Comparison of Microstructures Between Block Grafts from the Mandibular Ramus and Calvarium for Horizontal Bone Augmentation of the Maxilla: A Case Series Study

Alberto Monje, DDS1/Florencio Monje, MD, PhD2
Hsun-Liang Chan, DDS, MS1/Fernando Suarez, DDS1
Laura Villanueva-Alcojol, MD4/Agustin Garcia-Nogales5
Hom-Lay Wang, DDS, MS, PhD1

The primary purpose of this clinical study was to compare architectural metric parameters using microcomputed tomography (micro-CT) between sites grafted with blocks harvested from the mandibular ramus and calvarium for horizontal bone augmentation in the maxilla. The second aim was to compare the primary stability of implants placed in both types of block grafts. Ten consecutive healthy partially edentulous patients requiring extensive horizontal bone reconstruction in the maxilla were included. A total of 14 block grafts (7 each from the mandibular ramus and calvarium) were studied. After 4 to 6 months of healing, 41 implants were placed: 24 implants (58.5%) in calvarial (group 1) and 17 (41.5 %) in ramus grafts (group 2). A resonance frequency analysis (RFA) was performed to test implant stability. Furthermore, two biopsy specimens were randomly selected for histomorphometric analysis. Micro-CT analyses showed no significant difference in the morphometric parametric values analyzed between groups. Furthermore, RFA also showed no difference between groups. However, slightly higher RFA values were noted for implants placed in ramus grafts. Bone quality, as assessed by micro-CT and histomorphometric analyses, was similar in both ramus and calvarial block grafts. In addition, there was no difference in primary implant stability between groups. (Int J Periodontics Restorative Dent 2013;33:e153–e161. doi: 10.11607/prd.1664)
larger amount of intramembranous bone. However, general anesthesia is needed for harvesting the graft. After a block graft is placed, particulate grafts are often added around the block to fill the gaps, providing additional osteoconduction properties. Barrier membranes, either nonresorbable or resorbable ones, may be used to increase the amount of regenerated bone in the grafted area. Nonresorbable membranes have shown higher rates of membrane exposure, thus affecting the amount of bone regeneration. Collagen resorbable membranes, on the other hand, can stimulate DNA syntheses and thus may have some biologic advantage in bone regeneration.

Microcomputed tomography (micro-CT) is a well-documented method to study bone microstructures because it provides accurate three-dimensional (3D) images and is time efficient compared with conventional histomorphometry. Micro-CT images are the result of differences in radiation attenuation properties of bone, marrow spaces, and soft tissues. It can determine 3D bone structures at micrometer to submicrometer resolution and allows for quantification of architectural metric parameters, such as bone volume, total volume, and bone surface.

In 1996, resonance frequency analysis (RFA) was developed and implant stability quotient (ISQ) was used as a quantitative unit to assess implant stability. The RFA reading is a reflection of the combination of three main factors: (1) stiffness of the implant fixture and its interface with the surrounding tissues, (2) design of the transducer, and (3) total effective length above the bone level. The current version of an RFA device uses a small L-shaped transducer to “read” implant stability. This transducer comprises two piezoceramic elements, one vibrating by a sinusoidal sign (5 to 15 Hz), while the other serves as a receptor. The ISQ reading ranges from 0 to 100, with the higher number indicating higher stability. Although there is no definitive threshold value to differentiate a stable, integrated implant from a failing/failed implant, it has been reported that a successful implant had an ISQ ranging from 57 to 82 after 1-year loading. A value that is less than 50 indicates a potential risk of implant failure.

Therefore, the main purpose of this clinical study was to compare architectural metric parameters between sites grafted with blocks harvested from the mandibular ramus and calvarium for horizontal bone augmentation in the maxilla. The second aim was to compare ISQ values between implants placed in both types of block grafts.

Method and materials

Ten consecutive healthy partially edentulous patients requiring extensive horizontal bone reconstruction in the maxilla were included in this controlled clinical study from February 2011 to January 2012. A total of 14 onlay block grafts harvested from either the calvarium or the ramus were placed. Written consent from each subject was obtained prior to treatment.

