Published in final edited form as:

J Clin Periodontol. 2013 April ; 40(4): 387–395. doi:10.1111/jcpe.12065.

Determinants of alveolar ridge preservation differ by anatomic location

Binnaz Leblebicioglu¹, Mabel Salas^{1,*}, Yirae Ort¹, Ashley Johnson², Vedat O. Yildiz³, Do-Gyoon Kim², Sudha Agarwal⁴, and Dimitris N. Tatakis¹

¹Division of Periodontology, College of Dentistry, The Ohio State University, Ohio, USA

²Division of Orthodontics, College of Dentistry, The Ohio State University, Ohio, USA

³Center for Biostatistics, The Ohio State University, Ohio, USA

⁴Division of Oral Biology, College of Dentistry, The Ohio State University, Ohio, USA

Abstract

Aim—To investigate and compare outcomes following alveolar ridge preservation (ARP) in posterior maxilla and mandible.

Methods—Twenty-four patients (54 ± 3 years) with single posterior tooth extraction were included. ARP was performed with freeze-dried bone allograft and collagen membrane. Clinical parameters were recorded at extraction and re-entry. Harvested bone cores were analysed by microcomputed tomography (micro-CT), histomorphometry and immunohistochemistry.

Results—In both jaws, ARP prevented ridge height loss, but ridge width was significantly reduced by approximately 2.5 mm. Healing time, initial clinical attachment loss and amount of keratinized tissue at extraction site were identified as determinants of ridge height outcome. Buccal plate thickness and tooth root length were identified as determinants of ridge width outcome. In addition, initial ridge width was positively correlated with ridge width loss. Micro-CT revealed greater mineralization per unit volume in new bone compared with existing bone in mandible ($p < 0.001$). Distributions of residual graft, new cellular bone and immature tissue were similar in both jaws.

Conclusion—Within the limitations of this study, the results indicate that in different anatomic locations different factors may determine ARP outcomes. Further studies are needed to better understand determinants of ARP outcomes.

Keywords

alveolar bone; extraction; outcome; regeneration; wound healing

Address: Binnaz Leblebicioglu, Division of Periodontology, College of Dentistry, The Ohio State University, 305 West 12th Ave, Columbus OH 43210, USA, leblebicioglu.1@osu.edu.

*Current address: Department of Periodontology, School of Dentistry, University of Missouri-Kansas City

Conflict of interest

Authors report no conflict of interest.

The clinical and histological aspects of socket healing have been reported (Boyne 1966, Trombelli et al. 2008; Van der Weijden 2009, Moya-Villaescusa & Sanchez-Perez 2010, Ten Heggeler et al. 2011). Alveolar bone resorption post-extraction varies among individuals and sites; ridge height loss is greater in mandibular than maxillary sites and ridge width loss is greater on buccal aspects (Smukler et al. 1999, Iasella et al. 2003). Post-extraction ridge resorption leads to loss of function, inadequate bone for dental implants and prosthodontic difficulties (Bartee 2001). The need to avoid such complications has spurred significant interest in developing techniques and biomaterials to prevent ridge resorption. Alveolar ridge preservation (ARP) is a guided bone regeneration (GBR) procedure aiming to control post-extraction ridge resorption.

Current ARP protocols include bone autografts, allografts, xenografts, alloplasts and membranes of diverse origin (Nemcovsky & Serfaty 1996, Bartee 2001, Iasella et al. 2003, Fickl et al. 2008a,b). Despite numerous options, outcome data are scarce (Ten Heggeler et al. 2011). The limited evidence available suggests that mineralized freeze-dried bone allograft (FDBA) combined with collagen membrane may offer the best height preservation (Iasella et al. 2003, Ten Heggeler et al. 2011). No ARP protocol effectively prevents ridge width changes (Ten Heggeler et al. 2011). To the best of our knowledge, there are no studies providing insights regarding the factors, anatomical or other, which determine ARP outcomes. Therefore, the purpose of this study was to investigate and compare, in maxillary and mandibular sites, the clinical and histological outcomes of ARP, in an effort to identify treatment outcome determinants for these different anatomical locations.

Materials and Methods

Study design

The study was a prospective observational trial. Patients planned for tooth extraction and placement of single dental implant at maxillary or mandibular posterior sextants were examined before tooth extraction, at time of extraction and at time of implant placement and clinical parameters were recorded. Bone core samples were obtained at implant placement and used to assess bone healing by means of microcomputed tomography (micro-CT), histomorphometric and immunohistochemical analysis. The Institutional Review Board of The Ohio State University (OSU) approved the study protocol, data collection forms and informed consent form.

Study population

Patients were recruited among those attending the Graduate Periodontology Clinic at OSU. Eligibility criteria were single bounded posterior tooth (molar or premolar with adjacent teeth present) scheduled for extraction and implant placement; adult (aged ≥ 18 years); systemically and periodontally healthy; non-smoker; able and willing to comply with study procedures and visits. Exit criteria were voluntary withdrawal; non-compliance with study procedures and visits; development of systemic or oral diseases; required surgical protocol modification (e.g. indication for delayed GBR procedure or immediate implant placement). All study subjects provided written consent.

Clinical parameters

Subjects received complete periodontal examination. Probing depth (PD), gingival recession (GR) and gingival (GI) and plaque (PI) indices were recorded for the respective quadrant prior to tooth extraction. Clinical attachment level (CAL) was calculated. PD, GR and CAL were obtained (six sites/tooth) for the tooth assigned for extraction and the two adjacent teeth (mesial and distal). Keratinized tissue (KT) width (mucogingival junction to gingival margin) was determined on the mid-buccal aspect of the target tooth. Parameters were recorded to nearest millimeter. Clinical measurements were performed by a trained examiner, using UNC-15 probe. Examiner reliability was assessed in pilot study; intra-examiner differences for PD, GR and KG, as well as for various ridge measurements, were within 1 mm.

