PERIODONTAL REGENERATION

CONTENTS:

* INTRODUCTION
* HISTORY
* DEFINITIONS

INTRODUCTION

Intrabony and furcation defects are sequelae of periodontal disease. Ideally, these defects are managed in a timely fashion through periodontal regeneration.

In the past, the results of regenerative therapy were inconsistent and unpredictable.

The current status of regenerative therapy has dramatically changed and improved due to research and a better understanding of the biology of the tissues that comprise the periodontal attachment.

The various surgical approaches including bone replacement grafts, guided tissue regeneration (GTR), and a better understanding of biologic mediators and tissue engineering have improved the predictability of regeneration as another choice for therapy.

When the periodontium is damaged by inflammation or as a result of surgical treatment, the defect heals either through periodontal regeneration or repair.

DEFINITION

In periodontal regeneration, healing occurs through the reconstitution of a new periodontium, which involves the formation of alveolar bone, functionally aligned periodontal ligament, and new cementum.

Repair due to healing by replacement with epithelial and/or connective tissue that matures into various nonfunctional types of scar tissue is termed new attachment.

Histologically, patterns of repair include long junctional epithelium, ankylosis, and/or new attachment. Although the stability of periodontal repair is not clear, the ideal goal of periodontal surgical therapy is periodontal regeneration.

Today, several highly reproducible regenerative approaches are used, as evidenced by clinical attachment gain, decreased pocket probing depth, radiographic evidence consistent with bone fill, and overall improvements in periodontal health. These clinical improvements can be maintained over long periods (>10 years).

RECONSTRUCTIVE SURGICAL TECHNIQUES

* Reconstructive techniques can be subdivided into three major therapeutic approaches:
  1. non–bone graft–associated,
  2. graftassociated, and
  3. biologic mediator–associated new attachment and regeneration.
* All recommended techniques include careful case selection and complete removal of all irritants on the root surface.Although this can be done in some cases as a closed procedure, in most cases it should be done after exposure of the area with a flap.

NON–GRAFT-ASSOCIATED RECONSTRUCTIVE PROCEDURES

* New attachment or periodontal bone regeneration in response to non– graft-associated reconstructive surgical therapy is used in Europe and Asia where human bone graft is not available due to regulatory restraints.
* Of these procedures, GTR is the main procedure used in clinical practice.
* More recent evidence suggests that the laser-assisted new attachment procedure (LANAP) may also result in new attachment and regeneration, but further clinical trials are needed to test its efficacy and parameters for success.
* Several procedures are of historical interest:

(1) the removal of the junctional and pocket epithelium;

(2) the prevention of their migration into the healing area after therapy;

(3) clot stabilization, wound protection, and space creation; and

(4) biomodification of the root surfaces.

* Although these procedures are not used individually as reconstructive approaches, some of these strategies are currently incorporated into reconstructive surgery as adjuncts.

GUIDED TISSUE REGENERATION

* GTR is used for the prevention of epithelial migration along the cemental wall of the pocket and for maintaining space for clot stabilization.
* Derived from the classic studies of Nyman, Lindhe, Karring, and Gottlow, this method is based on the assumption that periodontal ligament and perivascular cells have the potential for regeneration of the attachment apparatus of the tooth.
* GTR consists of placing barriers of different types (membranes) to cover the bone and periodontal ligament, thus temporarily separating them from the gingival epithelium and connective tissue.
* Excluding the epithelium and the gingival connective tissue from the root surface during the postsurgical healing phase not only prevents epithelial migration into the wound but also favors repopulation of the area by cells from the periodontal ligament and the bone.
* In the United States, GTR is often performed with some type of bone graft as a scaffolding agent, so it is a combined therapy.
* As indicated earlier, in Europe and in other parts of the world, because of regulatory and religious constraints, human graft materials are not available, so it is performed as a traditional GTR procedure and may be occasionally used in conjunction with other graft materials as combined therapy.
* Initial animal experiments using Millipore filters (Millipore Sigma, Burlington, MA) and Teflon membranes resulted in regeneration of cementum and alveolar bone and a functional periodontal ligament.
* Clinical case reports indicate that GTR results in a gain in attachment level.
* Histologic studies in humans provided evidence of periodontal reconstruction in most cases, even with horizontal bone loss.
* The use of polytetrafluoroethylene (PTFE) membranes has been tested in controlled clinical studies in mandibular molar furcations and has shown statistically significant decreases in pocket depths and improvement in attachment levels after 6 months, but bone level measurements have been inconclusive.
* A study of maxillary molar furcations did not result in significant gain in attachment or bone levels.
* Problems such as membrane exposure, which resulted in no or limited regeneration and the need for a secondary procedure for surgical removal, resulted in the development of biodegradable membranes.
* Today in clinical practice, most GTR procedures use biodegradable membranes, whereas the nonresorbable membranes, especially those with titanium reinforcement struts, are used for regeneration of large intrabony defects and implant site development.
* Nevertheless, the historical research using nonresorbable membranes and the development of various types of biodegradable membranes are valuable.

NONRESORBABLE MEMBRANES

* In classic animal and human studies demonstrating the efficacy of GTR, cellulose acetate filters were used.
* As this technique became more prevalent, the first commercial membrane was produced from expanded PTFE (ePTFE).
* This membrane has all the properties necessary for GTR barriers in that it

(1) is a cellular barrier,

(2) is biocompatible,

(3) provides space for the healing tissue,

(4) permits tissue integration, and

(5) is clinically manageable.

* Much of our current understanding of GTR is based on studies using ePTFE membranes. Although these membranes are used less frequently now, they are still popular for guided bone regeneration and ridge preservation, so it is important to understand the clinical procedures for managing these membranes.
* The clinical effectiveness of ePTFE membranes is dependent on technique.
* Preservation of the keratinized gingiva and a relatively thick overlying surgical flap are critical to avoid perforation of the flap by the membrane during healing.
* After the surgical area has been flapped, the defect is degranulated and the root surface scaled and root planed. The ePTFE membrane is trimmed to adapt to tooth configuration, secured by ePTFE sutures, and the flap is repositioned.
* Although much of the emphasis in the literature is on adapting the membrane to the defect, no membrane can ever be perfectly adapted. Despite gaps, these membranes appear to be successful. After membrane placement, healing is allowed to proceed for 4 to 6 weeks.
* Barring any membrane exposure, a second surgery is performed to remove the membrane. During this removal, the healing tissue appears reddish and granulomatous.
* After membrane removal, the area should not be probed for 3 months.
* Radiographic evidence of bone fill is usually present after 6 months and should continue over the course of 1 year.
* Clinical studies have shown that ePTFE membranes used in GTR procedures are more effective than surgical debridement in correcting intrabony defects.
* In intrabony and furcation defects, gains are made in clinical attachment level (3 to 6 mm), improved bone levels (2.4 to 4.8 mm), and probing depth reductions (3.5 to 6 mm).
* Studies have demonstrated that these regenerative results can be maintained over the course of several years.
* The advent of titanium-reinforced ePTFE allowed for the formation of larger spaces, thus permitting correction of larger defects. This resulted in significant clinical improvements using titanium-reinforced ePTFE compared with ePTFE.
* To determine how regeneration can be enhanced with GTR technique, the prolonged retention of ePTFE membranes was evaluated.
* After allowing the membrane to be retained for 4 months, surgical reentry at 1 year determined that the mean bone fill of intrabony defects was 95%. This suggests that prolonged retention of a barrier membrane is desirable if no tissue perforation is present. This finding is consistent with many clinical reports of the improved bone quality associated with guided bone regeneration in implant site development.
* The major problem with using nonresorbable membranes is that the membrane is exposed to the oral environment during healing. On exposure, the membrane is contaminated and colonized by oral microflora.
* Several studies have shown that contamination of the surgical field can result in decreased formation of new attachment. If the membrane is exposed, the infection can be temporarily managed with topical application of chlorhexidine. This may minimize the infection and extend the time the membrane can be retained in place.