Inclusion and exclusion criteria

All patients recruited met the following inclusion criteria: between 18 and 85 years of age, no systemic diseases or conditions known to alter bone metabolism, and adequate oral hygiene. Patients were selected to undergo a calvarial or ramus block graft depending on the extent of the defect. Hence, patients who needed more extensive bone regeneration (≥ 20 mm) received calvarial grafts. For defects < 20 mm, patients received mandibular ramus grafts. Patients were excluded if they were pregnant, smokers, taking medications known to modify bone metabolism, or had taken antibiotics for more than 2 weeks in the past 3 months.

Ramus block graft harvesting procedures

Under local anesthesia and intravenous conscious sedation, an incision was made in the posterior mandible following the external oblique ridge. A full-thickness flap was reflected, exposing the lateral aspect of the ramus. Rectangular grafts were harvested using fissure burs to delineate the block and curved chisels and mallet to detach the graft. Sharp edges around the blocks were subsequently smoothened with a large bur.
Calvarial block graft harvesting procedures

Under general anesthesia with local infiltration, an incision was made in the parietal area, parallel to the cranial major axis. The length of the incision was made according to the quantity of bone needed. A rectangular bone block was outlined with a fissure bur and gently detached from the outer cortex using a chisel. The harvested block grafts were then segmented into smaller blocks to graft several defects.

Recipient site preparation and delayed implant placement

At the recipient site, a midcrestal incision was performed with intrasulcular and vertical releasing incisions, after which a full-thickness flap was reflected. The block graft, either from the ramus or calvarium, was adapted to the recipient sites and anchored to the residual ridge by one or two 1.5-mm-diameter titanium fixation screws (Level One 1.5 Neuro, KLS Martin). After achieving stability, sharp edges were smoothed using a fissure bur. A bone substitute of bovine origin (Bio-Oss, Geistlich Pharma) was packed around the graft to fill any voids (Fig 1). Then, a probe was placed close to the rod at the midfacial and buccal sides of the implant. The ISQ was generated and recorded for both sides. The two measurements were averaged to represent the primary stability of each implant.

Micro-CT analysis

Cylindrical bone core biopsy samples were obtained using a 2-mm-diameter trephine bur at the implant sites. The bone specimens were preserved at –20°C. They were scanned with a high-resolution micro-CT (SkyScan 1172) in 100 voltage and 100 microamperage (Figs 2 and 3). The exposure time was 450 ms. Images were reconstructed using software (Nrecon, SkyScan NV) that used the modified algorithm described by Feldkamp et al21 to obtain the axial sections of the specimen. The morphometric variables analyzed included: (1) bone volume (BV); (2) total volume (TB); (3) BV/TB; (4) trabecular thickness (Tb.Th); (5) trabecular separation (Tb.Sp); (6) trabecular number (Tb.N); (7) trabecular pattern factor or inverse connectivity (Tb.Pf), which is an inverse connectivity index: the higher it is the trabecules are less connected; (8) structure model index (SMI), which gives information about preponderance of trabecular morphology (0 is an ideal plate, whereas 3 is an ideal cylinder); and (9) degree of anisotropy (DA), which is the presence or absence of aligned trabecules in a particular direction (1 is considered isotropic, >1 is considered anisotropic).

Histomorphometric analysis

Two biopsy specimens, one from each group, were randomly selected. The mineralized biopsy specimens were progressively dehydrated in 60% ethanol for 2 hours, 80% ethanol for 4 hours, 90% ethanol for 12 hours, and 100% ethanol for 24 hours. After this, bone biopsy specimens were placed in a solution of 50% acetone and 98% ethanol for 4 to 7 months of healing (mean, 5.34 ± 1.66 months), implants were placed according to the initial treatment plan. The implant insertion torque ranged from 35 to 45 Ncm.
6 hours and later in 100% xylene for 2 hours. Finally, the specimens were drained and dried in an incubator at 60°C for 25 hours. Specimens were sectioned along their long axes to 50 ± 10 µm. The slides, including the middle portion of the specimens, were used for the analyses. In each group, one slide was prepared for low-vacuum surface scanning electron microscopy (SEM) (JSM-5500LV, Jeol) and the other for optic microscopic scans (OM). The slides were stained with toluidine blue before the optic microscopic analyses with ×20 and ×25 magnifications. Slide preparations and SEM scans followed standard protocols.