An alginate impression was obtained pre-extraction and stone cast was produced to prepare a template (stent). The stent was fabricated with standardized 3'' aluminium tubes integrated into a 0.06'' thermo-forming material, and was used both for standardization of clinical measurements after extraction (Figure 1) and for guidance during implant placement.

Ridge dimensions were recorded immediately following extraction and prior to implant placement (Fig. 1). Relative ridge height (stent occlusal plate to alveolar crest) was measured on buccal plate at three different positions: mesial, middle and distal aspects of extraction site. Buccolingual socket size (residual defect size), alveolar ridge width (buccal bone plate to lingual bone plate) and buccal and lingual bone plate thickness were measured at center of target site. Site center was determined as half the distance between mesial and distal teeth. UNC-15 probe was used for residual defect size measurement, while a calliper placed 1 mm apical to crest was used for remaining measurements. Buccolingual root diameter at cemento-enamel junction (CEJ) and root length (CEJ to longest root's tip) were determined post extraction using a calliper.

Surgical protocol

Surgical procedures were performed by residents of the Advanced Periodontics Training Program at OSU, under direct supervision by periodontal faculty, according to routine surgical protocol. Following local anaesthesia and mucoperiosteal flap reflection (due to defect size, need to stabilize membrane), atraumatic extraction and socket debridement (curettes, saline irrigation) were performed. ARP indication was confirmed based on site characteristics (site unsuitable for immediate implant placement; immediate implant indication consisted of four-wall residual defect (intact walls) allowing intimate implant–bone contact on 3 walls, and 3 mm of alveolar bone height available apically, to obtain primary stability). Flaps were released, as needed, to achieve primary closure whenever possible. Then, FDBA (DENTSPLY Tulsa Dental, Tulsa, OK, USA) and collagen membrane (Biomend Extend, Zimmer Dental., Carlsbad, CA, USA) were placed and flaps sutured. Membrane exposure, if any, was measured in buccolingual direction with UNC-15 probe. Patients were given prescriptions for antibiotics (7 days) and 0.12% chlorhexidine rinse (twice daily for 2 weeks). At the end of the healing period, mucoperiosteal flaps were again reflected to perform implant placement.

At re-entry time (Table 1), using the original measurement stent to determine implant location, an indentation mark (round bur) was made on the crest to both mark osteotomy site and to retain bone core sample orientation. Cylindrical bone cores (average diameter = 2.5 mm, length = 6–8 mm) were obtained using internally irrigated trephine bur.

Bone core processing and analysis

Upon harvesting, bone core samples were immediately frozen (liquid nitrogen) and stored (-80°C) until micro-CT scanning and further processing.

Micro-CT Scan analysis—For quantitative three-dimensional (3D) analyses, cores were thawed and scanned parallel to longitudinal axis (360° full scan). The instrument (Inveon[®] micro-CT, Siemens Medical Solutions, PA, USA) scanned at a voxel size of $10.11\ \mu\text{m}$ in all three dimensions. A filter (Al thickness = 1.5 mm) was used to account for beam hardening. Proprietary 3D software (Inveon[®] micro-CT) was used to gather volumetric and density data and to perform structural analysis. Bone and bone graft voxels were segmented from non-bone voxels using a heuristic algorithm as previously (Kim et al. 2004, Zauel et al. 2004). Total counts and CT attenuation values of bone and bone graft voxels were collected in the process of segmentation. CT attenuation values were converted to degree of bone mineralization (DBM) and a DBM histogram was constructed (Fig. 2). Specimen mean DBM values were computed (sum of DBM values divided by total bone and bone graft voxel counts). Mineralization and resorption velocities in given volumes of bone voxels were obtained by subtracting Low5 from Low25 (Low25-Low5) and High25 from High5 (High5-High25) respectively (Fig. 2; modified from Ruffoni et al. 2007).

Volume of interest (VOI) was drawn with a slice-based method, starting from surface of the core and moving towards its center. Microstructural indices were calculated directly from VOI ($4.60 \pm 1.02\ \text{mm}^3$). Total bone, new bone, graft material, mineralization per unit volume and trabeculation were analysed.

Histomorphometric and immunohistochemical analysis—On completion of micro-CT scan, cores were fixed in formalin, decalcified (formaldehyde and formic acid), embedded in paraffin and sectioned ($4\text{-}\mu\text{m}$ thickness) parallel to long axis. Three slides, each carrying six sections (18 sections/core), were obtained from the central portion of each core, stained (Masson's trichrome for collagen type I) and analysed under light microscopy. Another three slides were prepared for immunohistochemistry, labelled with fluorescent-labelled antibodies against CD34 (130-081-001 CD34 human, clone 136 mouse monoclonal (IgG2A) antibody; Life Technologies, Grand Island, NY, USA) and von Willebrand factor (ab8822 Anti-Von Willebrand factor [FITC] sheep antibody; Abcam, Cambridge, MA, USA) to identify endothelial cells. The apical, middle and coronal third portions of each section (total: 54 readings/core) were documented at $5\times$ magnification with digital photomicrographs (Figs 3 and 4). A 10×10 ocular grid was used to manually measure surface area occupied by graft material, new cellular bone and immature tissue. Graft material was included into mature bone if it stained only red (no resorption characteristics and no surrounding new bone). Graft material was included into new bone if it was partially stained red and blue with resorption characteristics and surrounded with new bone. Average

readings for each core were calculated for apical and coronal parts. In addition, software (Axiovision software, ©Carl Zeiss MicroImaging, Inc., Thornwood, NY, USA) was used to automatically calculate the same parameters.

Data management and statistical analysis

Clinical parameter measurements (PD, GR and CAL) (six surfaces per tooth) were averaged; the three buccal and the three lingual/palatal measurements were averaged to obtain a single buccal and a single lingual/palatal value per tooth. Histomorphometry and immunohistochemistry data are presented as distributions (percent) and as number of positively labelled cells, respectively.

