318–326.

Donos, N., Mardas, N. & Chadha, V. (2008) Clinical outcomes of implants following lateral bone augmentation: systematic assessment of available options (barrier membranes, bone grafts, split osteotomy). *Journal of Clinical Periodontology* **35** (Suppl.), 173–202.

Fugazzotto, P. A. (1997) Success and failure rates of osseointegrated implants in function in regenerated bone for 6 to 51 months: a preliminary report. *International Journal of Oral & Maxillofacial Implants* **12**, 17–24.

Haney, J. M., Nilvéus, R. E., McMillan, P. J. & Wikesjö, U. M. (1993) Periodontal repair in dogs: expanded polytetrafluoroethylene barrier membranes support wound stabilization and enhance bone regeneration. *Journal of Periodontology* **64**, 883–890.

Hämmerle, C. H., Chiantella, G. C., Karring, T. & Lang, N. P. (1998) The effect of a deproteinized bovine bone mineral on bone regeneration around titanium dental implants. *Clinical Oral Implants Research* **9**, 151–162.

Hämmerle, C. H., Jung, R. E., Yaman, D. & Lang, N. P. (2008) Ridge augmentation by applying bioresorbable membranes and deproteinized bovine bone mineral: a report of twelve consecutive cases. *Clinical Oral Implants Research* **19**, 19–25.

Hämmerle, C. H. F., Olah, A. J., Schmid, J., Fluckiger, L., Gogolewski, S., Winkler, J. R. & Lang, N. P. (1997) The biological effect of deproteinized bovine bone on bone neoformation on the rabbit skull. *Clinical Oral Implants Research* **8**, 198–207.

Hockers, T., Abensur, D., Valentini, P., Legrand, R. & Hammerle, C. H. (1999) The combined use of bioresorbable membranes and xenografts or autografts in the treatment of bone defects around implants. A study in beagle dogs. *Clinical Oral Implants Research* **10**, 487–498.

Iezzi, G., Scarano, A., Mangano, C., Cirotti, B. & Piattelli, A. (2008) Histologic results from a human implant retrieved due to fracture 5 years after insertion in a sinus augmented with anorganic bovine bone. *Journal of Periodontology* **79**, 192–198.

Jensen, S. S., Aaboe, M., Pinholt, E. M., Hjørting-Hansen, E., Melsen, F. & Ruyter, I. E. (1996) Tissue reaction and material characteristics of four bone substitutes. *International Journal of Oral & Maxillofacial Implants* **11**, 55–66.

Juodzbalys, G., Raustia, A. M. & Kubilius, R. (2007) A 5-year follow-up study on one-stage implants inserted concomitantly with localized alveolar ridge augmentation. *Journal of Oral Rehabilitation* **34**, 781–789.

Karnovsky, M. J. (1965) A formal dehydeglutaroldehyde fixture of high osmolarity for use in electron microscopy. *Journal of Cell Biology* **3**, 112–119.

Klokkevold, P. R., Johnson, P., Dadgostari, S., Caputo, A., Davies, J. E. & Nishimura, R. D. (2001) Early endosseous integration enhanced by dual acid etching of titanium: a torque removal study in the rabbit. *Clinical Oral Implants Research* **12**, 350–357.

Klokkevold, P. R., Nishimura, R. D., Adachi, M. & Caputo, A. (1997) Osseointegration enhanced by chemical etching of the titanium surface. A torque removal study in the rabbit. *Clinical Oral Implants Research* **8**, 442–447.

Kozlovsky, A., Tal, H., Laufer, B. Z., Leshem, R., Rohrer, M. D., Weinreb, M. & Artzi Z (2007) Impact of implant overloading on the peri-implant bone in inflamed and non-inflamed peri-implant mucosa. *Clinical Oral Implants Research* **18**, 601–610.

Lee, D. W., Pi, S. H., Lee, S. K. & Kim, E. C. (2009) Comparative histomorphometric analysis of extraction sockets healing implanted with bovine xenografts, irradiated cancellous allografts, and solvent-dehydrated allografts in humans. *International Journal of Oral & Maxillofacial Implants* **24**, 609–615.

Lundgren, S., Rasmusson, L., Sjostrom, M. & Sennerby, L. (1999) Simultaneous or delayed placement of titanium implants in free autogenous iliac bone grafts. Histological analysis of the bone graft-titanium interface in 10 consecutive patients. *International Journal of Oral & Maxillofacial Surgery* **28**, 31–37.

McAllister, B. S. & Haghighat, K. (2007) Bone augmentation techniques. *Journal of Periodontology* **78**, 377–396.