BIODEGRADABLE MEMBRANES

* Bioresorbable membranes have replaced the routine use of ePTFE membranes in GTR.
* There are basically three types of bioresorbable membranes:

(1) polyglycoside synthetic polymers (i.e., polylactic acid, polylactate-polygalactate copolymers), (2) collagen, and

(3) calcium sulfate.

* Polyglycoside membranes degrade as the result of random nonenzymatic cleavage of the polymer, to produce polylactide and polyglycolide, which are converted to lactic acid and pyruvate, respectively, and metabolized by the enzymes of the Krebs cycle.
* Porcine collagen membranes are degraded by collagenases and subsequently by gelatinases and peptidase.
* A resurgence has occurred in the use of calcium sulfate as a regeneration material because it can be used as a pavable resorbable barrier in combination with bone or bone substitutes. The calcium sulfate is bioresorbed through a giant cell reaction.
* Several features make these bioresorbable membranes easier to manage clinically:

(1) they are more tissue compatible than nonresorbable membranes,

(2) the timing for resorption can be regulated by the amount of cross-linkage in the synthetic polymer and collagen membrane or the amount of heat-processed calcium sulfate chips in calcium sulfate barrier, and

(3) a second surgical procedure is not required to retrieve the nonresorbable membrane.

* One GTR study compared the use of bioresorbable membranes (polylactate-polygalactate copolymer) versus ePTFE membranes, with surgical debridement as a control.
* After 1 year, significant gains in clinical attachment level (CAL) were observed in all three groups. There was no difference in CAL gain between the two membrane groups, with both of them gaining 2 mm or more. In both membrane groups, 83% of the sites improved 4 mm or more, which was significantly better than the surgical debridement control group. These findings indicate that GTR procedures are equally effective when using resorbable and nonresorbable membranes. This finding has been confirmed by other investigators.
* A large, multicenter clinical study reported the use of bioresorbable membranes in 203 consecutively treated intrabony defects. After 1 year, investigators found that CAL improved by 79%, and 78% of the sites improved by 4 mm or more. An average of 3 mm of bone fill was measured radiographically. Compromised clinical results occurred when membranes were exposed or patients had poor plaque control.
* The search for resorbable membranes has included trials and tests with numerous materials and collagens from different species such as bovine, porcine, Cargile membrane derived from the cecum of an ox, polylactic acid, polyglactin 910 (Vicryl, Johnson & Johnson Dental Care Division, New Brunswick, NJ), synthetic skin (Biobrane, Smith & Nephew, London, United Kingdom), and freeze-dried dura mater.
* Clinical studies with a mixture of copolymers derived from polylactic acid and acetyl tributyl citrate resorbable membranes (Guidor membrane, no longer on the market) and a poly-D,L-lactide-co-glycolide (Resolut membrane, also no longer on the market) showed significant gains in clinical attachment and bone fill.
* The potential of using autogenous periosteum as a membrane and also to stimulate periodontal regeneration was explored in two controlled clinical studies, one of grade II furcation involvements in mandibular molars and another of interdental defects.
* The periosteum was obtained from the patient's palate by means of a window flap.
* Both studies reported that autogenous periosteal grafts can be used in GTR and result in significant gains in clinical attachment and osseous defect fill.

LASER-ASSISTED NEW ATTACHMENT PROCEDURE:

* The role of laser in periodontal therapy remains controversial.
* Nevertheless, the use of neodymium:yttriumaluminum-garnet (Nd:YAG) to perform surgical LANAPs has been reported for the management of chronic periodontitis, and it can potentially result in new attachment and periodontal regeneration.
* Many questions remain about LANAP. The first refers to the exact mechanism and parameter by which healing by new attachment versus regeneration occurs with LANAP therapy.
* The frequency, consistency, and extent of regeneration have not been defined, nor has this approach been compared with other established regenerative therapies. This comparison, along with other randomized controlled trials, will be needed for meta-analysis to determine whether LANAP is equivalent or superior to other conventional therapy. As with all periodontal therapy, the longterm stability of the regeneration also needs to be explored.

Non–Graft-Associated Procedures of Historical Interest

Removal of Junctional and Pocket Epithelium

* Since the earliest attempts at periodontal new attachment, the presence of junctional and pocket epithelium has been perceived as a barrier to successful therapy because its presence interferes with the direct apposition of connective tissue and cementum, thus limiting the height to which periodontal fibers can insert to the cementum.
* Several methods have been recommended to remove the junctional and pocket epithelium. These include curettage, chemical agents, ultrasonics, lasers, and surgical techniques.

CURETTAGE

* Results of removal of the epithelium by means of curettage vary from complete removal to persistence of as much as 50%.
* Although curettage is not a reliable procedure, occasional bone regeneration does occur.
* Ultrasonic methods, lasers, and rotary abrasive stones have also been used, but their effects cannot be controlled because of the clinician's lack of vision and tactile sense when using these methods.

CHEMICAL AGENTS

* Chemical agents have also been used to remove pocket epithelium, usually in conjunction with curettage.
* The drugs used most often have been sodium sulfide, phenol camphor, antiformin, and sodium hypochlorite.
* However, the effect of these agents is not limited to the epithelium, and their depth of penetration cannot be controlled. These drugs are mentioned here for their historical interest.

BIOMODIFICATION OF ROOT SURFACE

* Changes in the tooth surface wall of periodontal pockets (e.g., degenerated remnants of Sharpey fibers, accumulation of bacteria and their products, disintegration of cementum and dentin) interfere with new attachment.
* Although these obstacles to new attachment can be eliminated by thorough root planing, the root surface of the pocket can be treated to improve its chances of accepting the new attachment of gingival tissues.
* Several substances have been proposed for this purpose, including citric acid, fibronectin, and tetracycline.

Citric Acid.

One in a series of studies applied citric acid to the roots to demineralize the surface, thereby attempting to induce cementogenesis and attachment of collagen fibers.

The following actions of citric acid have been reported:

1. Accelerated healing and new cementum formation occur after surgical detachment of the gingival tissues and demineralization of the root surface by means of citric acid.

2. Topically applied citric acid on periodontally diseased root surfaces has no effect on nonplaned roots, but after root planing, the acid produces a 4-µm-deep demineralized zone with exposed collagen fibers.

3. Root-planed, non–citric acid–treated roots are left with a surface smear layer of microcrystalline debris. Citric acid application not only removes the smear layer, exposing the dentinal tubules, but also makes the tubules appear wider and with funnel-shaped orifices.