The site was the unit of statistical analysis, performed using computer software (SAS 9.2; SAS Institute Inc., Cary, NC, USA and GraphPad Prism 5; GraphPad Software Inc., La Jolla, CA, USA). Descriptive statistics were calculated for demographic, clinical and bone analysis parameters, and data presented as mean $\pm$ SE (median; range) based on parametric or non-parametric analysis. Similarly, parametric or non-parametric methods were chosen based on data distribution within study population; data normality was tested by Shapiro–Wilks test and was also visually inspected by examining the normal probability plots and histograms. Chi-square test was used to reveal the relationship between two categorical variables. Within-group differences between baseline and re-entry were analysed by paired *t*-test or by Wilcoxon signed rank sum test if the data were not normally distributed. Between-group (maxilla and mandible) differences at baseline and re-entry were analysed by unpaired *t*-test or Mann–Whitney *U* test if data was not normally distributed.

For outcome analyses (ridge height or width gain/loss), sites were grouped by jaw. The stepwise method was used to select variables that confounded the relationship between outcome variables and the location (site) variable. Univariate model was initially completed with location variable. Then each variable was added into the model to determine whether the second variable changed the location coefficient in the model. Finally, multiple regression model was used with the variables that confounded the relationship between outcome and location. Spearman correlation coefficient was applied to determine the level of association between various initial parameters and outcome values. The level of statistical significance was set at $\alpha=0.05$.

Results

Study population

Thirty-six subjects were recruited and 24 completed the study; 12 subjects failed to comply with study protocol (by choosing to delay implant-placement surgery). Each patient contributed a single site, except for one patient who contributed two maxillary sites, resulting in 25 total sites analysed (10 maxilla, 15 mandible). Demographic and other study population data are presented in Table 1. There were no differences between maxilla and mandible groups, except for tooth type distribution (maxilla: 8 premolars, 2 molars; mandible: 4 premolars, 11 molars; $p=0.03$; chi-square). The main reason for tooth extraction was non-restorability (severe decay). At re-entry, all sites were suitable for

implant placement and implants were successfully placed at all sites; two sites (one maxillary, one mandibular) required bone grafting during implant placement because dehiscence occurred.

Clinical Parameters

Initial post-extraction relative ridge height and ridge width (Table 2) did not differ between jaws ($p > 0.05$; unpaired t -test). Buccolingual socket size (residual defect size) was 8.5 ± 0.4 (8.5; 6–11) mm and 8.5 ± 0.4 (9; 6–11) mm for maxillary and mandibular sites, respectively ($p > 0.05$). Root diameter and root length for extracted maxillary teeth were 7 ± 0.5 (6.5; 5–9) mm and 13 ± 0.7 (13; 10–16) mm, respectively, while for mandibular teeth, they were 8.2 ± 0.4 (8; 5–10) mm and 13.4 ± 0.6 (14; 8–16) mm; neither root dimension differed between jaws ($p > 0.05$).

Buccal bone plate thickness was 1.5 ± 0.2 (1.5; 0.5–2) mm for maxillary and 1.5 ± 0.2 (1.5; 0.5–2.5) mm for mandibular sites. Palatal (lingual) bone plate thickness was 1.3 ± 0.13 (1; 1–2) mm for maxillary and 2 ± 0.3 (1.5; 0.5–5) mm for mandibular sites. Differences between jaws were not significant ($p > 0.05$).

The post-extraction alveolar ridge width measurement (Table 2) was compared to the respective sum of buccal plate thickness, defect size and palatal (lingual) plate thickness for maxillary [11.1 ± 0.5 (11; 9–15) mm] and mandibular [11 ± 0.6 (11; 7–15) mm] sites; these comparisons revealed negligible differences. Membrane exposure at ARP procedure completion (Table 1) was common (>75% of sites).

At re-entry, maxillary sites had experienced 0.2 ± 0.3 mm of height loss while mandibular sites presented a height gain of 1 ± 0.3 mm (Table 2); neither change was statistically significant ($p > 0.05$). However, under aspect-specific analysis, the height gain at the distal buccal aspect of mandibular sites was statistically significant [both within the mandible compared to initial relative height ($p = 0.01$) and, as an outcome difference between mandible and maxilla at reentry ($p = 0.02$)] (Table 2).

At re-entry, maxillary and mandibular sites had lost 2–2.5 mm of width, on average (Table 2), a statistically significant change for both jaws ($p = 0.01$), with no significant difference between jaws ($p > 0.05$).

Correlation between initial clinical parameters and ARP outcomes

Initial clinical periodontal parameters (PD, GR, CAL and KT), extracted tooth type and root dimensions, buccal and palatal plate thickness, socket size, gender, age and healing time were all investigated as possible factors affecting ARP outcomes.

When maxilla and mandible were compared for ridge height outcome (gain/loss), there was statistically significant difference between jaws at the distal buccal aspect of extraction site after adjustment of univariate model for covariates ($p = 0.01$). Root diameter and buccal PD were the only parameters that confounded the relationship between ridge height outcome (gain/loss) and location (maxilla/mandible); however, neither parameter had a significant direct effect on ridge height outcome ($p > 0.05$).

An analysis to determine possible factors affecting ridge height outcome was also performed by pooling all sites ($n = 25$). The analysis revealed that healing time ($p = 0.05$), mid-buccal CAL ($p = 0.001$) and mid-buccal KT ($p = 0.005$) had a statistically significant effect. However, none of these parameters was significantly correlated with ridge height outcome when analysed individually (healing time: $r = 0.21$, $p = 0.3$; CAL: $r = 0.38$, $p = 0.06$; KT: $r = -0.14$, $p = 0.5$).

