Bone Loss Around Immediately Loaded Transitional Implants: Histologic and Microcomputed Tomographic Analysis—A Case Report

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Immediate loading of dental implants can significantly decrease treatment time and thus increase patient acceptance. However, there is still a need to investigate whether an implant in which primary stability is achieved can be immediately loaded without the formation of connective fibrous tissue at the interface. Three transitional implants were placed in an edentulous mandible: two implants loaded immediately and one left unloaded. All transitional implants were retrieved after a period of 12 weeks to perform qualitative and quantitative analysis of the peri-implant tissue and bone-implant interface. Bone biopsy specimens containing the transitional implant were analyzed using microcomputed tomography (micro-CT). Subsequently, the same samples were analyzed using standard undecalcified histology. Micro-CT analysis showed that bone tissue was slightly detached from the surface of the loaded implant. Histology demonstrated the presence of a soft tissue layer inside the socket. Morphometric values of total bone volume, bone-implant contact, and bone connectivity were higher for the unloaded implant, which appeared to be covered by an almost continuous layer of bone. Micro-CT evaluation of some morphometric parameters and histologic results pertaining to small-diameter transitional implants showed that uncontrolled loading may produce untoward effects on peri-implant bone healing. (Int J Periodontics Restorative Dent 2012;32:e195–e203.)

Titanium dental implants are documented to be predictably successful when proper guidelines are followed. According to the Brånemark two-stage protocol, after implant placement, a period of 3 to 6 months in the absence of occlusal loading is required to achieve successful osseointegration.1,2 Such a strict protocol compels lengthy treatment periods, involving use of a provisional prosthesis between surgery and prosthetic rehabilitation. Over the past two decades, some of the original concepts have been reassessed to satisfy patients’ expectations for shorter rehabilitation periods and increased comfort during healing. Efforts have been undertaken to predictably reduce surgical interventions and to shorten the timeframe between surgery and prosthesis delivery.3 A reduction in treatment time can be achieved by completing prosthetic rehabilitation at an early stage (at a second procedure earlier than the conventional healing period of 3 to 6 months) or even immediately (2 to 3 days after implant placement).4

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Because of such modifications to long-established loading protocols, further understanding of the biomechanical aspects of the bone-implant interface, particularly in immediate loading of dental implants, is strongly needed. Bone healing is regulated by interactions between mechanical factors, such as load magnitudes and directions, and the biologic environment. Clinical and experimental studies have demonstrated that without controlled mechanical loading, the failure rate of early loaded implants is higher than that of implants that have been allowed to heal undisturbed for 3 to 6 months, as in conventional delayed protocols. Thus, it is important to analyze the peri-implant bone architecture and its relationship to applied load to better clarify the biomechanical implications in dental implant failures.

Dental research in the field of implants, biomaterials, and regenerative procedures employs morphometry as a method to evaluate bone integration of dental implants and biomaterials. Qualitative and quantitative measurements of the peri-implant tissues around retrieved implants are typically done by means of light microscopy on thin histologic sections containing both the undecalcified bone tissue and the implant. Bone histomorphometry allows the evaluation of many parameters, such as bone volume and bone structure. Dynamic parameters can also be evaluated (e.g., bone formation and resorption rate, bone balance) by labeling the bone with tetracyclines. These parameters give many indications regarding bone development around implants during unloaded healing or functional loading. Bone morphometric analysis has traditionally been assessed in two-dimensional (2D) histologic sections, with a third dimension added on the basis of stereology. The histologic processing of bone biopsy specimens containing alloplastic implants is a destructive procedure, and the measurement technique for bone histomorphometric analysis is tedious and time consuming.

In an attempt to better evaluate bone connectivity, other three-dimensional (3D) procedures have been proposed. Microcomputed tomography (micro-CT) scanning is a nondestructive alternative approach to outline and quantify bone in three dimensions, allowing higher-resolution 3D images and quantitative measurements of the trabecular bone structure.

Transitional implants are inserted between submerged implants at the time of implant placement to provide support for interim removable partial dentures during healing. Transitional implants are designed with narrower widths compared to standard implants so that they can fit between simultaneously placed definitive implants and are supposed to be removed after a few months of loading.

In this case report, three transitional implants (two immediately loaded and one unloaded) were retrieved after 3 months and analyzed using micro-CT and traditional histologic examinations.

