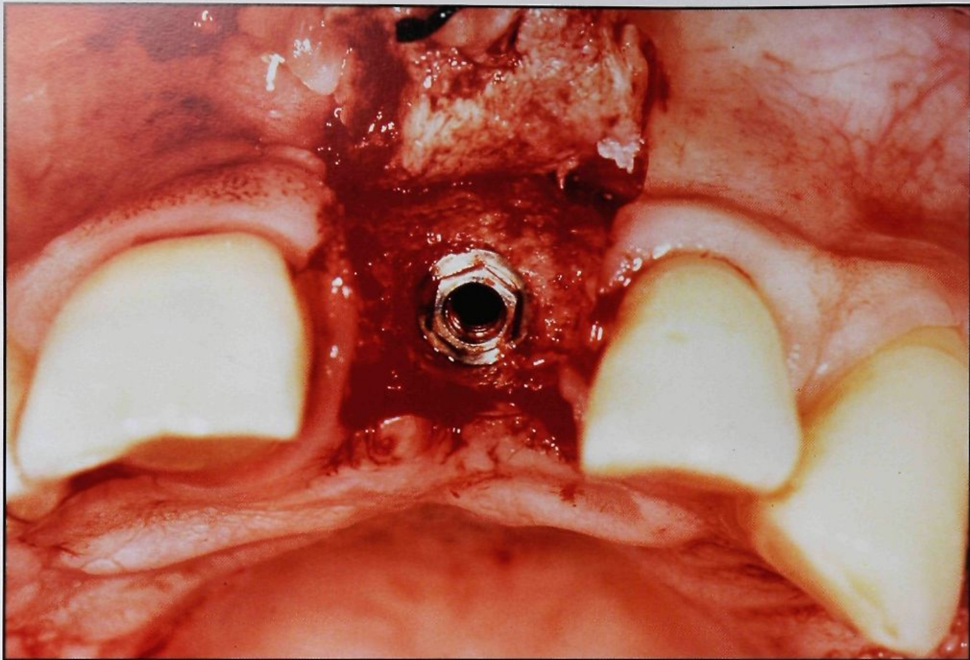
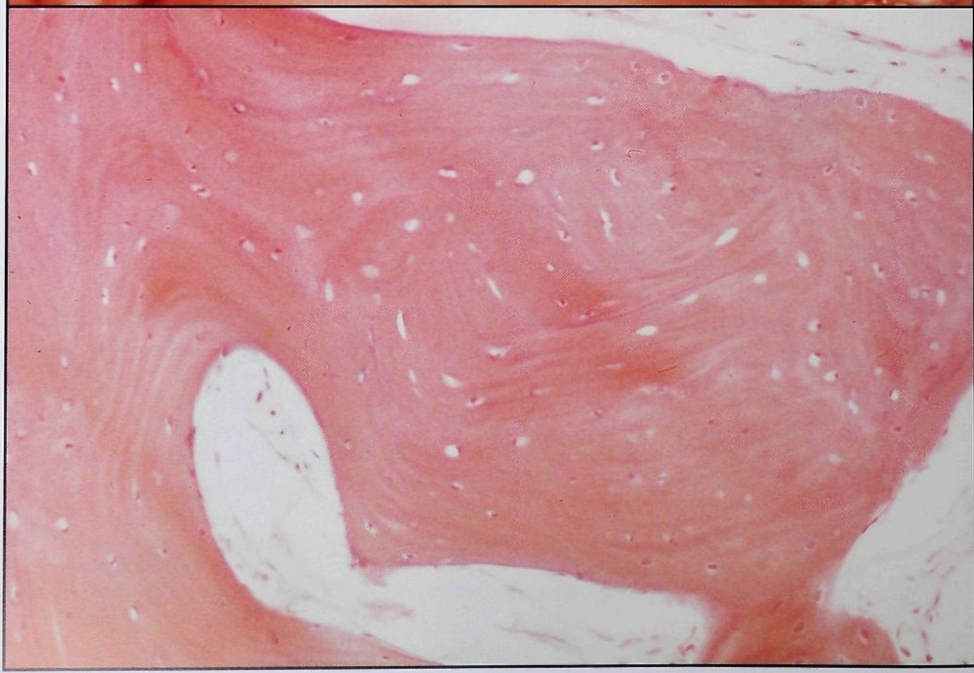
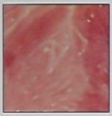