4. Citric acid has also been shown in vitro to eliminate endotoxins and bacteria from the diseased tooth surface.

5. An early fibrin linkage to collagen fibers exposed by the citric acid treatment prevents the epithelium from migrating over treated roots.

This technique using citric acid has been extensively investigated in animals and humans. Studies in dogs have shown encouraging results, especially for the treatment of furcation lesions, but the results in humans have been contradictory.

The recommended citric acid technique is as follows:

1. Raise a mucoperiosteal flap and thoroughly instrument the root surface, thus removing calculus and underlying cementum.

2. Apply cotton pledgets soaked in a saturated solution of citric acid (pH of 1.0) for 2 to 5 minutes.

3. Remove pledgets, and irrigate root surface profusely with water.

4. Replace the flap and suture.

The use of citric acid has also been recommended in conjunction with coverage of denuded roots using free gingival grafts.

Fibronectin.

* Fibronectin is the glycoprotein that fibroblasts require to attach to root surfaces.
* The addition of fibronectin to the root surface may promote new attachment.
* However, increasing fibronectin above plasma levels produces no obvious advantages.
* Adding fibronectin and citric acid to lesions treated with GTR in dogs did not improve the results.

Tetracycline.

* In vitro treatment of the dentin surfaces with tetracycline increases binding of fibronectin, which in turn stimulates fibroblast attachment and growth while suppressing epithelial cell attachment and migration.
* Tetracycline also removes an amorphous surface layer and exposes the dentin tubules. In vivo studies, however, have not shown favorable results.
* A human study showed a trend for greater connective tissue attachment after tetracycline treatment of roots.
* Tetracycline gave better results when used alone than when combined with fibronectin.
* Tetracycline has been used as an adjunctive procedure in preparation of the root in regenerative procedures and is a recommended step for use with biologic mediators.

Surgical Techniques

* Surgical techniques have been recommended to eliminate the pocket and junctional epithelium.
* The excisional new attachment procedure (ENAP) consists of an internal bevel incision performed with a surgical knife, followed by removal of the excised tissue.
* No attempt is made to elevate a flap.
* After careful scaling and root planing, interproximal sutures are used to close the wound.
* This approach has been modified and is used in conjunction with the ND:YAG laser in the previously described LANAP procedure.
* Glickman and Prichard advocated performing a gingivectomy to the crest of the alveolar bone and debriding the defect.
* Excellent results have been obtained with this technique in uncontrolled human studies.
* The modified Widman flap, as described by Ramfjord and colleagues, is similar to the excisional new attachment procedure but is followed by elevation of a flap for better exposure of the area. The internal bevel incision eliminates the pocket epithelium.

Preventing or Impeding the Epithelial Migration

* Elimination of the junctional and pocket epithelium may not be sufficient because the epithelium from the excised margin may rapidly proliferate to become interposed between the healing connective tissue and the cementum.
* For experimental purposes, several investigators have analyzed, in animals and humans, the effect of excluding the epithelium by amputating the crown of the tooth and covering the root with the flap (“root submergence”).
* This experimental technique not only excludes the epithelium but also prevents microbial contamination of the wound during the reparative stages.
* Successful repair of osseous lesions in the submerged environment was reported, but obviously this method has little or no clinical application.
* Two other methods have been proposed to prevent or impede the migration of the epithelium. One consists of the total removal of the interdental papilla covering the defect and its replacement with a free autogenous graft obtained from the palate.
* During healing, the graft epithelium necroses and is slowly replaced by proliferating epithelium from the gingival surface.
* The graft simply delays the epithelium from proliferating into the healing area. This method has not been widely used.
* The second approach is the use of coronally displaced flaps, which increase the distance between the epithelial wound edge and the healing area.
* This technique is particularly suitable for the treatment of mandibular molar furcations and has been used mostly in conjunction with citric acid treatment of the roots.
* Periodontal regeneration after the use of this technique has been demonstrated histologically in humans.

Clot Stabilization, Wound Protection, and Space Creation

* Some investigators have attributed the successful results reported with graft materials, barrier membranes, and coronally displaced flaps to the findings that these techniques protect the wound and create a space for undisturbed and stable maturation of the clot.
* This hypothesis suggests that preservation of the root surface fibrin clot interface prevents apical migration of the gingival epithelium and allows for connective tissue attachment during the early wound healing period.
* The importance of space creation for bone repair has long been recognized in orthopedic and maxillofacial surgery.
* Transference of this concept to periodontal therapy has been explored for treatment of periodontal and peri-implant osseous defects and for root coverage.
* The space can be created by using a titanium-reinforced ePTFE membrane to prevent its collapse.
* For the study of reconstructive techniques, these membranes were placed over experimentally created supra-alveolar bone defects in dogs, and considerable bone reconstruction was reported.
* The following is a discussion of the GTR technique.

Graft Materials and Procedures

* Numerous therapeutic grafting modalities for restoring periodontal osseous defects have been investigated.
* Periodontal reconstruction can be attained without the use of bone grafts in meticulously treated three-wall defects (intrabony defects) and in periodontal and endodontic abscesses.
* New attachment is more likely to occur when the destructive process has occurred rapidly, such asafter treatment of pockets complicated by acute periodontal abscesses and after treatment of acute necrotizing ulcerative lesions.
* The use of graft materials at one time was to provide regenerative inductive effect, but it should be viewed primarily as providing a scaffold for healing.
* The following classifications of bone graft material are important. Grafts are categorized either by their origins or function during healing. Categorizations by origin include the following:
* (1) autografts are bone obtained from the same individual;
* (2) allografts are bone obtained from a different individual of the same species; and
* (3) xenografts are bone from a different species.
* Bone graft materials are also evaluated based on their osteogenic, osteoinductive, or osteoconductive potential.
* Osteogenesis refers to the formation or development of new bone by cells contained in the graft.
* Osteoinduction is a chemical process by which molecules contained in the graft (e.g., bone morphogenetic proteins) convert the neighboring cells into osteoblasts, which in turn form bone.
* Osteoconduction is a physical effect by which the matrix of the graft forms a scaffold that favors outside cells to penetrate the graft and form new bone.
* Periodontal defects as sites for transplantation differ from osseous cavities surrounded by bony walls.
* Saliva and bacteria may easily penetrate along the root surface, and epithelial cells may proliferate into the defect, thus resulting in contamination and possible exfoliation of the grafts.
* Therefore the principles established to govern transplantation of bone or other materials into closed osseous cavities are not fully applicable to transplantation of bone into periodontal defects.
* Schallhorn defined the considerations that govern the selection of a material as follows: biologic acceptability, predictability, clinical feasibility, minimal operative hazards, minimal postoperative sequelae, and patients' acceptance.
* It is difficult to find a material with all these characteristics and to date, no ideal material or technique exists.
* Graft materials have been developed and tried in many forms. To familiarize the reader with various types of graft material, as defined by either the technique or the material used, a brief discussion of each is provided.
* All grafting techniques require presurgical scaling, occlusal adjustment as needed, and exposure of the defect with a fullthickness flap.
* The flap technique best suited for grafting purposes is the papilla preservation flap because it provides complete coverage of the interdental area after suturing. The use of antibiotics after the procedure is generally recommended.