Method and materials

Case presentation

A nonsmoking, healthy 63-year-old woman with a mandibular complete denture was referred to the Department of Surgery and Stomatology, University of Bari, Bari, Italy, to receive an implant-supported prosthetic rehabilitation. The patient accepted treatment with an implant-supported overdenture. Therefore, the patient consented to the placement of two submerged implants that would be used for the definitive rehabilitation after a free-load healing period and placement of three transitional implants, two of which were immediately loaded and one of which was left unloaded, which would be removed after 3 months. The immediately loaded transitional implants were used to support the old denture with the goal of improving prosthesis stability. Informed consent was obtained after explaining the treatment plan, and the clinical study was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2000.

The patient showed good oral hygiene and no systemic diseases or use of medication that contraindicated oral surgical procedures. Complete clinical and radiographic examinations were carried out to obtain a treatment plan of the future rehabilitation. Edentulous sites scheduled for implant-supported rehabilitation were treated conventionally. Briefly, two submerged implants (Hex 3.3 Threaded Implant,
IntraLock) were placed in the mandibular lateral incisor sites according to the manufacturer’s protocol. Transitional implants (3.0-mm MILO Fine Pitch Implants, IntraLock) were placed for temporary retention of the old denture at the mandibular right central incisor and left first premolar sites. Another identical transitional implant was left unloaded in the mandibular left central incisor site. All implants were placed with 32 Ncm of torque and achieved primary stability. The bone quality at insertion was normal (D2/D3) for all implants placed. Soft tissues were sutured over the two submerged implants and around the three protruding heads of the transitional implants. Following placement, implants at the right central incisor and left first premolar sites were loaded and the old denture was relined. Amoxicillin and clavulanic acid (2 g/day for 5 days) were prescribed, and chlorhexidine mouthwash was suggested for 4 weeks. Care was taken to avoid any pressure on the unloaded transitional implant. No special dietary recommendations were provided to the patient, who was recalled after 1, 4, 8, and 12 weeks for clinical and radiographic follow-up.

After 12 weeks of healing, a gingival flap was raised. The submerged implants received standard abutments were checked for clinical mobility, whereas the transitional implants were removed. The implant at the right central incisor site was lost after 12 weeks during clinical follow-up because of progressive bone loss, while the implants at the left central incisor and first premolar sites were retrieved with the surrounding bone using a 5-mm-diameter trephine (Bontempi). Soft tissues were sutured, and standard prosthetic procedures were carried out up to definitive prosthesis delivery.

Sample preparation

After retrieval, bone biopsy specimens were rinsed in saline and fixed with neutral-buffered formalin. The specimens were dehydrated in an ascending series of alcohol rinses and then embedded in acrylic resin. After polymerization, they were analyzed using a micro-CT scanner (micro-CT-20, Scanco Medical) and subsequently sent to the laboratory of the Biomaterial Clinical and Histological Research Association, Pescara, Italy, to be processed for standard undecalcified histology.

Micro-CT processing

Specimens were scanned with the high-resolution micro-CT system in multislice mode. Each 3D image dataset consisted of approximately 400 micro-CT slice images (1,024 × 1,024 pixels with 16-bit gray levels). Specimens were scanned in high-resolution mode with an x, y, and z resolution of approximately 20 μm. The voxel size was 15 × 15 × 15 μm³. Scanning time for each specimen was approximately 4 hours. Micro-CT measurement of the bone-implant contact (BIC) was obtained working on the threshold of the gray levels. After scanning, the 3D dataset was segmented using two different thresholds for bone and titanium to separate the different materials. The threshold values were determined by analyzing the gray-level distribution (histogram analysis) and picking up the intermediate gray-level value between the two peaks of the materials to be distinguished.

Histologic processing

After micro-CT scanning of the samples was completed, specimens were processed to obtain thin ground sections. They were sectioned at 200 to 250 μm using a Micromet high-speed rotating-blade microtome (Buehler) and subsequently ground to approximately 40 to 50 μm using an LS2 grinding machine (Remet). The histologic slides were routinely stained with toluidine blue and basic fuchsin. Three sections were produced for each specimen. The histomorphometric analysis was performed by digitizing the images from the microscope using a JVC TK-C1380 Color Video Camera (JVC Victor) and a frame grabber. Subsequently, the digitized images were routinely analyzed using image analysis software IAS 2000 (Delta Sistemi). The images were acquired with a 5X objective all around the implant surface.
Morphometric measurements

The morphometric parameters obtained with micro-CT included bone volume, BIC, and connectivity density. Micro-CT measurements were obtained as a mean of 400 2D sections of the analyzed sample.