GBR in Human Extraction Sockets and Ridge Defects Prior to Implant Placement: Clinical Results and Histologic Evidence of Osteoblastic and Osteoclastic Activities in DFDBA



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This study evaluated new bone formation in 3 types of osseous defects following treatment with demineralized freeze-dried bone allografts (DFDBA) and cell-occlusive membranes. For 8 patients electing to receive implant treatment, a distinction was made among 3 clinical situations: (1) existing alveolar ridge defects; (2) extraction sockets with lost buccal plate; and (3) extraction sockets with an intact alveolus. Implants were placed a mean of 6 months after the regenerative procedure. Clinical examination of bone width and height at the time of implant placement showed sufficient augmentation or preservation, and implants were inserted without incident. Histologic examination of hard tissue biopsies obtained from the implant sites revealed no discernible differences among the 3 types of defects. Specifically, all sites demonstrated DFDBA particles surrounded by woven or lamellar bone. No fibrous encapsulation of DFDBA or inflammatory reaction was observed. Osteoblasts were found lining marrow spaces. Howeship's lacunae, with and without resident osteoclasts, were clearly seen in several DFDBA particles; this finding supports the belief that DFDBA undergoes osteoclasis in vivo. These results demonstrate that commercially available DFDBA has osteoconductive properties that lead to appositional new bone growth in both self-contained and non-self contained osseous defects. (Int J Periodontics Restorative Dent 1999;19:259–267.)

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Human demineralized freeze-dried bone allografts (DFDBA) are widely used in periodontal regeneration procedures.[1-8] In addition, DFDBA is used in the preservation of extraction sockets and in the repair of resorbed alveolar ridges to provide sufficient quantity of bone for the placement of endosseous implants.[9-17] In these various clinical situations, DFDBA is believed to act as a space-maintaining agent and also as a bone-growth promoter.

Although the combined use of DFDBA and cell-occlusive membranes has been associated with new bone growth in alveolar sockets following tooth extraction,[9,11,13,14] the therapeutic value of DFDBA as a graft material has come under scrutiny.[18] Furthermore, the question of whether intact extraction sockets are suitable sites to test for bone-growth activity of DFDBA has been raised.[19] Such extraction sockets, it can be argued, are self-healing and would likely

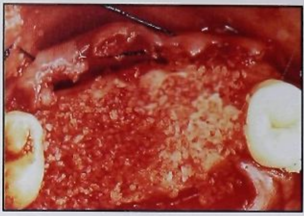

Fig 1a (left) *Socket preservation of the maxillary right first and second molar alveoli in 28-year-old white man. Full-thickness mucoperiosteal flap is reflected and molars are sectioned and extracted atraumatically. Sockets are thoroughly debrided to remove residual granulation tissue.*

Fig 1b (right) *Commercially available DFDBA is placed into the sockets until a slight overfilling is achieved, and then covered by an oval # 9 nonresorbable e-PTFE titanium-reinforced membrane (not shown).*

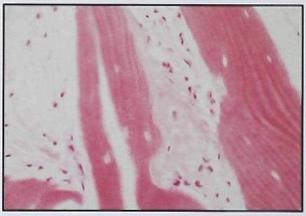

Fig 1c (left) *DFDBA particles are intimately associated with woven and lamellar bone. (Original magnification × 50; hematoxylin-eosin stain.)*

Fig 1d (right) *Higher mangnification of DFDBA fragments demonstrates a focus of woven bone with osteoblastic rimming and Howeship's lacunae. There is a notable lack of inflammation within the mildly fibrotic marrow. (Original magnification × 100; hematoxylin-eosin stain.)*

heal with or without the grafting procedure.