Microphotographs of the entire specimens were obtained with a digital microphotograph system (Nikon, Kodak) and analyzed with a software package (Image-Pro AMS 5.1, Media Cybernetics and Adobe 7 portable, Adobe). The software was used to define the areas of interest and to calculate the percentage of bone tissues and Bio-Oss.

### Statistical analysis

A statistical package (SPSS 13.0, IBM and STATISTICA, version 7.1, StatSoft) was used to analyze the data. Descriptive statistical analysis for continuous and categorical variables was performed. Mixed models for dependent data were applied to analyze ISQ values and to compare morphometric variables. A P value < .05 was considered statistically significant.

### Results

The mean age of patients was 42.4 years, with a 1:1 male/female distribution. A total of 14 block grafts (7 each from the mandibular ramus and calvarium) were placed. Samples were equally distributed between the two groups. Six were placed in the anterior area, two in the posterior area, and the rest...
occupied the anterior and posterior areas. After 4 to 6 months of healing, 41 implants were placed: 24 implants (58.5%) in calvarial (group 1) and 17 (41.5%) in ramus grafts (group 2) (Table 1). All implants were clinically stable.

**Table 1** Biopsy specimen distribution of the onlay grafts

<table>
<thead>
<tr>
<th>Biopsy no.</th>
<th>Donor site</th>
<th>Age (y)</th>
<th>Sex</th>
<th>Recipient site</th>
<th>Implants (n)</th>
<th>Mean ISQ</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Calvarium</td>
<td>22</td>
<td>M</td>
<td>Anterior</td>
<td>2</td>
<td>73.5</td>
</tr>
<tr>
<td>2</td>
<td>Calvarium</td>
<td>22</td>
<td>M</td>
<td>Anterior</td>
<td>2</td>
<td>76</td>
</tr>
<tr>
<td>3</td>
<td>Calvarium</td>
<td>49</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>70.2</td>
</tr>
<tr>
<td>4</td>
<td>Calvarium</td>
<td>49</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>65</td>
</tr>
<tr>
<td>5</td>
<td>Calvarium</td>
<td>56</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>70.5</td>
</tr>
<tr>
<td>6</td>
<td>Calvarium</td>
<td>56</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>70</td>
</tr>
<tr>
<td>7</td>
<td>Calvarium</td>
<td>38</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>73</td>
</tr>
<tr>
<td>8</td>
<td>Ramus</td>
<td>53</td>
<td>F</td>
<td>Anterior</td>
<td>2</td>
<td>75.5</td>
</tr>
<tr>
<td>9</td>
<td>Ramus</td>
<td>49</td>
<td>M</td>
<td>Anterior</td>
<td>2</td>
<td>68.5</td>
</tr>
<tr>
<td>10</td>
<td>Ramus</td>
<td>41</td>
<td>M</td>
<td>Anterior</td>
<td>2</td>
<td>79</td>
</tr>
<tr>
<td>11</td>
<td>Ramus</td>
<td>41</td>
<td>M</td>
<td>Posterior</td>
<td>2</td>
<td>76</td>
</tr>
<tr>
<td>12</td>
<td>Ramus</td>
<td>42</td>
<td>F</td>
<td>Anterior/posterior</td>
<td>4</td>
<td>76.7</td>
</tr>
<tr>
<td>13</td>
<td>Ramus</td>
<td>33</td>
<td>M</td>
<td>Anterior</td>
<td>2</td>
<td>76</td>
</tr>
<tr>
<td>14</td>
<td>Ramus</td>
<td>43</td>
<td>F</td>
<td>Posterior</td>
<td>3</td>
<td>73.6</td>
</tr>
</tbody>
</table>