Autogenous Bone Grafts

* Historically, extraoral sites for bone harvesting have been from the iliac crest, but this approach is seldom performed due to medical and legal concerns.
* Intraoral sites can be effective, especially when donor sites adjacent to the defects are available.
* Despite the popularity of using allograft, this should always be a consideration, especially as one reviews the historical development of the use of autografts from intraoral sites.

Bone from Intraoral Sites

* In 1923, Hegedüs 113 attempted to use bone grafts for the reconstruction of bone defects produced by periodontal disease.
* The method was revived by Nabers and O'Leary 203 in 1965, and numerous efforts have been made since that time to define its indications and technique.
* Sources of bone include bone from healing extraction wounds, bone from edentulous ridges, bone trephined from within the jaw without damaging the roots, newly formed bone in wounds especially created for the purpose, bone removed from tuberosity, and the ramus and bone removed during osteoplasty and ostectomy.

Osseous Coagulum.

* Robinson 249 described a technique using a mixture of bone dust and blood that he termed osseous coagulum.
* The technique uses small particles ground from cortical bone.
* The advantage of the particle size is that it provides additional surface area for the interaction of cellular and vascular elements.
* Sources of the graft material include the lingual ridge on the mandible, exostoses, edentulous ridges, the bone distal to a terminal tooth, bone removed by osteoplasty or ostectomy, and the lingual surface of the mandible or maxilla at least 5 mm from the roots.
* Bone is removed with a carbide bur #6 or #8 at speeds between 5000 and 30,000 rpm, placed in a sterile dappen dish and used to fill the defect.
* The obvious advantage of this technique is the ease of obtaining bone from an area already exposed during surgery.
* The disadvantages are its relatively low predictability and the inability to procure adequate material for large defects.
* Although notable success has been reported by many individuals, studies documenting the efficacy of the technique are still inconclusive.

Bone Blend.

* Some disadvantages of osseous coagulum derive from the inability to use aspiration during accumulation of the coagulum.
* Another problem is the unknown quantity and quality of the bone fragments in the collected material. To overcome these problems, the bone blend technique has been proposed.
* The bone blend technique uses an autoclaved plastic capsule and pestle.
* Bone is removed from a predetermined site, triturated in the capsule to a workable, plastic-like mass, and packed into bony defects.
* Froum and associates found osseous coagulum–bone blend procedures to be at least as effective as iliac autografts and open curettage.

Cancellous Bone Marrow Transplants

* Cancellous bone can be obtained from the maxillary tuberosity, edentulous areas, and healing sockets.
* The maxillary tuberosity frequently contains abundant cancellous bone, particularly if the third molars are not present.
* After a ridge incision is made distally from the last molar, bone is removed with a curved rongeur.
* Care should be taken not to extend the incision too far distally to avoid entering the mucosal tissue of the pharyngeal area.
* In addition, the location of the maxillary sinus must be analyzed on the radiograph to avoid entering or disturbing it.
* Edentulous ridges can be approached with a flap, and cancellous bone and marrow are removed with curettes, back-action chisels, or trephine.
* Extraction sockets are allowed to heal for 8 to 12 weeks before reentering and removing the newly formed bone from the apical portion, which is used as the donor material.

Bone Swaging

* The bone swaging technique requires an edentulous area adjacent to the defect, from which the bone is pushed into contact with the root surface without fracturing the bone at its base.
* Bone swaging is technically difficult, and its usefulness is limited.

Bone From Extraoral Sites

* The use of fresh or preserved iliac cancellous marrow bone has been extensively investigated.
* This material has been used by orthopedic surgeons for years.
* Data from human and animal studies support its use, and the technique has proved successful in osseous defects with various numbers of walls.
* It has also been successful in furcations and even supracrestally to some extent.
* However, because of numerous problems associated with its use, the technique is no longer in use.
* Some of the problems were postoperative infection, bone exfoliation, sequestration, varying rates of healing, root resorption, and rapid recurrence of the defect.
* Other problems were increased patient expense and difficulty in procuring the donor material.

BONE ALLOGRAFTS

* Obtaining donor material for autograft purposes necessitates inflicting surgical trauma on another part of the patient's body.
* Obviously, it would be to the patient's and therapist's advantage if a suitable substitute could be used for grafting purposes that would offer similar potential for repair and not require the additional surgical removal of donor material from the patient.
* However, both allografts and xenografts are foreign to the patient and therefore have the potential to provoke an immune response.
* Attempts have been made to suppress the antigenic potential of allografts and xenografts by radiation, freezing, and chemical treatment.
* Bone allografts are commercially available from tissue banks.
* They are obtained from cortical bone within 12 hours of the death of the donor, defatted, cut in pieces, washed in absolute alcohol, and deep-frozen.
* The material may then be demineralized, subsequently ground and sieved to a particle size of 250 to 750 µm, and freeze-dried.
* Finally, it is vacuum-sealed in glass vials.
* Numerous steps are also taken to eliminate viral infectivity.
* These include exclusion of donors from known high-risk groups and various tests on the cadaver tissues to exclude individuals with any type of infection or malignant disease.
* The material is then treated with chemical agents or strong acids to inactivate the virus, if still present. The risk of human immunodeficiency virus (HIV) infection has been calculated as 1 in 1 to 8 million and is therefore characterized as highly remote.

Freeze-Dried Bone Allograft

* Several clinical studies by Mellonig, Bowers, and coworkers reported bone fill exceeding 50% in 67% of the defects grafted with freeze-dried bone allograft (FDBA) and in 78% of the defects grafted with FDBA in combination with autogenous bone.
* FDBA, however, is considered an osteoconductive material, whereas demineralized FDBA (DFDBA) is considered an osteoinductive graft.
* Laboratory studies have found that DFDBA has a higher osteogenic potential than FDBA and is therefore preferred.

Demineralized Freeze-Dried Bone Allograft

* Experiments by Urist established the osteogenic potential of DFDBA.
* Demineralization in cold, diluted hydrochloric acid exposes the components of bone matrix, which are closely associated with collagen fibrils and have been termed bone morphogenetic proteins (BMPs).
* In 1975, Libin and colleagues reported three patients with 4 to 10 mm of bone regeneration in periodontal osseous defects.
* Subsequent clinical studies were made with cancellous DFDBA and cortical DFDBA.
* DFDBA resulted in more desirable results (2.4 mm vs. 1.38 mm of bone fill).
* Bowers and associates, in a histologic study in humans, showed new attachment and periodontal regeneration in defects grafted with DFDBA.
* Mellonig and colleagues tested DFDBA against autogenous materials in the calvaria of guinea pigs and showed it to have similar osteogenic potential.
* These studies provided strong evidence that DFDBA in periodontal defects results in significant probing depth reduction, attachment level gain, and osseous regeneration.
* The combination of DFDBA and GTR has also proved to be very successful; however, limitations of the use of DFDBA include the possible, although remote, potential of disease transfer from the cadaver.
* A bone-inductive protein isolated from the extracellular matrix of human bones, termed osteogenin or BMP-3, has been tested in human periodontal defects and seems to enhance osseous regeneration.