In contrast, non-self healing defects would be more suitable sites to test the bone-growth potential of DFDBA. These defects include fresh extraction sockets without buccal plates or ridge defects resulting from prior extractions. Therefore, the purpose of this investigation was to compare the clinical and histologic results of DFDBA and membrane treatment among self-contained extraction sockets, non-self contained extraction sockets, and ridge defects.

Method and materials

Patient selection

Eight patients seeking treatment in the Postgraduate Periodontology Clinic at Tufts University School of Dental Medicine participated in this study. The group consisted of 3 women and 5 men, ranging

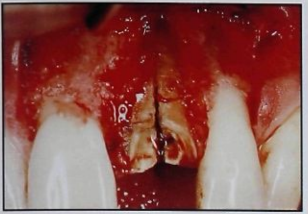

Fig 2a *Socket augmentation of maxillary left central incisor alveolus in 59-year-old white man. Loss of the buccal plate and vertical fracture of the root is evident.*

Fig 2b *To reshape the lost profile of the ridge DFDBA is packed as described in the methods section.*

Fig 2c *Oval # 4 nonresorbable e-PTFE membrane is trimmed and positioned over the buccal and coronal aspects of the socket, extending ≥ 2 mm in all directions onto sound bone, with care taken to avoid contact with the adjacent teeth.*

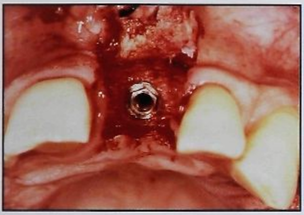

Fig 2d *Standard-diameter, 15-mm long implant is placed ad modum Brånemark 7 months after extraction.*

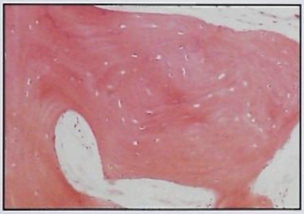

Fig 2e *Fragments of DFDBA are seen adjacent to woven and lamellar bone with interposed cement (reversal) lines. The marrow demonstrates mild fibrosis. (Original magnification × 66; hematoxylin-eosin stain.)*

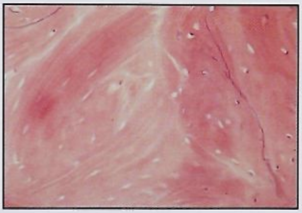

Fig 2f *Higher magnification clearly demonstrates intimate apposition of DFDBA (empty lacunae, center left) and new bone (cell-filled lacunae, center right). Striking presence of cement (reversal) lines is also evident. (Original magnification × 80; hematoxylin-eosin stain.)*

from 24 to 65 years of age (mean 42 years). All patients were medically healthy with no underlying systemic disease, as assessed by medical history screening. All patients exhibited good oral hygiene and had no contraindications for dental treatment. Informed consent was obtained prior to treatment.

Based on clinical and radiographic findings, each patient had more than 1 hopeless tooth to be extracted and/or a resorbed edentulous ridge secondary to previous extraction. All patients chose to have the extracted or missing teeth replaced by endosseous implants, following preimplant bone augmentation procedures. The distribution of sites was: *(1)* 4 extraction sockets in which the alveolar plates were intact (socket preservation, Fig 1); *(2)* 3 extraction sockets in which the buccal plate was missing (socket augmentation, Fig 2); and *(3)* 2 edentulous ridges with Seibert Class I buccolingual defects (ridge augmentation, Fig 3).[20]

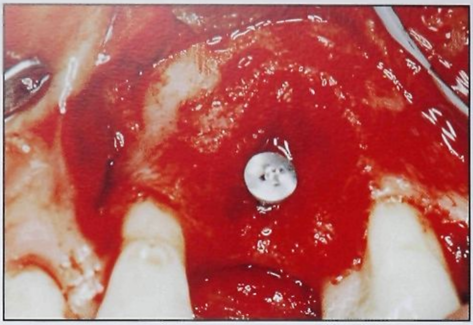

Fig 3a Ridge augmentation of the maxillary left canine alveolus in a 50-year-old white man. Missing maxillary left canine had been extracted by a private practitioner several months prior to the augmentation procedure. After careful debridement of the defect, an OsteoMed titanium screw of 1.6-mm diameter and 6-mm length is positioned perpendicular to the buccal plate in the center of the defect to support and tent the overlying cell-occlusive membrane.