**Fig 4** Mean ISQ values for the calvarial and ramus groups.

**Fig 5** Median BV/TV values for the calvarial and ramus groups.

RFA

The mean ISQ value was 70.58 ± 4.35 and 75.17 ± 6.42 for groups 1 and 2, respectively (Fig 4). The minimum ISQ value for both groups was 62, while the maximum value was 79 and 86 for groups 1 and 2, respectively. The median ISQ value for groups 1 and 2 was 70 and 75, respectively (Fig 5). There was no significant difference in the ISQ value between both groups but a slightly higher ISQ value ($P = .09$) was observed in group 2.
Table 2  Micro-CT analysis and statistical differences of the biopsy samples

<table>
<thead>
<tr>
<th>Donor site</th>
<th>Biopsy no.</th>
<th>BV/TV</th>
<th>BS/BV</th>
<th>BS/TV</th>
<th>Tb.Th</th>
<th>Tb.Sp</th>
<th>Tb.N</th>
<th>Tb.Pf</th>
<th>SMI</th>
<th>DA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calvarium</td>
<td>1</td>
<td>57.51</td>
<td>8.43</td>
<td>14.67</td>
<td>0.29</td>
<td>0.27</td>
<td>1.95</td>
<td>−1.53</td>
<td>0.97</td>
<td>6.73</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>89.77</td>
<td>10.74</td>
<td>11.96</td>
<td>0.50</td>
<td>0.19</td>
<td>1.76</td>
<td>−10.84</td>
<td>−6.90</td>
<td>3.62</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>64.24</td>
<td>17.93</td>
<td>27.92</td>
<td>0.20</td>
<td>0.24</td>
<td>3.20</td>
<td>−20.11</td>
<td>−3.28</td>
<td>2.76</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>44.51</td>
<td>23.88</td>
<td>10.59</td>
<td>0.16</td>
<td>0.21</td>
<td>2.77</td>
<td>3.69</td>
<td>1.19</td>
<td>1.71</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>30.09</td>
<td>23.96</td>
<td>7.21</td>
<td>0.18</td>
<td>0.30</td>
<td>1.67</td>
<td>7.19</td>
<td>1.94</td>
<td>2.05</td>
</tr>
<tr>
<td></td>
<td>6</td>
<td>29.35</td>
<td>20.59</td>
<td>6.04</td>
<td>0.22</td>
<td>0.36</td>
<td>1.33</td>
<td>7.98</td>
<td>2.50</td>
<td>1.75</td>
</tr>
<tr>
<td></td>
<td>7</td>
<td>44.03</td>
<td>19.21</td>
<td>8.46</td>
<td>0.20</td>
<td>0.26</td>
<td>2.20</td>
<td>2.15</td>
<td>1.06</td>
<td>2.89</td>
</tr>
<tr>
<td>Mean</td>
<td></td>
<td>51.35</td>
<td>17.82</td>
<td>12.40</td>
<td>0.25</td>
<td>0.26</td>
<td>2.12</td>
<td>−1.63</td>
<td>3.07</td>
<td>3.07</td>
</tr>
<tr>
<td>SD</td>
<td></td>
<td>21.28</td>
<td>6.08</td>
<td>7.44</td>
<td>0.12</td>
<td>0.05</td>
<td>0.65</td>
<td>10.32</td>
<td>1.75</td>
<td>1.75</td>
</tr>
<tr>
<td>Ramus</td>
<td>8</td>
<td>90.74</td>
<td>14.66</td>
<td>16.16</td>
<td>0.26</td>
<td>0.07</td>
<td>3.44</td>
<td>−36.17</td>
<td>−14.48</td>
<td>1.44</td>
</tr>
<tr>
<td></td>
<td>9</td>
<td>40.97</td>
<td>3.14</td>
<td>7.68</td>
<td>0.14</td>
<td>0.19</td>
<td>2.82</td>
<td>3.94</td>
<td>1.54</td>
<td>4.07</td>
</tr>
<tr>
<td></td>
<td>10</td>
<td>42.41</td>
<td>25.09</td>
<td>10.61</td>
<td>0.32</td>
<td>0.20</td>
<td>2.65</td>
<td>4.71</td>
<td>1.43</td>
<td>1.53</td>
</tr>
<tr>
<td></td>
<td>11</td>
<td>24.93</td>
<td>22.35</td>
<td>5.52</td>
<td>0.19</td>
<td>0.38</td>
<td>1.27</td>
<td>9.63</td>
<td>2.58</td>
<td>1.88</td>
</tr>
<tr>
<td></td>
<td>12</td>
<td>67.49</td>
<td>4.94</td>
<td>7.32</td>
<td>0.37</td>
<td>0.23</td>
<td>1.84</td>
<td>−7.69</td>
<td>0.85</td>
<td>1.89</td>
</tr>
<tr>
<td></td>
<td>13</td>
<td>44.90</td>
<td>4.58</td>
<td>10.19</td>
<td>0.15</td>
<td>0.18</td>
<td>3.02</td>
<td>−0.30</td>
<td>0.91</td>
<td>1.64</td>
</tr>
<tr>
<td>Mean</td>
<td></td>
<td>49.65</td>
<td>11.56</td>
<td>10.65</td>
<td>0.23</td>
<td>0.24</td>
<td>2.38</td>
<td>−3.54</td>
<td>1.97</td>
<td>1.97</td>
</tr>
<tr>
<td>SD</td>
<td></td>
<td>22.17</td>
<td>9.14</td>
<td>4.44</td>
<td>0.08</td>
<td>0.12</td>
<td>0.80</td>
<td>15.33</td>
<td>0.94</td>
<td>0.94</td>
</tr>
<tr>
<td>P</td>
<td></td>
<td>.86</td>
<td>.18</td>
<td>.63</td>
<td>.90</td>
<td>.65</td>
<td>.53</td>
<td>.64</td>
<td>.70</td>
<td>.29</td>
</tr>
</tbody>
</table>