Xenografts

* Bone products from other species have a long history of use in periodontal therapy.
* A few of these xenograft products are mentioned here for historical purposes but are no longer used today.
* Bovine-derived bone (Bio-Oss, Geistlich Pharma, Princeton, NJ) is used in combination with GTR for periodontal regeneration.
* This material is also used in combination with autologous bone for ridge augmentation.
* Calf bone (Boplant), treated by detergent extraction, sterilized, and freeze-dried, has been used for the treatment of osseous defects.
* Kiel bone is calf or ox bone denatured with 20% hydrogen peroxide, dried with acetone, and sterilized with ethylene oxide.
* Anorganic bone is ox bone from which the organic material has been extracted by means of ethylenediamine; it is then sterilized by autoclaving.
* These materials have been tried and discarded for various reasons.
* Currently, an anorganic, bovine-derived bone marketed under the brand name Bio-Oss (Geistlick Pharma) has been successfully used both for periodontal defects and in implant surgery.
* It is an osteoconductive, porous bone mineral matrix from bovine cancellous or cortical bone.
* The organic components of the bone are removed, but the trabecular architecture and porosity are retained.
* The physical features permit clot stabilization and revascularization to allow for migration of osteoblasts, leading to osteogenesis.
* Bio-Oss is biocompatible with the adjacent tissues, and it elicits no systemic immune response.
* Several studies have reported successful bone regeneration and new attachment with Bio-Oss in periodontal defects, as well as regeneration around implants and sinus grafting.
* Periodontally, Bio-Oss has been used as a graft material covered with a resorbable membrane (Geistlich Bio-Gide, Geistlich Pharma).
* The membrane prevents the migration of fibroblasts and connective tissues into the pores and between the granules of the graft.
* Histologic studies of this technique have shown significant osseous regeneration and cementum formation.
* Yukna and associates have used Bio-Oss in combination with a cell-binding polypeptide (P-15) that is a synthetic analogue of a 15- amino acid sequence of type I collagen marketed as PepGen P-15 (Dentsply Sirona, York, PA); this combination seems to enhance the bone regenerative results of the matrix alone in periodontal defects.

TISSUE ENGINEERING WITH BIOLOGIC MEDIATORS

* In wound healing, the natural healing process usually results in tissue scarring or repair. By using tissue engineering, the wound healing process is manipulated so that tissue regeneration occurs.
* This manipulation usually involves one or more of the three key elements: the signaling molecules, scaffold or supporting matrices, and cells.
* The use of tissue engineering for periodontal regeneration and dental implant site preparation has been reviewed.
* Early clinical examples involving tissue engineering principles include the use of bone allografts and autologous platelet-rich plasma (PRP).
* Investigations indicated that the success rates with these materials were inconsistent. With the development of recombinant growth factors and morphogens, and the use of synthetic scaffolds, the level of success has improved.
* Once considered experimental, tissue engineering is now clinically applicable with two commercially available tissue engineering systems for periodontal regeneration that involve the use of enamel matrix derivative (EMD) and platelet-derived growth factor-BB (PDGF-BB)–beta-TCP (β-TCP).
* The ability of BMP type I collagen sponge to enhance periodontal regeneration has been studied, but the mixed results and the concern for ankylosis have relegated this differentiation factor to be used primarily for implant site development.
* The development of a fourth promising system using basic fibroblast growth factor (FGF-2) is completing multicenter clinical trials. Because tissue engineering approaches are likely to improve clinical results, clinicians need to understand the biology and clinical parameters and limitations of these techniques.
* In the following sections, each of the three key elements of tissue engineering and how they are applied to the spectrum of periodontal and other oro-facial surgery procedures are reviewed.

Enamel Matrix Derivative for Periodontal Regeneration

* EMD has been effective in the treatment of infrabony defects (Fig. 63.2).
* The histologic evidence of EMD-induced periodontal regeneration has been confirmed in a clinical case report.
* A mandibular lateral incisor destined for orthodontic extraction was treated with acid etching and EMD. After 4 months, the tooth was extracted and examined histologically. Regenerated cementum covered 73% of the defect, and regenerated alveolar bone covered 65%. This histologic finding was later confirmed in other case reports, whereas new connective tissue attachment was reported in another case series where EMD was used in combination with a bone-derived xenograft.
* EMD has been shown to be safe for clinical use. Evidence of clinical efficacy was first reported in a multicenter study consisting of 33 patients with at least 2 defects, which were treated in a splitmouth design. The experimental site was treated with acid etching and EMD, whereas the control site was treated with a placebo. Patients were examined at 8, 16, and 36 months after surgery. Increased bone fill of the osseous defect was observed over time for 25 of the 27 (93%) EMD-treated teeth, but no bone fill was detected in the controls. The mean radiographic bone fill was greater for the EMD-treated defects compared with the control sites treated with open flap debridement (2.7 mm vs. 0.7 mm, respectively). Statistically significant improvements were also observed for EMD treated sites over control sites in mean pocket reduction (3.1 mm vs. 2.3 mm, respectively) and mean attachment level gain (2.2 mm vs. 1.7 mm, respectively). These clinical findings have been supported by additional studies.
* However, one randomized, double-masked, placebo-controlled clinical trial failed to show significant differences in clinical and radiographic measure between EMD and control.
* Long-term stability of EMD regenerative therapy was reported in a case series that followed EMD-treated defects in 90 patients. The data suggest that radiographic bone level, clinical attachment level gain, and reduced pocket depth reached near maximal response after 1 year, and the results remained stable over 5 years. Other long-term studies have confirmed these findings.
* Several studies have compared the use of EMD alone or in conjunction with other regenerative approaches. When EMD treatment was compared with GTR using bioresorbable membranes, the clinical results were comparable and stable over as much as a 10-year period.
* In a study comparing EMD, GTR, or EMD in combination with GTR with open flap debridement, all three had results superior to those of open flap surgery, with no additional improvement when EMD was used in conjunction with GTR. Other investigators have confirmed this finding.
* The use of EMD in combination with other graft materials is controversial. When EMD is used in conjunction with autogenous bone, DFDBA, xenograft, and bioactive glass, additional improvements in clinical parameters were observed as compared with the use of either EMD or DFDBA alone. However, others studies failed to demonstrate clinical improvement when EMD was used in conjunction with TCP or bioactive glass.
* EMD remains a very intriguing biologic mediator. As we better understand the mechanism of action of the potpourri of proteins and growth factors, this may strengthen the biologic rationale for clinical use of this material.
* The concern remains whether commercial batches of EMD will be consistent and provide comparable clinical results in all cases. Perhaps the message is that the achievement of maximum regenerative response will require a mixture of biologic mediators. With further characterization of EMD, we may better develop a synergistic blend that will provide an optimal result.