As already mentioned, alveolar ridge width loss did not differ between jaws. Thus, statistical analysis was repeated after pooling all sites ($n = 25$). Following corrections for covariates, buccal plate thickness and root length were found to be confounders for the observed alveolar ridge width loss ($p = 0.03$), with both factors negatively affecting the outcome. There was a statistically significant correlation between buccal plate thickness and ridge width loss ($r = -0.49$, $p = 0.01$); sites with thicker buccal plate lost more ridge width. Root length was not significantly correlated with ridge width loss ($r = -0.35$, $p = 0.09$). Ridge width loss was correlated with initial ridge width ($r = -0.42$, $p = 0.03$), that is, initially wider ridges lost more width. Initial ridge width did not correlate with width loss when loss was expressed as percentage of initial width ($p > 0.05$).

When outcomes for ridge height and width were correlated with each other, there was a negative but statistically non-significant correlation ($r = -0.19$, $p = 0.35$).

Bone healing assessment

Micro-CT scan analysis—The average healing time for analysed bone cores was 124 ± 3 days and 133 ± 4 days for maxilla and mandible, respectively (cores excluded from two subjects with 8 months of healing time). The micro-CT-based analysis revealed no significant difference in amount of total bone, new bone, graft material and trabeculation, and mean DBM between maxilla and mandible ($p > 0.24$). However, mineralization/unit volume was higher in newly forming (less mineralized) bone than resorption per unit volume in mature bone and mineralized bone graft in mandible ($p < 0.001$); this difference was not significant in maxilla ($p = 0.158$) (Table 3).

Histomorphometric analysis—Histomorphometric analysis (manually) revealed similar distributions of mature bone, new bone and immature tissue in the apical and coronal portions of both maxillary and mandibular bone cores (Table 3). Following adjustment of the univariate model for covariates, there was statistically significant difference between maxilla and mandible for new cellular bone at the coronal core portion ($p = 0.03$). The confounders for this difference were tooth type, root diameter, buccal PD and buccal CAL of extracted tooth. There were no direct statistically significant differences between jaws for the histomorphometric parameters ($p > 0.05$; Table 3). Results were similar when histomorphometric measurements were obtained automatically (using computer software program; data not shown).

Immunohistochemical analysis—Immunohistochemical analysis revealed angiogenic activity at various levels for all sites, regardless of antibody used to identify newly forming endothelial wall (Table 3). The apical bone core portion exhibited a non-significant trend for higher angiogenic activity (Table 3).

Discussion

The purpose of this study was to investigate healing outcomes following ARP performed on posterior sites, using FDBA and collagen membrane, and to identify site determinants of ARP outcomes. The results showed that in both maxillary and mandibular posterior sites, ARP maintains ridge height, while approximately 2.5 mm ridge width loss occurs despite ARP. Using similar ARP approaches, others have earlier reported comparable outcomes (Nishimura et al. 1987, Simon et al. 2000, Clem 2000, Iasella et al. 2003). Although reported ridge width loss even in the presence of ARP is significant, this amount is less than what is reported at extraction sites healed without ARP (Barone et al. 2012, Cardaropoli et al. 2012). The ARP procedure outcome (reduced width loss combined with height loss prevention) is of significant clinical benefit, as demonstrated by the fact that in all cases it was possible, at re-entry, to successfully place a dental implant. This study identified healing time, CAL and KT at extraction site as determinants of ridge height preservation, and buccal plate thickness and tooth root length as determinants of ridge width loss. In addition, initial ridge width was positively correlated with ridge width loss. To the best of our knowledge, this is the first study to identify possible determinants of ARP outcomes.

ARP outcomes in this study differed between maxilla and mandible at the distal buccal aspect of the extraction site, with mandibular sites experiencing height gain. This is consistent with previous studies' results, where mandibular sites exhibited less height loss than maxillary ones (Iasella 2003; Hoffmann et al. 2008). Although bucco-lingual root diameter and buccal PD were identified as confounders for the difference between maxilla and mandible, the direct effect of these two parameters on height outcome was not statistically significant. When pooled data from both jaws were analysed for ARP ridge height outcome, healing time, CAL and KT were identified as confounders for ridge height outcome. However, these parameters did not have statistically significant direct correlation with height outcome.

Ridge width loss was approximately 2.5 mm at both anatomical sites (no significant difference between jaws). When maxillary and mandibular sites were pooled, buccal plate bone thickness and extracted tooth root length were identified as confounders for the observed ridge width outcome, with buccal plate thickness being significantly correlated with ridge width loss ($r=-0.49$, $p=0.01$). This result, where thicker buccal plate was associated with greater ridge width loss, is in conflict with other studies. Studies have reported a positive effect of thicker buccal plate on surgical outcomes following immediate implant placement (Tomasi et al. 2010, Ferrus et al. 2010). Similarly, the potential positive impact of buccal plate thickness on post-extraction ridge width as physiological healing outcome has been previously discussed (Smukler et al. 1999, Iasella 2003). Although flap elevation, surgical trauma on buccal plate and jeopardized blood supply (membrane placement between flap and bone) have been listed as possible factors affecting ridge width following ARP (Iasella 2003, Covani et al. 2011), this is the first study to report a statistical association between residual buccal bone plate thickness and post-ARP ridge width change. The reasons for the discrepancy in the contribution of the buccal plate to ridge dimension outcomes in studies investigating immediate implants (Tomasi et al. 2010, Ferrus et al. 2010) and studies investigating ARP (present study) are speculative at this point. The reasons could

be physiological (residual buccal bone more likely to receive mechanical stimulation when implants are placed), methodological or anatomical. The exclusion, from this study, of sites eligible for immediate implant placement suggests that the two types of studies (ARP *versus* immediate implants) may select for sites that differ anatomically. The statistically significant positive correlation between initial ridge width and ridge width loss is consistent with the aforementioned correlation of buccal plate thickness and width loss (buccal plate thickness is a component of initial ridge width). Another hitherto unreported finding was that sites of teeth with longer roots experienced more ridge width loss following ARP, although the direct effect of root length on ridge width outcome did not reach statistical significance. Studies on tooth anatomy and its effect on extraction, surgical techniques and residual alveolar bone are limited to impacted third molars and the prognosis for adjacent second molars (Karapataki et al. 2000a, b). These results regarding root length should be interpreted cautiously because only the longest tooth root was measured, which does not provide information on root position relative to buccal/lingual plate.