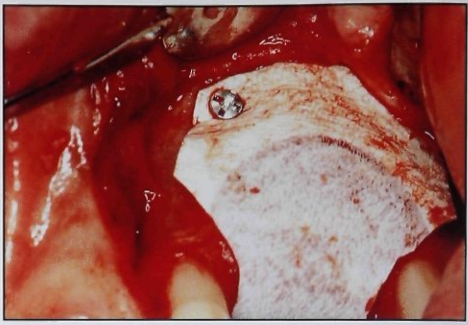

Fig 3b Oval # 6 nonresorbable e-PTFE membrane is fixed to the bone surface with an OsteoMed fixation screw of 1.2-mm diameter and 4-mm length. Complete soft tissue closure is achieved.

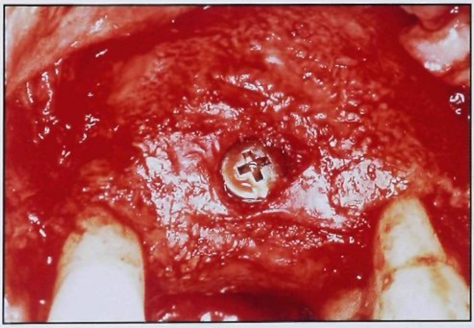

Fig 3c After surgical exposure and removal of the membrane and titanium screw, clinical evaluation shows complete regeneration of the defect. A standard-diameter Brånemark implant 15 mm in length has been placed.

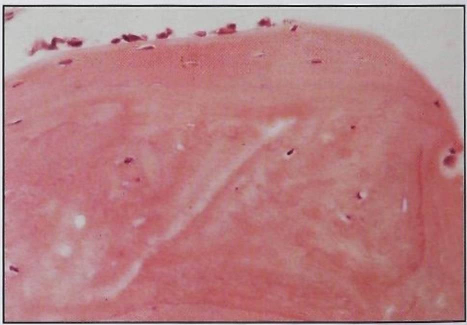

Fig 3d Fragment of DFDBA (far right) shows the presence of an osteoclast within a Howeship's lacuna juxtaposed with woven bone separated by a prominent cement (reversal) line. Note rim of osteoblasts adjacent to lamellar bone. (Original magnification × 66; hematoxylin-eosin stain.)

Surgical technique

Socket preservation and socket augmentation: Bone grafting in self-contained and non-self contained sockets. Under local anesthesia, a sulcular incision was made on the facial aspect beginning one tooth mesial and extending one tooth distal to the tooth to be extracted. Vertical releasing incisions were placed at the mesial and distal line angles of the approximating teeth (Figs 1a and 2a). A full-thickness mucoperiosteal flap was reflected to expose the tooth to be extracted and the adjacent teeth. A lingual sulcular incision beginning one tooth before and extending one tooth beyond the tooth to be extracted was also made, and the flap was released and reflected. The hopeless tooth was extracted atraumatically. The socket was thoroughly debrided to remove residual granulation tissue. The status of the ridge was then recorded by intraoral photography. Intrasocket bleeding was promoted by intramarrow penetration prior to placement of the graft material. Commercially available DFDBA, 250 to 710 µm particle size (LifeNet Transplant Service) was rehydrated with sterile saline 30 minutes prior to the procedure. To facilitate handling of the graft material, a sterile disposable syringe was used to apply bone into the socket. The bone was then manually compressed several times, with a spatula and saline-wetted gauze, to minimize dead spaces between the DFDBA particles. This process was repeated until a slight overfilling of the socket was achieved (Figs 1b and 2b). In the socket augmentation cases, particular care was taken to reshape the lost buccal profile (Fig 2b).