BV/TV = bone volumetric fraction; BS/BV = bone specific surface; BS/TV = bone surface density; Tb.Th = trabecular thickness; Tb.Sp = trabecular spacing; Tb.N = trabecular number; Tb.Pf = trabecular pattern factor; SMI = structural model index; DA = degree of anisotropy.

**Fig 6**  Histomorphometric analysis by SEM. (a) Histomorphometric analysis of an onlay calvarial block graft (biopsy sample no. 7) by SEM (original magnification ×40). (b) Histomorphometric analysis of an onlay ramus block graft (biopsy sample no. 10) by SEM (original magnification ×40).

**Micro-CT**

Table 2 lists the mean, minimum, maximum, and median values of all measured parameters for groups 1 and 2. None of the morphometric parameters analyzed were found to be significantly different between the groups (Table 2). Actually, as demonstrated by the BV/TV, both groups displayed similar bone volume (51.35 ± 21.28 and 49.65 ± 22.17 for groups 1 and 2, respectively).

**Histomorphometric evaluation**

Under SEM, sample no. 7 (group 1) showed 33.6% bone tissues and 6.2% Bio-Oss; while sample no. 13 (group 2) had 22.3% bone and 15.9% Bio-Oss (Fig 6). Under optic
microscopy, sample no. 7 (group 1) revealed 32.6% bone and 3.9% Bio-Oss and sample no. 10 (group 2), 15.2% bone and 9.2% Bio-Oss (Fig 7).

**Discussion**

This study showed similar microstructures between sites grafted with mandibular ramus and calvarial block grafts, as evidenced by micro-CT analyses. None of the parametric values analyzed were found to be different between the block grafts. Nonetheless, the results should be interpreted with caution because of the nature of a case series study: small sample size, nonmasked examiner, and nonrandomization of the treatment group assigned.