As the post-extraction ridge loses width, it is inevitable that any standardized position height measurement will be made at a location that is more apical post-operatively. This observation, made also by other investigators (Brownfield & Weltman 2011, Iasella et al. 2003), suggests that there should be a negative correlation between ridge height and width outcomes. In this study, the investigated correlation between height and width outcome was indeed negative, but non-significant. Additional studies are necessary to determine the relationship between ridge height and width changes.

In this study, the healing time between ARP procedure and implant placement was approximately 4 months, chosen based on earlier studies (Hammerle et al. 1996). Iasella et al. (2003) reported minimal differences in ridge height or width changes between 4- and 6-month healing times after ARP. The healing time was a confounder for ridge height outcome in this study. However, it did not have statistically significant direct effect on height outcome.

There was no statistically significant difference between jaws for trabeculation or amount of bone formation (micro-CT). Similarly, histological analysis revealed no statistically significant differences between jaws regarding percentage of mature bone, new bone, new cellular bone or immature tissue. Interestingly, there was statistically higher rate of mineralization per unit volume in newly forming bone compared with the resorption of mature bone (and mineralized bone graft) in mandible (micro-CT). Finally, immunohistochemistry revealed higher, but not statistically significant, angiogenic activity in mandible compared with maxilla. Collectively, the above healing aspects observed in the mandible might partly explain the better ridge height outcomes in the lower jaw.

When histological outcome was analysed for possible confounders, a statistically significant difference between jaws was found for coronal new cellular bone. Following adjustment for covariates, the possible confounders for this differential outcome were tooth type, root diameter, buccal PD and buccal CAL of the extracted tooth. However, there was no significant direct effect of these factors on histological outcome. The coronal aspect of the wound is almost always exposed to greater inflammatory challenge and this may affect

histological outcome (Romano et al. 1997, Artzi et al. 2000). Greater pre-extraction PD and CAL reflect greater periodontal tissue loss and possibly greater inflammation locally.

The high rate of membrane exposure encountered in this study is an expected ARP limitation (Tal et al. 2008). Exposures were generally small (4 mm). Zubillaga et al. (2003) found no significant detrimental effect of membrane exposure on height outcomes following ARP. Wound size, flap elevation and closure were controlled in this study by site selection (single posterior tooth with intact adjacent teeth).

Factors such as age (Amler 1977, Amler 1993), gender (Engeland et al. 2006, August et al. 2001) and environmental factors (e.g. smoking, Haber & Kent 1992, Riebel et al. 1995) can differentially affect oral soft and hard tissue healing. In this study, subjects were randomly selected based on their need for extraction and ARP, and the two groups (maxilla, mandible) did not differ with respect to age or gender. Furthermore, smokers were excluded. Therefore, age, gender or smoking cannot account for observed inter-group differences.

Two limitations of this study are sample size and tooth type differences between jaws. In this study, a third of recruited subjects opted to delay their implant placement surgery, mostly for financial reasons, resulting in significant dropout rate and reduced sample size. Although a larger study population would be desirable, to account for anticipated wound-healing variability, the final sample size was within the size indicated by a priori power analysis to detect a 2-mm difference in ridge dimensions. Despite the statistically significant difference between maxilla and mandible for tooth type, there were no inter-group differences in defect size following extraction, and thickness of buccal or palatal (lingual) plate. Furthermore, tooth type did not emerge as a confounder in any of the clinical outcome analyses. A possible additional limitation to consider is surgeon's experience. Although this variable may affect procedure outcomes, it did not appear to be a factor in this study, as the results obtained were comparable to those of other studies. Furthermore, this variable was controlled by standardization of ARP indication and surgical protocol; inclusion of surgeons in their second or third year of training and working under close supervision by a board-certified periodontist (full-time faculty).

Within the limitation of this study, the results suggest that in different anatomical locations, different factors may determine the clinical outcome following ARP treatment. These results may help clinicians to better predict ARP treatment outcomes. Further studies are needed to better understand grafted alveolar socket wound healing at the cellular level, to determine clinical limitations of ARP, and to improve ARP protocols.

Acknowledgements

Authors would like to thank Dr. M. Knopp and Ms. M. Carlton from Microimaging Lab, Department of Radiology, College of Medicine, The Ohio State University (OSU) for their assistance in micro-CT scanning, Ms. M. Lloyd from Oral Pathology, OSU for preparation of histological slides, Drs. Susan and Joe Travers from Oral Biology, OSU for morphometric analysis and Mr. Gary S. Phillips from the Center for Biostatistics, OSU for statistical analysis support.

source of funding

This study was supported by OSU College of Dentistry start-up funds provided to Dr. Leblebicioglu and by the OSU Division of Periodontology.