Recombinant Human Platelet-Derived Growth Factor for Periodontal Regeneration

* PDGF is one of the earliest growth factors studied for its effect on wound healing because it is a potent mitogenic and chemotactic factor for mesenchymal cells in cell culture.
* Histologic evidence of periodontal regeneration was first reported in experimental defects in beagle dog.
* During the development of PDGF for clinical use, recombinant human PDGF (rhPDGF) was used in conjunction with allogenic bone to correct class II furcations and interproximal intrabony defects on hopeless teeth.
* Histologic evidence of successful periodontal regeneration in the furcation lesion with excellent fill has been noted.
* A human clinical trial was conducted using rhPDGF and recombinant human insulin-like growth factor 1 (rhIGF-1). Using a split-mouth design, defects were treated with either a low dose (50 µg/mL) or high dose (150 µg/mL) of rhPDGF–rhIGF-1. After 9 months, the high-dose rhPDGF–rhIGF-1 induced 2.08 mm of new bone and 43.2% defect fill, compared with 0.75 mm vertical bone height and 18.5% bone fill in controls. Low-dose rhPDGF–rhIGF-1 results were statistically similar to those of the controls. Additionally, this study demonstrated that no adverse immunologic or clinical reaction resulted from use of these agents.
* A primate study examined the regenerative effects of PDGF–IGF-1 individually and in combination. PDGF alone was found to be as effective as the PDGF–IGF-1 combination in producing new attachment after 3 months. No significant effect was found when IGF was used alone. This study suggests that IGF may not be important at the dose level tested.
* Subsequently, the effectiveness of 0.3 mg/mL of rhPDGF in combination with β-TCP to improve attachment level gain, bone level, and bone volume significantly compared with β-TCP alone was demonstrated after 6 months in a multicenter clinical trial. A subset of these patients was followed for 24 months, and a representative case series was reported to be stable, with increases in radiographic bone fill compared with the end results after 6 months (Fig. 63.3). A review of these cases indicates that the results were stable after 3 and 5 years.
* Another case series suggested that rhPDGF with freeze-dried bone allograft can be combined to achieve excellent results in severe periodontal intrabony defects. These findings were confirmed by another randomized control trial.
* The combination of rhPDGF with a β-TCP carrier is now commercially available (GEM 21S, Osteohealth, Shirley, NY). These preliminary studies using rhPDGF-TCP suggest that it is easy to use, requires no barrier membranes, and has results comparable or superior to those of other regenerative graft materials.
* The potential for using rhPDGF for regeneration of furcation defects and implant site preparation still needs to be evaluated.
* Additionally, considerable clinical interest has been expressed in combining rhPDGF-BB with other bone replacement grafts, particularly bone allografts and xenografts.

Combined Techniques

* Periodontal new attachment and bone reconstruction have been challenges for clinicians throughout the history of periodontal therapy.
* To take advantage of the different bone graft materials and biologic mediators, clinicians have combined these graft materials with the use of membranes in an attempt to find a predictable technique to regenerate bone.
* Several clinicians have proposed a combination of the techniques previously described in an attempt to enhance their results.
* A classic paper published by Schallhorn and McClain in 1988 described a combination technique using graft material, root conditioning with citric acid, and coverage with a nonresorbable membrane (the only available one at the time).
* More recently, with the advent of osteopromotive agents, such as the EMD (Emdogain, Straumann, Andover, MA) and osteoconductive bovine-derived anorganic bone (Bio-Oss) graft materials, other combination techniques have been advocated.
* The combined use of these products, along with autogenous bone with resorbable membrane coverage, has resulted in an increased percentage of cases with successful new attachment and periodontal reconstruction.
* Whereas the use of combination technique may be appealing, it is important for clinicians to remember that these added materials often escalate the cost of the procedure and should be balanced with the quality and the long-term stability of the clinical results.

Factors That Influence Therapeutic Success

* Some of the therapeutic factors that have been implicated or shown to influence periodontal regenerative therapy adversely include
* (1) the selection of the appropriate surgical technique, accurate assessment of the periodontal defect, and the clinician's clinical experience (Fig. 63.4);
* (2) the importance of the tooth in the overall restorative treatment plan; and
* (3) the patient's selection of the regenerative options.

Therapeutic Considerations

* The selection of an appropriate therapeutic approach is one that is based on accurate assessment of the periodontal defect, one's past clinical experience, familiarity with the various regenerative and resective techniques, and the patient's selection of the regenerative options.
* Successful regenerative surgery requires delicate and timely tissue management to minimize tissue shrinkage.
* Some of these important surgical considerations are good passive flap closure for encasement of the graft materials and a flap design to allow tension-free suture placement.
* These concepts of using conservative and minimally invasive flap approaches have been introduced as minimally invasive surgery (MIS).
* By using these surgical principles, several modifications have developed and evolved into the MIS technique (MIST), the modified MIST (M-MIST), and the single-flap approach (SFA).
* The flap design for these techniques minimizes the number of incisions and as a result the surgical trauma, especially when the operation is performed with the use of magnification and microsurgical techniques.
* Several minimally invasive approaches have shown statistical improvements when used in conjunction with EMD, although with no difference when used with rhPDGF.
* Most of these studies were of short duration, whereas one study reported that the 12-month result remained stable at 6 years.
* Of special interest is that the recession values for these conservative or minimally invasive procedures tend to be lower than those reported with other access flap surgery techniques.
* GTR studies suggest that the MIS approach may be better suited for regeneration in the aesthetic zone.
* Large scale randomized controlled trials are needed to evaluate emerging treatment approaches such as MIS regarding its safety, efficacy, and patient-reported outcomes including pain, postoperative complications, aesthetics, and patients' satisfaction.

Tooth and Defect Related Considerations

* Therapeutic success is influenced by the tooth's importance in prosthetic rehabilitation, its endodontic status, and the osseous defect characteristics.
* The critical question to be addressed is whether the involved dentition is strategically important to treat and maintain by using regenerative periodontal theapy.
* If the tooth has little or no importance in the overall treatment plan, extraction may be indicated to avoid potential technical difficulties, postsurgical complications, and expenses. Strategic extraction may also improve access for better plaque or biofilm removal by the patient and compliance.
* If a tooth is deemed essential, it is important to assess its endodontic status before the clinician proceeds with periodontal regenerative therapy.
* A positive endodontic status is necessary before one proceeds with regenerative therapy.
* The financial consideration is also important for undertaking regenerative therapy.
* In a clinical scenario, if endodontic therapy, periodontal regenerative therapy, and restorative therapy are all necessary to retain a tooth, the financial commitment may indicate the possible extraction and replacement with a dental implant or a prosthesis.
* Characteristics of the defect, such as the overall osseous pocket depth, width, and walls, can influence clinical outcome in response to regenerative surgery.
* Studies have consistently indicated that a deeper defect is correlated with increased clinical attachment level and probing depth.
* One is more likely to achieve improved regenerative results when the osseous defects are narrow, circumferential, and with a three- or two-walled configuration.
* Conversely, a wide osseous lesion with one wall or no wall is less amenable to regenerative procedures.
* Even with the use of iliac and autologous grafts, current regenerative approaches have not been successful in regenerating one- or zero-walled osseous defects.
* The role of tooth mobility in regenerative therapy remains controversial.
* Clinical experience suggests that regenerative therapy is successful when the tooth is not mobile before regenerative therapy.
* Numerous studies support the positive attribute of controlling mobility through presurgical splinting as compared with mobile teeth before regenerative therpy.
* A study by Schultz and colleagues evaluated mobile teeth stabilized by splinting versus mobile teeth before regenerative surgery.
* Presplinting of mobile teeth treated with bone replacement graft resulted in significantly improved clinical parameter as compared with nonsplinted teeth after a year.
* These studies suggest the importance of appropriate presurgical management of tooth mobility before regenerative procedures.