Block graft healing starts with revascularization, where periphery blood vessels grow into the grafts.22 Once the grafts survive the surgical trauma and period of hypoxia, the process of bone remodeling commences. Ramus and calvarial bone grafts are primarily cortical bone. As such, there are few if any viable cells that contribute to bone formation (unlike cancellous bone grafts). Graft survival from hypoxia is not an issue with cortical bone grafts. Cancellous block grafts undergo “creeping substitution,” the process in which new bone is deposited first, followed by resorption of necrotic bone. On the other hand, cortical block grafts undergo “reverse creeping substitution,” whereas bone resorption precedes bone apposition.23 The calvarial and ramus block grafts harvested were mostly cortical bone; therefore, it was understandable that no differences could be identified in bone composition from sites grafted with the two studied autogenous blocks. Literature on bone density from calvaria-grafted sites is limited. Vinci et al22 observed that 4 months after the augmentation, the BV/TV was 38.50%. In this study, the BV/TV of the calvarial graft was higher (51.35% ± 21.28%). The higher bone density in the present study might be due to the longer healing time (4 to 6 months).

Absence of clinical mobility immediately after implant placement represented good primary implant stability.24 Many methods have been proposed to assess initial osseointegration,25 but most of them are no longer used because they are invasive and inaccurate.25 RFA is a valid method to quantify initial implant stability. Primary stability is related to the percentage of bone-to-implant contact and the bone density around the implant.26 RFA was shown to correlate with the amount of cortical bone height.27 In this study, a marginal difference \( P = .08 \) was found between both groups, favoring the ramus graft, therefore suggesting that both calvarial and ramus block grafts provided implants with similar initial stability. The fact that the bone density was similar between the two groups further supported the finding of similar RFA results between the two groups.

Since clinical performances of both studied block grafts were similar, the choice between the two was dependent on other factors. The amount of grafts that could be harvested was larger from the calvarium compared to the ramus. Calvarial bone thickness ranged between 6.8 and 7.7 mm at the thickest and safest area, which was the central part of the parietal bone.29
In addition, the resorption of calvarial bone (< 10%) was generally smaller than that of ramus bone, which ranged from 17% to 25% 6 months after surgery. A recent study by Smolka et al evaluated bone volume and density changes of calvarial split bone grafts after alveolar ridge reconstruction. They found a mean volume reduction of 16.2% after 6 months of healing (15 patients) and 19.2% at a 1-year follow-up (5 patients). Both techniques demonstrated minimal risk of complications and donor site morbidity for experienced operators. In addition, the use of the mandibular ramus was less objectionable for the patient and local anesthesia was sufficient for harvesting.

Regarding micro-CT accuracy, it was reported that the mean difference in density between micro-CT and histomorphometric analysis was 2.5%. Furthermore, the feasibility between the SEM and blue toluidine stains in detecting bone and surrounding tissue were comparable. However, SEM seemed to be more precise for determining areas with high calcium concentration. These histomorphometric analyses were added to the study to confirm and evaluate bone morphology. Even though these results were not conclusive due to the small sample size analyzed, the present study showed similar results to the one performed by Proussaefs and colleagues, who found 34% bone formation at 4 to 8 months after grafting with mandibular ramus blocks.

**Conclusion**

Both calvarial and ramus block grafts can be successfully used to augment a deficient horizontal alveolar ridge and behave similarly with regard to their bone-related morphometric parameters and primary implant stability.

**Acknowledgments**

The authors wish to thank the FEDICOM Foundation (Foundation for the Study of Implantology, Oral and Maxillofacial Surgery), Badajoz, Spain, for their financial support. Also, they want to thank to Ms Purificación Barragán, Center of Implantology, Oral and Maxillofacial Surgery (CICOM), Badajoz, Spain, for her assistance in data collection and Dr Jia-Hui Fu (Assistant Professor, Department of Periodontics, Faculty of Dentistry, National University of Singapore, Singapore) for editing this manuscript. The authors reported no conflicts of interest related to this study.

**References**