References

- Amler MH. The age factor in human extraction wound healing. *Journal of oral surgery*. 1977; 35:193–197. [PubMed: 264520]
- Amler MH. Age factor in human alveolar bone repair. *The Journal of oral implantology*. 1993; 19:138–142. [PubMed: 8246300]
- Artzi Z, Tal H, Dayan D. Porous Bovine Bone Mineral in healing of human extraction sockets. Part 1: Histomorphometric Evaluations at 9 months. *Journal of Periodontology*. 2000; 71:1015–1023. [PubMed: 10914806]
- August M, Chung K, Chang Y, Glowacki J. Influence of estrogen status on endosseous implant osseointegration. *Journal of Oral and Maxillofacial Surgery*. 2001; 59:1285–1289. discussion 1290–1291. [PubMed: 11688027]
- Barone A, Ricci M, Tonelli P, Santini S, Covani U. Tissue changes of extraction sockets in humans: a comparison of spontaneous healing versus ridge preservation with secondary soft tissue healing. *Clinical Oral Implants Research*. 2012 doi: 10.1111/j.1600-0501.2012.02535.x.
- Bartee BK. Extraction site reconstruction for alveolar ridge preservation. Part 2: membrane-assisted surgical technique. *The Journal of oral implantology*. 2001; 27:194–197. [PubMed: 12500878]
- Boyne PJ. Osseous repair of the postextraction alveolus in man. *Oral Surgery, Oral medicine, and Oral Pathology*. 1966; 21:805–813.
- Brownfield LA, Weltman RL. Ridge Preservation With or Without an Osteoinductive Allograft. A Clinical, Radiographic, Micro-CT, and Histologic Study Evaluating Dimensional Changes and New Bone Formation of the Alveolar Ridge. *Journal of Periodontology*. 2011; 83:581–589. [PubMed: 21942791]
- Cardaropoli D, Tamagnone L, Roffredo A, Gaveglia L, Cardaropoli G. Socket preservation using bovine mineral and collagen membrane: a randomized controlled clinical trial with histologic analysis. *The International Journal of Periodontics & Restorative Dentistry*. 2012; 32:421–430. [PubMed: 22577648]
- Clem D. Implant site development using guided bone regeneration techniques. *Compendium of Continuing Education in Dentistry*. 2000; 21(424–8):430.
- Covani U, Ricci M, Bozzolo G, Mangano F, Zini A, Barone A. Analysis of the pattern of the alveolar ridge remodeling following single tooth extraction. *Clinical Oral Implants Research*. 2011; 8:820–825. [PubMed: 21198897]
- Engeland CG, Bosch JA, Cacioppo JT, Marucha PT. Mucosal wound healing: the roles of age and sex. *Archives of Surgery*. 2006; 141:1193–1197. discussion 1198. [PubMed: 17178961]
- Ferrus J, Cechinato D, Pjetursson EB, Lang NP, Sanz M, Lindhe J. Factors influencing ridge alterations following immediate implant placement into extraction sites. *Clinical Oral Implants Research*. 2010; 21:22–29. [PubMed: 19912273]
- Fickl S, Zuhr O, Wachtel H, Bolz W, Huerzeler M. Tissue alterations after tooth extraction with and without surgical trauma: a volumetric study in beagle dog. *Journal of Clinical Periodontology*. 2008a; 35:356–363. [PubMed: 18353082]
- Fickl S, Zuhr O, Wachtel H, Stappert CFJ, Stein JM, Huerzeler MB. Dimensional changes of the alveolar ridge contour after different socket preservation techniques. *Journal of Clinical Periodontology*. 2008b; 35:906–913. [PubMed: 18713258]
- Haber J, Kent RL. Cigarette smoking in a periodontal practice. *Journal of Periodontology*. 1992; 63:100–106. [PubMed: 1552463]
- Hammerle CH, Schmid J, Olah AJ, Lang NP. A novel model system for the study of experimental guided bone regeneration in humans. *Clinical Oral Implants Research*. 1996; 7:38–47. [PubMed: 9002821]
- Hoffmann O, Bartee BK, Beaumont C, Kasaj A, Deli G, Zafiropoulos GG. Alveolar bone preservation in extraction sockets using non-resorbable dPTFE membranes: a retrospective non-randomized study. *Journal of Periodontology*. 2008; 79:1355–1369. [PubMed: 18672984]
- Iasella JM, Greenwell H, Miller RL, et al. Ridge preservation with freeze-dried bone allograft and a collagen membrane compared to extraction alone for implant site development: a clinical and histologic study in humans. *Journal of Periodontology*. 2003; 74:990–999. [PubMed: 12931761]