Meijndert, L., Raghoebar, G. M., Meijer, H. J. & Vissink, A. (2008) Clinical and radiographic characteristics of single-tooth replacements preceded by local ridge augmentation: a prospective randomized clinical trial. *Clinical Oral Implants Research* **19**, 1295–1303.

Meijndert, L., Raghoebar, G. M., Schüpbach, P., Meijer, H. J. & Vissink, A. (2005) Bone quality at the implant site after reconstruction of a local defect of the maxillary anterior ridge with chin bone or deproteinised cancellous bovine bone. *International Journal of Oral & Maxillofacial Surgery* **34**, 877–884.

Norton, M. R., Odell, E. W., Thompson, I. D. & Cook, R. J. (2003) Efficacy of bovine bone mineral for alveolar augmentation: a human histologic study. *Clinical Oral Implants Research* **14**, 775–783.

Nyman, S., Lang, N. P., Buser, D. & Bragger U (1990) Bone regeneration adjacent to titanium dental implants using guided tissue regeneration: a report of two cases. *International Journal of Oral & Maxillofacial Implants* **5**, 9–14.

Polyzois, I., Renvert, S., Bosshardt, D. D., Lang, N. P. & Claffey, N. (2007) Effect of Bio-Oss on osseointegration of dental implants surrounded by circumferential bone defects of different dimensions: an experimental study in the dog. *Clinical Oral Implants Research* **18**, 304–310.

Rasmusson, L., Meredith, N., Cho, I. H. & Sennerby, L. (1999) The influence of simultaneous versus delayed placement on the stability of titanium implants in onlay bone grafts. A histologic and biomechanic study in the rabbit. *International Journal of Oral & Maxillofacial Surgery* **28**, 224–231.

Rosen, V. B., Hobbs, L. W. & Spector, M. (2002) The ultrastructure of anorganic bovine bone and selected synthetic hydroxyapatites used as bone graft substitute materials. *Biomaterials* **23**, 921–928.

Schenk, R. K., Buser, D., Hardwick, W. R. & Dahlin, C. (1994) Healing pattern of bone regeneration in membrane-protected defects: a histologic study in the canine mandible. *International Journal of Oral & Maxillofacial Implants* **9**, 13–29.

Simion, M. (2003) Procedures used to augment the deficient alveolar ridge. In: Lindhe, J., Karring, T. & Lang, N. P. (eds). *Clinical Periodontology and Implant Dentistry*, pp. 897–914. Oxford, UK: Blackwell Munksgaard Publication.

Skoglund, A., Hising, P. & Young, C. (1997) A clinical and histologic examination in humans of the osseous response to implanted natural bone mineral. *International Journal of Oral & Maxillofacial Implants* **12**, 194–199.

Spector, M. (1994) Anorganic bovine bone and ceramic analogs of bone mineral as implants to facilitate bone regeneration. *Clinics in Plastic Surgery* **21**, 437–444.

Spector, M. (1999) Basic principles of tissue engineering. In: Lynch, S. E., Genco, R. J. & Marx, R. E. (eds). *Tissue Engineering*, pp. 3–16. Carol Stream, IL: Quintessence Publishing Co.

Tapety, F. I., Amizuka, N., Uoshima, K., Nomura, S. & Maeda T (2004) A histological evaluation of the involvement of Bio-Oss in osteoblastic differentiation and matrix synthesis. *Clinical Oral Implants Research* **15**, 315–324.

Tonetti, M. S. & Hämmerle, C. H. (2008) Advances in bone augmentation to enable dental implant placement: consensus report of the sixth European workshop on periodontology. *Journal of Clincal Periodontology* **35** (Suppl.), 168–172.

Veis, A. A., Papadimitriou, S., Trisi, P., Tsirlis, A. T., Parissis, N. A. & Kenealy, J. N. (2007) Osseointegration of Osseotite and machined-surfaced titanium implants in membrane-covered critical-sized defects: a histologic and histometric study in dogs. *Clinical Oral Implants Research* **18**, 153–160.

Address:
Zvi Artzi
Department of Periodontology
School of Dental Medicine
Tel Aviv University
Ramat Aviv
Tel Aviv 69978
Israel
E-mail: zviartzi@tau.ac.il

Clinical Relevance

Scientific rationale for the study: The timing of implant placement and bone augmentation as a combined or a two-stage event is of clinical importance.
Principal findings: Over time, a similar amount of osseointegration was established using both techniques. Newly formed bone was enhanced proximal to the rough surface of the implant. The staged approach showed a higher degree of newly formed bone and osteoconduction, less CBR, and smaller vertical bone defect over time when compared with the combined simultaneous technique.
Practical implications: Although the staged approach showed enhanced bone level and higher bone density, timing of the augmentation procedure did not influence the degree of osseointegration or the clinical outcome.