Results

Clinical findings

Soft tissue healing occurred uneventfully from the first surgical phase to the uncovering of the definitive implants (Fig 1). The transitional implants did not disturb the osseointegration of the definitive implants, and the patient reported no pain or discomfort with the immediate provisional prosthesis. No bone resorption was observed at exposure of the definitive implants. However, both loaded implants (right central incisor and left first premolar sites) showed signs of progressive bone loss without pain or any sign of inflammation. Periapical radiographs showed that the two immediately loaded implants had deep infrabony defects (Fig 2). After 12 weeks, the implant at the right central incisor site was mobile and was removed unintentionally when testing its stability. The retrieval procedure of the remaining loaded implant in the left first premolar site was difficult because the trephine bur came up against hard bone and the implant body. Subsequently, strong traction with forceps was needed to remove the implant. The unloaded implant at the left central incisor site did not show any sign of bone loss, and the bone quality during removal was perceived as normal. The bone defects consequent to biopsy healed uneventfully and were spontaneously filled with bone.

Histologic and micro-CT findings

Unloaded transitional implant

Bone volume, as measured using micro-CT, was 37.03%. Micro-CT 2D analysis showed a bone shell surrounding most of the implant surface in connection with thick trabeculae (Fig 3a) and a high BIC (Fig 3b). By means of 3D analysis, the mean BIC was recorded (73.19%) and visualized using the subtraction technique (Figs 3c and 3d, Table 1). Three-dimensional sections showed a thick cortical bone layer coronally in contact with the implant collar, while large marrow spaces were identified more apically around the implant surface. Thick trabeculae that were horizontally oriented like overlapped plates with infrequent oblique interconnections were in contact with a bone shell surrounding most of the implant. Thus, the surface of the implant seemed to exert a positive effect on the amount of bone facing the implant body, which could be described as conductive (Fig 3e). Histologic analysis confirmed a high BIC and an almost continuous thin layer of bone covering most of the implant surface, surrounded by large marrow spaces (Fig 3f).
Fig 3a  Cross-sectional micro-CT 2D image of the unloaded transitional implant. Bone can be seen surrounding the implant surface, which is connected by a few thin horizontal trabeculae to the denser bone.

Fig 3b  Micro-CT 2D analysis (longitudinal sections) of an immediately loaded transitional implant. The implant surface was in contact with mineralized bone (red) and with soft bone marrow (green).

Fig 3c  Micro-CT 3D reconstruction of the entire biopsy specimen. The BIC value was evaluated using the 3D micro-CT subtraction technique. Red = BIC; green = lack of BIC.

Fig 3d  BIC of the unloaded implant as calculated using micro-CT. A high grade of osseointegration was noted in the area of the implant collar as well as at the implant surface. The implant showed the same degree of BIC along the implant axis (yellow line).

Fig 3e  Higher magnification with micro-CT allowed better identification of the anatomy of bone plates (orange) and marrow spaces (blue).

Fig 3f  Histologic analysis showing the BIC obtained at the transitional unloaded implant.

### Table 1  Morphometric measurements obtained with micro-CT

<table>
<thead>
<tr>
<th></th>
<th>Total BIC (%)</th>
<th>BIC above socket area (%)</th>
<th>BIC below socket area (%)</th>
<th>BV (%)</th>
<th>CD (1/mm³)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Loaded</td>
<td>80.75</td>
<td>4.42</td>
<td>89.46</td>
<td>51.75</td>
<td>61.17</td>
</tr>
<tr>
<td>Unloaded</td>
<td>73.19</td>
<td>No socket</td>
<td>No socket</td>
<td>37.03</td>
<td>40.68</td>
</tr>
</tbody>
</table>

BIC = bone-implant contact; BV = bone volume; CD = connectivity density.
Loaded transitional implant

Micro-CT analysis showed that bone tissue was slightly detached from the implant surface. A deep and narrow bone defect around the coronal part of the implant was evident (Fig 4a). It was possible to virtually settle the sample to calculate the BIC, which was approximately 80.75%. BIC was 4.42% in the apical part of the implant.