- Karapataki S, Hugoson A, Falk H, Laurell L, Kugelberg CF. Healing following GTR treatment of intrabony defects distal to mandibular 2nd molars using resorbable and non-resorbable barriers. *Journal of Clinical Periodontology*. 2000a; 27:333–340. [PubMed: 10847537]
- Karapataki S, Hugoson A, Kugelberg CF. Healing following GTR treatment of bone defects distal to mandibular 2nd molars after surgical removal of impacted 3rd molars. *Journal of Clinical Periodontology*. 2000b; 27(5):325–332. [PubMed: 10847536]
- Kim DG, Christopherson GT, Dong XN, Fyhrie DP, Yeni YN. The effect of microcomputed tomography scanning and reconstruction voxel size on the accuracy of stereological measurements in human cancellous bone. *Bone*. 2004; 35:1375–1382. [PubMed: 15589219]
- Moya-Villaescusa MJ, Sanchez-Perez A. Measurement of ridge alterations following tooth removal: a radiographic study in humans. *Clinical Oral Implants Research*. 2010; 21:237–242. [PubMed: 20070757]
- Nemcovsky CE, Serfaty V. Alveolar ridge preservation following extraction of maxillary anterior teeth. Report of 23 consecutive cases. *Journal of Periodontology*. 1996; 67:390–395. [PubMed: 8708965]
- Nishimura I, Damiani PJ, Atwood DA. Resorption of residual ridges (RRR) in rats. *Journal of Dental Research*. 1987; 66:1753–1757. [PubMed: 3479475]
- Riebel GC, Boden SD, Whitesides TE, Hutton WC. The effect of nicotine on incorporation of cancellous bone graft in an animal model. *Spine*. 1995; 20:2198–2202. [PubMed: 8545712]
- Romano PR, Caton JG, Puzas JE. The reversal line may be a key modulator of osteoblast function: observations from an alveolar bone wound-healing model. *Journal of Periodontal Research*. 1997; 32:143–147. [PubMed: 9085225]
- Ruffoni D, Fratzl P, Roschger P, Klaushofer K, Weinkamer R. The bone mineralization density distribution as a fingerprint of the mineralization process. *Bone*. 2007; 40:1308–1319. [PubMed: 17337263]
- Simon BI, Von Hagen S, Deasy MJ, Faldu M, Resnansky D. Changes in alveolar bone height and width following ridge augmentation using bone graft and membranes. *Journal of Periodontology*. 2000; 71:1774–1791. [PubMed: 11128929]
- Smukler H, Landi L, Setayesh R. Histomorphometric evaluation of extraction sockets and deficient alveolar ridges treated with allograft and barrier membrane: a pilot study. *International Journal of Oral and Maxillofacial Implants*. 1999; 14:407–416. [PubMed: 10379115]
- Tal H, Kozlovsky A, Artzi Z, Newcovsky CE, Moses O. Long-term bio-degradation of cross-linked and non-cross-linked collagen barriers in human guided bone regeneration. *Clinical Oral Implants Research*. 2008; 19:295–302. [PubMed: 18190562]
- Ten Heggeler JM, Slot DE, Van der Weijden GA. Effect of socket preservation therapies following tooth extraction in non-molar regions in humans: a systematic review. *Clinical Oral Implants Research*. 2011; 22:779–788. [PubMed: 21091540]
- Trombelli L, Farina R, Marzola A, Bozzi L, Liljeborg B, Lindhe J. Modeling and remodeling of human extraction sockets. *Journal of Clinical Periodontology*. 2008; 35:630–639. [PubMed: 18498382]
- Tomasi C, Sanz M, Cecchinato D, Pjetursson B, Ferrus J, Lang NP, Lindhe J. Bone dimensional variations at implants placed in fresh extraction sockets: a multilevel multivariate analysis. *Clin. Oral Implants Res*. 2010; 21:30–36. [PubMed: 20070744]
- Van der Weijden F, Dell'Acqua F, Slot DE. Alveolar bone dimensional changes of postextraction sockets in humans: a systematic review. *Journal of Clinical Periodontology*. 2009; 36:1048–1058. [PubMed: 19929956]
- Zauel R, Yeni YN, Christopherson GT, Cody DD, Fyhrie DP. Segmentation algorithm for accurate 3D representation of microcomputed tomographic images of human vertebral bodies. *Transactions of Orthopaedic Research Society*. 2004; 29:1018.
- Zubillaga G, Von Hagen S, Simon BI, Deasy MJ. Changes in alveolar bone height and width following postextraction ridge augmentation using a fixed bioabsorbable membrane and demineralized freeze-dried bone osteoinductive graft. *Journal of Periodontology*. 2003; 74:965–975. [PubMed: 12931758]

Clinical relevance**Scientific rationale for the study**

ARP is routinely used procedure. However, confounding factors that determine clinical and histological outcomes are not well defined. This study investigates whether initial anatomical factors such as location, size and periodontal support of the extracted root affect treatment outcome following ARP.

Principal findings

Different factors may be confounders of ARP outcome at different anatomical locations.

Practical implications

The presented data may help improve our understanding on factors that affect ARP outcome.

Fig. 1. Clinical image of template being used during ridge height measurements. Note the discernible three incorporated aluminium tubes defining the mesial (probe in place), central and distal third of the buccal aspect of the target extraction site.

Fig. 2. Representative histogram of degree of bone mineralization (DBM) obtained from micro-CT scan of typical mandibular bone core specimen. The calculated mean DBM value (arrow), and the mineralization (Low 25-Low5) and resorption (High5-High25) velocities are identified.

Fig. 3. Photomicrograph of representative bone core section (Mason Trichrome; Original magnification 5×). Identified areas represent residual bone graft material (A), new bone forming around graft material (B), immature tissue (C) and new cellular bone (D).

Fig. 4. Photomicrographs of representative bone core section examined by immunohistochemistry (Original magnification 5 $\times$; Call outs: original magnification 20 $\times$). Top panel: stained for CD34. Bottom panel: stained for von Willebrand factor.

Table 1

Demographics of maxilla and mandible groups

	Maxilla		Mandible		<i>p</i> value
Age (years)					
Median (range)	52 (24–79)		61 (24–83)		<i>p</i> > 0.05
Mean ± SE	52 ± 5 (<i>n</i> = 9)		55 ± 5 (<i>n</i> = 15)		
Gender (M:F)	2:7		6:9		<i>p</i> > 0.05
Tooth type (Premolar:Molar)	8:2		4:11		<i>p</i> = 0.03
Extraction	4 fractures		3 fractures		
Reason	1 endodontic		2 endodontic		
	0 periodontal		2 periodontal		
	5 non-restorable		8 non-restorable		
CAL (mm)	Buccal	Palatal	Buccal	Lingual	
Median (range)	2 (0–4)	3 (0–3)	2 (1–4)	2 (2–6)	<i>p</i> > 0.05
Mean ± SE	2 ± 0.3	2 ± 0.4	2 ± 0.2	3 ± 0.3	
PD (mm)					
Median (range)	2 (1–2)	2 (1–3)	2 (1–4)	2 (2–6)	<i>p</i> > 0.05
Mean ± SE	2 ± 0.2	2 ± 0.3	2 ± 0.2	3 ± 0.3	
GR (mm)					
Median (range)	0 (–1 to 2)	0 (–1 to 1)	0 (0–1)	0 (0–2)	<i>p</i> > 0.05
Mean ± SE	0 ± 0.3	0 ± 0.2	0 ± 0.1	0 ± 0.2	
KT (mm)					
Median (range)	4 (3–6)		4 (3–8)		<i>p</i> > 0.05
Mean ± SE	4 ± 0.4		5 ± 0.4		6 ± 1
Healing time (days)					
Median (range)	125 (111–246)		127 (111–192)		<i>p</i> > 0.05
Mean ± SE	136 ± 12 (<i>n</i> = 10)		137 ± 12 (<i>n</i> = 15)		
Membrane exposure (mm)					
Median (range)	1 (0–2)		2 (0–4)		<i>p</i> > 0.05
Mean ± SE	1.2 ± 0.2		2 ± 0.3		
	[<i>n</i> = 8 exposures]		[<i>n</i> = 11 exposures]		

CAL, Clinical attachment level; PD, probing depth; GR, gingival recession; KT, keratinized tissue width.