A nonresorbable expanded polytetrafluoroethylene membrane (e-PTFE; Gore-Tex Augmentation Material, 3i/WL Gore) of suitable size was then positioned over the buccal and coronal aspects of the socket, extending ≥ 2 mm in all directions onto sound bone (Fig 2c), with care taken to avoid contact with adjacent teeth. Nonresorbable, interrupted e-PTFE sutures were used to approximate flap margins; the buccal flap was further released when necessary to ensure primary closure.

Ridge augmentation: Bone grafting in the edentulous ridge. After administration of an appropriate local anesthetic, initial incisions were made according to the surgical approach used. In the maxilla, a palatal incision beveled to the midcrest on the edentulous area was performed. In the mandible, a buccal approach was used. Vertical releasing incisions were placed at the mesial and distal line angles of the approximating teeth; in a contiguous edentulous area the incisions were placed at a reasonable distance from the anticipated edges of the membrane. A full-thickness mucoperiosteal flap was reflected to expose the defect. The status of the ridge was recorded by intraoral photography. Titanium screws 1.6 mm in diameter (OsteoMed) of appropriate length were selected and positioned in the center of the defect, perpendicular to the buccal aspect of the residual bone. This maintained space and helped support the overlying membrane (Fig 3a). The DFDBA was handled in the manner described above and was carefully packed around the central screw, reshaping the lost alveolar ridge. Also as described above, a nonresorbable e-PTFE membrane of suitable size was then trimmed and positioned over the defect. The membrane was then fixed to the bone surface with titanium fixation screws 1.2 mm in diameter (OsteoMed) (Fig 3b). Primary closure of flaps was achieved with nonresorbable, interrupted e-PTFE sutures.

Postsurgical care

All patients received doxycycline, 100 mg daily, for 3 weeks postsurgical. Sutures were removed 2 weeks postsurgical. During the first month after graft and membrane placement, patients were seen weekly and chlorhexidine gluconate (Peridex, Proctor & Gamble) was prescribed 2 times a day to enhance plaque control. All membranes were kept in place for at least 6 weeks postsurgical. Membranes were removed using techniques described above. Briefly, the patient was anesthetized and a full-thickness mucoperiosteal flap was elevated. The e-PTFE membranes were then carefully separated from the underlying tissue and the flaps were positioned and sutured. Routine postoperative care was provided. The healing was normal and unremarkable.

Bone core sampling and histology

Endosseous implants were inserted a mean of 6 months after the graft and membrane placement (range ≈ 3 to 9 months). At the time of implant surgery, the patients again received an appropriate local anesthetic followed by elevation of a full-thickness mucoperiosteal flap. Preservation and/or augmentation of ridge height and width, as well as localization of the grafted sites, was established intraoperatively by comparison with initial photographs and radiographs (data not shown).

As part of the implant site preparation, a surgical trephine 2 mm in net diameter (Implant Innovations) was used to harvest 6 mm × 2 mm bone cores, which were immediately fixed in 10% formalin. Following completion of implant site preparation, the implants were inserted and the flaps were repositioned and sutured. The sutures were removed after 2 weeks and postoperative healing was uneventful.

Core samples were obtained from 9 sites in the 8 patients. Approximately 3 months after grafting, 2 core biopsies were obtained from 2 different socket preservation sites in the same patient. Another socket preservation biopsy was obtained from one patient approximately 5 months after grafting. One socket augmentation biopsy from one patient and one ridge augmentation biopsy from another patient were obtained 6 months after grafting. One socket augmentation core was harvested at 7 months (from one patient), and the 3 remaining biopsies were collected at 9 months (one socket preservation, one socket augmentation, and one ridge augmentation core from 3 different patients). All core samples were decalcified by ≤ 3 hours incubation in 100 mL of a standard decalcifying solution consisting of disodium ethylenediaminetetraacetic acid (EDTA), potassium tartrate, and diluted hydrochloric acid in distilled water, pH ≤ 1 (RDO solution; DuPage Kinetic Laboratories). Sections were cut to a thickness of 6 µm, stained with hematoxylin-eosin, and examined using light microscopy.