The International Journal of Periodontics & Restorative Dentistry
Discussion

This case report documented impressive bone loss around two immediately loaded small-diameter implants, while another identical implant inserted in a comparable position but left unloaded showed absence of bone resorption. The bone loss observed around the loaded transitional implants could be caused by several factors, such as erroneous technique for implant placement, infection, lack of primary stability, and overload. The hypothesis of an erroneous osteotomy during implant site preparation was discarded since the handpieces employed and the technique chosen for implant placement were the same for all transitional implants. Moreover, all implants were inserted in the anterior mandible, where homogenous bone quality is often found. All transitional implants were placed with a high insertion torque (30 to 35 Ncm). The implant insertion torque is a value commonly used to assess implant primary stability, which was very good at the time of placement and at the 12-week follow-up evaluation. Another striking observation was that no clinical or histologic signs of infection were observed. All data supported the hypothesis that the progressive bone loss at loaded implants was a result of overload. Indeed, implants were stable and clinically osseointegrated, but loading conditions were unfavorable: There were only two loaded implants and a full functional occlusal load was applied immediately. Implants were short and narrow, and for this reason, applied loads were concentrated around a reduced bone-implant interface compared to standard implants. Additionally, the bone quality at implant insertion was normal, and for this reason, the BIC expected at implant placement was lower compared to that attainable in hard bone. It is conceivable that this demanding biomechanical condition could lead to peri-implant bone damage caused by bone fatigue. Studies dealing with immediate loading of dental implants and bone biomechanics showed that bone remodeling depends on continuous adaptation to functional loading and repair of damage subsequent to overload at the bone-implant interface. Orthopedic literature on bone biomechanics has reported that applied functional load over the thresholds of bone resistance can cause microdamage. Bone microdamage and trabecular bone repair were also observed histologically and described in the literature but in a controlled loading condition. Moreover, it should be noted that failure of poorly osseointegrated implants occurred during the first 12 months of function. Surprisingly, a higher BIC was registered around the apical portion of the overloaded implant, and bone quality seemed to be improved in only 12 weeks. This finding supports previous studies that showed how the density of peri-implant bone can be modified, even in a short period, if stimulated by adequate loading conditions.

Biomechanical competence of trabecular bone is dependent not only on the absolute amount of bone present but also on the trabecular microstructure. For this purpose, connectivity density was measured even if it did not provide any additional information on peri-implant bone behavior beyond that provided by measurements of bone mass and BIC. Despite the limitations of this report, micro-CT may be considered a nondestructive, fast, and precise procedure that allows the measurement of morphometric indices in unprocessed biopsy specimens. These indices could be helpful in understanding the mechanisms and timing of bone loss in failing implants. Micro-CT helped discriminate between bone and soft tissue at the interface to precisely calculate BIC percentage, and since it is a nondestructive method, it could be used in addition to histology for this purpose. In this report, BIC was determined by means of micro-CT without using standard histology. Even if micro-CT is more precise on 3D measurements since it allows calculation of morphometric parameters on a large number of sections, standard histology has a higher resolution and image contrast. Differences between the bone morphometric values obtained histologically and with micro-CT were reported. This was ascribed to the limitation of micro-CT in measuring only mineralized tissues over a certain threshold of mineralization. On the contrary, histomorphometry can also measure
minimally mineralized or nonmineralized bone tissues, such as osteoid and certain degrees of development of woven bone. Another possible explanation was thought to be the histologic technique used for hard biomaterial evaluation, which allows evaluation of a restricted number of sections, whereas micro-CT assessment allows the automatic evaluation of a large number of 2D sections and a complete 3D assessment.\textsuperscript{18}

Conclusions

Immediately loaded small-diameter implants showed severe bone loss in the coronal portion, but the density of bone tissue and the BIC were higher in the apical portion in comparison to the unloaded implant. This pattern could represent a bone reaction to more demanding biomechanical conditions. Even if denser bone could improve implant stability, this situation could be harmful since deep peri-implant bone loss can cause infiltration of pathogenic bacteria in addition to augmentation of rigidity and a lengthening of the crown-implant ratio, thus causing unfavorable levers leading to implant fracture. Further studies are needed to confirm these observations and to improve knowledge about the mechanical causes of peri-implant bone loss.

References