Patient-Related Considerations

* Classic studies of poor plaque control and poor postoperative recall compliance have indicated that much of the therapeutic gain from periodontal surgery will deteriorate.
* Similarly, the positive results from GTR regenerative procedure have been shown to deteriorate with poor compliance.
* Progressive deterioration in these patients have been demonstrated to be associated with a higher incidence of infection with putative periodontal pathogens (Porphyromonas gingivalis, Prevotella intermedia, and Aggregatibacter actinomycetemcomitans)
* Areas such as furcation and root proximity situations have also been shown to be difficult to maintain, and as a result, the risk for deterioration is higher.
* As a clinician in practice, it is important to remember that the patient presented to the clinician with behavioral (plaque or biofilm) and anatomic problems, which resulted in periodontal disease.
* After therapy, the difficult challenge is to motivate patients to be skilled, enthusiastic, and passionate about their oral hygiene and compliant with periodontal maintenance.
* Smoking is a behavioral challenge that the therapist must always assess if the patient is a smoker. Smoking not only promotes disease progression, but also results in adverse therapeutic outcomes.
* It has been implicated as having a detrimental effect on periodontal wound healing following surgical procedures, as well as to influence periodontal regeneration adversely.
* Studies examining the regenerative outcomes in diabetic patients are lacking due to ethical considerations.
* Despite the lack of direct evidence in humans, animal studies confirm the detrimental effects on poor wound healing capacity in diabetic animals as compared with controls.
* Of special interest is that the use of biomimetic agents, such as EMD, did not improve the compromised healing response of animals with diabetes.

Clinical Guidelines to Guide Clinicians in Their Patient Management

* Clinical guidelines for the management of patients with periodontal disease are depicted in Fig. 63.4. 136
* The ideal management of periodontal defects consists of the early diagnosis and appropriate addressing of the defect (Fig. 63.4, A).
* When defects are detected early, before the formation of intrabony and furcation lesions, a predictable outcome can be obtained with scaling, root planing, and conventional osseous surgery (Fig. 63.4, B).
* Even early narrow intrabony (3 mm, periodontal regeneration should be considered (Fig. 63.4, C).
* Assessment of defect morphology and the patient's clinical and systemic-behavioral determinants is critical for regenerative success.
* Consideration of these issues, in addition to the patient's desires, will define the selection of the regenerative approach to be used (Fig. 63.4, D).
* Long-term stability is possible, but the individual outcome is influenced by patient-related considerations such as smoking and compliance with periodontal maintenance and monitoring.
* Should patient-related or clinical determinants be unfavorable for periodontal regeneration, appropriate therapy must be selected in place of regeneration that may consist of long-term maintenance or the removal of the tooth and replacement with a prosthesis such as a dental implant or another form of prosthesis. (Fig. 63.4, E).
* Before regenerative therapy, it is important to perform an endodontic assessment. This is to eliminate the possibility that the defect is the result of an endodontic-periodontal lesion. Should this be case, endodontic treatment may resolve that portion of the defect due to the endodontic lesion.
* If a residual defect still persists, periodontal therapy should be initiated.
* A common misconception is that regenerative therapy ends with a postoperative assessment a few months after treatment.
* Most therapeutic approaches have maximal healing results after 12 months. As such, postoperative monitoring should occur at least 12 months later.
* Additionally, these regenerated areas should be monitored at every recall visit because poor hygiene, uncorrectable tooth anatomy, and undiagnosed endodontic problems will cause these areas to relapse. Should failures due to these causes be determined, it may be prudent to consider strategic extraction.
* The endpoint for active periodontal therapy should comprise a stable periodontal attachment level, absence of inflammation or bleeding, and a periodontal anatomic environment that is conducive for the patient and the clinician to maintain excellent oral hygiene.
* A successful long-term periodontal outcome is also dependent on a patient who will be compliant with the maintenance visits.

Future Directions for Periodontal Regeneration

* In wound healing, the natural healing process usually results in tissue scarring or repair.
* By using tissue engineering, the wound healing process is manipulated so that tissue regeneration occurs. This manipulation usually involves one or more of the three key elements:
* (1) the signaling molecules,
* (2) scaffold or supporting matrices, and
* (3) cells (see Fig. 63.1).
* The cellular responses to these biologic mediators in vitro have been studied and are summarized in Table 63.2.
* Some of these biologic mediators are commercially available (recombinant human bone morphogenetic protein [rhBMP], rhPDGF, EMD).

Bone Morphogenetic Proteins for Periodontal and Implant Site Regeneration

* BMPs comprise a group of regulatory glycoproteins that are members of the transforming growth factor beta superfamily that function as differentiation factors.
* These proteins induce cellular differentiation of stem cells into chondroblastic and osteogenic cells. Much of the research interest has focused on BMP-2 (OP-2), BMP-3 (osteogenin), and BMP-7 (OP-1).
* BMPs have been demonstrated to be present in FDBA and DFDBA, but the levels are so low that BMP is not biologically active.
* In fact, the amount of BMP is so low that it takes approximately 10 kg of bovine bone to yield only 2 µg of BMP.
* It is only through recombinant DNA technology that BMP has been made available for clinical use.
* Although early studies using crude preparations of BMP-2 and BMP-3 applied in surgically induced furcation defects appeared to stimulate periodontal regeneration, more recent study with rhBMP-2 indicated that periodontal regeneration was associated with areas of ankylosis.
* Healing through ankylosis has been a concern, so most of the research using rhBMPs has involved correction of intrabony, supra-alveolar, furcation, and fenestration defects, †† as well as implant site preparation.

Use of Recombinant Human Fibroblast Growth Factor 2 for Periodontal Regeneration

* The potential use of recombinant human FGF-2 (rhFGF-2) for periodontal regeneration has been reviewed.
* Preliminary beagle and nonhuman primate studies demonstrated that topical application of FGF-2 into intraosseous defects in alveolar bones induces significant periodontal tissue regeneration.
* Histologic observation revealed new cementum with Sharpey fibers, new functionally oriented periodontal ligament fibers, and new alveolar bone.
* These findings suggest that topical application of FGF-2 may be efficacious in regeneration of human periodontal tissue that has been destroyed by periodontitis.
* A randomized, controlled, double-masked Phase II clinical trial at 13 Japanese dental facilities compared therapeutic response to varying doses of FGF-2 as compared with control. Eighty patients with a two- or three-walled vertical bone defect 3 mm or more from the alveolar crest were randomly divided into four groups: (group A) placebo control, (group B) 0.03% FGF-2, (group C) 0.1% FGF-2, and (group D) 0.3% FGF-2. The subjects underwent periodontal surgery during which 200 µL of the investigational drug or placebo carrier was applied to each test site. After 9 months, a significant increase (P = 0.021) in alveolar bone height was demonstrated by standardized radiographs between group A (23.92%) and group D (58.62%). No adverse effects were observed throughout this multicenter trial. This finding suggests that topical application of FGF-2 can be efficacious in regenerating periodontal tissue of patients with two- or three-walled intrabony defects. This has led to a subsequent larger clinical trial that has been completed, and the results are forthcoming.
* These trials will provide crucial information regarding the safety and efficacy of using FGF-2 for periodontal regeneration.