Bold indicates significant *p* value.

Table 2

Alveolar ridge preservation: clinical outcomes

	Initial	Re-entry	Change (Loss/gain)*	CHANGE(%)**	p-value
<i>Relative Ridge Height (mm)</i>					
Maxilla (n = 10)					
Mesial					
Mean ± SE	10 (7–13)	10 (7–13)	1 ((–)2–2)		p > 0.05
Median (range)	10 ± 0.6	10 ± 0.5	0 ± 0.5		
Middle					
Mean ± SE	11 (6–15)	10 (9–17)	0 ((–)4–2)		p > 0.05
Median (range)	10 ± 1	11 ± 0.8	–0.6 ± 0.72		
Distal [†]					
Mean ± SE	10 (7–12)	11 (9–14)	0 ((–)2–1)		p > 0.05
Median (range)	10.3 ± 0.5	11 ± 0.5	–0.33 ± 0.4		
Mandible (n = 15)					
Mesial					
Mean ± SE	11 (6–15)	10 (7–13)	2 ((–)4–5)		p > 0.05
Median (range)	11.3 ± 0.8	10.4 ± 0.4	1.4 ± 0.7		
Middle					
Mean ± SE	12 (8–15)	11 (9–12)	1 ((–)2–4)		p > 0.05
Median (range)	11.7 ± 0.6	11 ± 0.2	0.7 ± 0.5		
Distal [†]					
Mean ± SE	12 (7–15)	10 (8–12)	1 ((–)1–4)		p = 0.01
Median (range)	11.8 ± 0.6	10.3 ± 0.2	1.4 ± 0.5		
<i>Ridge Width (mm)</i>					
Maxilla (n = 10)					
Mean ± SE	11 (9–15)	8.5 (6–12)	–3 ((–)5–1)	27 (40–(–11))	p = 0.01
Median (range)	11.1 ± 0.5	9 ± 0.6	–2.2 ± 0.7	19 ± 6	
Mandible (n = 15)					
Mean ± SE	11 (7–15)	8 (6–12)	–3 ((–)6–1)	20 (46–(–14))	p < 0.01
Median (range)	11 ± 0.6	8.6 ± 0.6	–2.4 ± 0.5	21 ± 5	

Values in bold indicate significant p values.

* Negative values represent loss of ridge dimension from initial to re-entry.

** Negative values represent gain of ridge dimension from initial to re-entry.

[†] p = 0.02; Outcome difference between maxilla and mandible.

Table 3

Alveolar ridge preservation: histomorphometric and histological outcomes

	Low25-Low5 (mg/cm ³)*	High5-High25 (mg/cm ³)**		<i>p</i> -value
Micro-CT				
Maxilla (<i>n</i> = 9)				
(mean ± SE)	89.08 ± 11.79	71.96 ± 8.76		0.158
Mandible (<i>n</i> = 11)				
(mean ± SE)	109.18 ± 10.51	58.22 ± 4.85		<0.001
Histomorphometry				
	Mature bone	New bone	New cellular bone	Immature tissue
Apical part of bone core				
Maxilla (<i>n</i> = 9)				
Median (range)	0% (0–30)	26% (8–43)	9% (0–25)	16% (3–36)
Mean ± SE	7 ± 4%	26 ± 5%	11 ± 2%	19 ± 4%
Mandible (<i>n</i> = 11)				
Median (range)	1% (0–8)	31% (3–54)	14% (0–23)	22% (8–43)
Mean ± SE	2.3 ± 1%	28 ± 4%	12 ± 2%	22 ± 3%
<i>p</i> -value	0.7	0.8	0.6	0.4
Coronal part of bone core				
Maxilla (<i>n</i> = 9)				
Median (range)	1% (0–34)	29% (7–56)	7% (0–30)	30% (8–57)
Mean ± SE	7 ± 4%	29 ± 5%	11 ± 3%	27 ± 5%
Mandible (<i>n</i> = 11)				
Median (range)	2% (0–5)	22% (0–47)	8% (2–38)	18% (3–54)
Mean ± SE	2 ± 1%	24 ± 5%	12 ± 4%	21 ± 5%
<i>p</i> -value	0.2	0.4	0.8	0.4
Immunohistochemistry				
	CD34+ cell count		von Willebrand factor+ cell count	
	Apical part of bone core	Coronal part of bone core	Apical part of bone core	Coronal part of bone core
Maxilla (<i>n</i> = 7)				
Median (range)	70 (28–96)	48 (26–99)	71 (25–93)	42 (25–83)
Mean ± SE	69 ± 9	53 ± 8	66 ± 8	49 ± 8
Mandible (<i>n</i> = 8)				
Median (range)	86 (10–166)	75 (11–100)	92 (13–146)	76 (9–147)
Mean ± SE	81 ± 18	65 ± 13	85 ± 17	74 ± 17
<i>p</i> -value	0.6	0.5	0.4	0.2

Bold indicates significant *p* value.

* Mineralization velocity of newly forming tissue (Low25-Low5).

** Resorption velocity of matured tissue (High5-High25).