Results

Clinically, sufficient bone was present in every case to allow the placement of implants completely into bone (Figs 2d and 3c). Histologically, a total of 48 sections were obtained from the 9 core biopsies, with the following distribution: 3 sections each from the 2 biopsies taken at 3 months; 9 sections from the biopsy taken at 5 months; 10 sections from the 2 biopsies taken at 6 months; 11 sections from the biopsy taken at 7 months; and 4 sections each from the 3 biopsies taken at 9 months.

The decalcified sections clearly showed foci of DFDBA particles surrounded by woven and lamellar bone (Figs 1c, 1d, 2e, 2f, and 3d). Distinct cement lines were evident at the interface between DFDBA and new bone. The lacunae of DFDBA were acellular, whereas osteocyte-containing lacunae were prevalent within the new bone. Osteoblasts were seen lining endosteal spaces and the new bone marrow exhibited a mild degree of fibrosis without signs of inflammatory reaction. Evidence of osteoclastic activity on DFDBA surfaces was also seen in several sections (Figs 1d and 3d). There was, in addition, a notable lack of fibrous encapsulation of the allografts.

Discussion

Periodic assessment of treatment outcomes is the hallmark of any clinical profession. Relative to the present report, it is noteworthy that the role of DFDBA in periodontics has recently been challenged.[18] This has led to reevaluation not only of osseous graft materials, but also of intraoral test sites. In particular, Mellonig and Towle[19] have questioned whether intact extraction sockets are appropriate sites to test bone growth-promoting activities of any graft material. They postulate that only 2 responses can occur in such self-healing defects: either the graft material has no influence on the natural healing of the socket, or the graft material impairs the healing process. Consistent with the former possibility, test and control sites would give the same final results. Consistent with the latter possibility, test sites would likely heal slower than control sites, with or without clinically worse outcomes. Within this conceptual framework, therefore, osseous grafting of intact extraction sockets may help maintain space but does not necessarily enhance bone fill.

Thus, non-self healing defects can be considered more suitable test sites. For this reason we examined 3 different clinical situations: *(1)* extraction sockets in which the alveolus was intact (socket preservation); *(2)* extraction sockets in which the alveolus was altered by a pathologic or traumatic event, such as loss of buccal plate after extraction (socket augmentation); and *(3)* Seibert Class I[20] edentulous ridge defects in which the buccolingual osseous dimension was altered by a previous pathologic or traumatic event (ridge augmentation). The latter 2 clinical situations share features that distinguish them from intact extraction sockets. In all 3 defect types, however, we expected DFDBA to act as a space maintainer underneath the cell-occlusive membranes, and to serve as a scaffold for migrating host osteogenic cells (ie, osteoconduction).

From a clinical standpoint, sufficient osseous width and height were present after the preservation and augmentation procedures to allow implant placement in all 8 patients. Histologically, no differences in new bone formation were seen among the 3 classes of defects. Individual particles of DFDBA were discernible up to 9 months in situ. In all samples, DFDBA was well incorporated within new bone, which exhibited osteocyte-containing lacunae. The presence of osteoblasts and osteoclasts was also noted. As expected, the basic healing processes were similar and independent of the osseous topography seen clinically.

The present report confirms and extends previous findings by the authors.[9] In that earlier study we evaluated self-healing extraction sockets treated with DFDBA and found results similar to those reported here. Moreover, our observations now include non-self healing defects such as sockets with no buccal plate and edentulous ridge defects. These results demonstrate that DFDBA has osteoconductive properties that are useful for implant site development in a variety of clinical situations.

Acknowledgment

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