Cell Therapy

* Cell therapy has been used in periodontal surgery (Osteocel Plus, NuVasive, San Diego, CA).
* Stem cells have the potential to improve current bone regeneration.
* These cells can expedite cell recruitment, be target cells for growth factor delivery, and promote early extracellular matrix formation.
* All of these cellular activities increase the bioactivity of the graft. The concentration of multipotential stromal cells (MSCs) in a commercially available cellular bone allograft was compared with fresh age-matched iliac crest bone and bone marrow aspirate.
* Without cultivation or expansion, this allograft contains cells with cell surface markers called cluster dif erentiation (CD) markers that are found with immunotyping of osteoprogenitor cells and osteoblasts.
* These cells displayed an “osteoinductive” molecular signature and the presence of CD45 -CD271 +CD73 +CD90 +CD105 + MSCs surface markers that were more than 100-fold of what are found in iliac crest bone.
* In comparison with bone marrow, MSC numbers enzymatically released from 1 g of cellular allograft were equivalent to approximately 45 mL of bone marrow aspirate.
* This MSC cellular allograft bone represents a unique, nonimmune material rich in MSCs, osteoblasts, and osteocytes.
* This osteoinductive cellular graft represents an attractive alternative to autograft bone by eliminating a secondary surgical harvest site and morbidity risk.
* This stem cell preparation with bone scaffold has been used for implant site preparation by increasing increase alveolar ridge volume and sinus grafting, Additionally, this cellular allograft has been used successfully in regenerative treatment of periodontal defects in both a single-rooted tooth and a multirooted tooth.
* In the single-rooted case, a significant reduction in probing was obtained with radiologic evidence of approximately 4 mm of vertical bone fill at 6 months following grafting.
* In the multirooted case, clinical evidence showed decreased probing depths and radiographic bone improvement at 6 months. A cone beam computed tomography scan taken at 14 months demonstrated three-dimensional bone fill.
* A similar result was presented in a case report, where this allograft cellular bone matrix was used in the successful treatment of a severe periodontal defect. These case reports indicate a potential resolution of periodontal defects using cellular allograft material.

Scaffold or Supporting Matrices

* The use of scaffolding matrices to deliver growth factors to promote periodontal tissue regeneration has been an active area of research.
* Supporting matrices for engineering bone and soft tissue have included processed bone allografts, synthetic and natural polymers, synthetic ceramics, bovine type I collagen, and calcium sulfate.
* The major roles for the supporting matrices are to:
* 1. Provide physical support for the healing area so that no collapse of the surrounding tissue into the wound site occurs. Examples of this would be bone allografts and synthetic ceramics such as TCP.
* 2. Serve as a barrier to restrict cellular migration selectively. This is best exemplified by principles of GTR and guided bone regeneration (GBR) where nonresorbable PTFE and resorbable polylactate, polyglycolic acid, and calcium sulfate are used. The exclusive properties of cell barrier membranes have been reviewed and are not addressed here.
* 3. Serve as a scaffold for cellular migration and proliferation. Examples include the collagen matrix. Potentially, this scaffold can be further enhanced by selectively defining the types of cells permitted to attach to and proliferate on this matrix with the additions of adhesins and/or integrins.
* 4. Potentially serve as a time-release mechanism for signaling molecules.

Allogenic and Alloplastic Bone-Grafting Materials

* The classic approach to orofacial regeneration since the 1980s has been the use of bone grafts or substitutes to repair periodontal and maxillofacial defects.
* The literature contains several excellent reviews on the use of autografts, allografts, and alloplastic graft materials.
* Since 2000, mineralized and demineralized freeze-dried bone allografts (FDBA and DFDBA) have often been the regeneration material of choice.
* In addition to their availability and putative osteogenic potential, various clinical studies indicate that 2 to 3 mm of bone fill is possible with FDBA and DFDBA.
* However, other studies have questioned the osteogenic potential of bone allografts by suggesting that this may vary depending on the bone bank or batch, processing procedures used, and donor characteristics.
* Due to varying osteoinductive properties, which are not areas the US Food and Drug Administration regulates, growth factors and morphogens with FDBA and DFDBA are not commercially available.
* However, off-label use of this combination is common both in orthopedic and oral-periodontal surgical procedures.
* Alternatively, a variety of xenograft and alloplastic grafting materials is available as scaffolding agents for tissue engineering.
* Alloplastic bone grafts consist of ceramics, such as HA, porous HA, β-TCP, and biocompatible composite polymers (e.g., hard tissue replacement).
* Of these allografts, β-TCP is used in combination with rhPDGF-BB. In the development of rhPDGF-BB for clinical use, the biosynthetic β-TCP was used because it possessed defined and reproducible properties as required by the Food and Drug Administration.
* Allografts were not desirable because of varying osteogenic potential and properties. Extensive animal and human studies demonstrated biocompatibility of β-TCP with no reports of adverse reactions.
* This material physically fills bone defects, provides a scaffold for new bone formation, and prevents soft tissue collapse into the wound site.
* Clinically, β-TCP is osteoconductive and supports early healing. β-TCP resorbs at varying rates depending on the chemical structure, porosity, and particle size.
* Absorption, release, and bioactivity studies indicate that either βTCP or calcium sulfate can be an effective carrier for PDGF-BB. Approximately 45% of the adsorbed PDGF-BB was released after 10 days.
* In clinical studies with rhPDGF-BB, this material resorbs and is replaced with regenerated periodontium.
* Superficial granules at the soft tissue interface appear to resorb at a slower rate.
* More recently, the use of β-TCP coated with recombinant human growth and differentiation factor-5 was evaluated for its osteoinductive and osteoconductive properties in an experimental rat calvarial critical size defect.
* Histomorphometric results suggest that this proprietary coating of growth factor on β-TCP achieved superior bone regeneration compared with conventional materials. These latter two studies indicate that the absorption and release kinetics of signaling agents are areas that require further elucidation if we are to achieve optimal regenerative response.

Collagen Carriers

* Collagen is the main structural protein for tissue support. It also plays an essential role in wound healing by providing a biologic scaffold for cellular activities such as cell attachment, migration, and proliferation.
* Collagen has been widely used in tissue engineering for seeding of mesenchymal stem cells, as well as incorporation of growth factors.
* Because most collagens are derived from bovine dermal or skeletal tissue, concerns related to the purity, quality, immunogenicity, and potential for prion transmission have been raised.
* Studies addressed these issues by further purifying bovine collagen and modifying the molecules to remove antigenic N- and C-terminal telopeptides.
* Two variations of this collagen were produced to create an atelocollagen scaffold where both constructs were effective in supporting rhBMPinduced bone formation.

Calcium Sulfate

* Calcium sulfate is one of the oldest bone graft materials.
* Early clinical and animal studies indicate that calcium sulfate is biocompatible, degrades over time, is subsequently replaced with regenerated bone, and may be used in an infected area with no complications.
* More recent studies indicate that it also has barrier properties, enhances angiogenesis, and may be effective as a delivery vehicle for antibiotics, as well as growth factors.
* Rosenblum and associates demonstrated that FGF was observed to be released at a rate directly proportional to the rate of calcium sulfate dissolution.
* A secondary benefit of calcium sulfate dissolution is a local decrease in pH.
* An interesting study in the orthopedic literature reported that when an experimental sheep distal femoral cancellous defect was filled with calcium sulfate, increased immunostaining for BMP-2, BMP-7, transforming growth factor beta, and PDGF-BB was observed.
* All of these growth factors have been demonstrated to stimulate bone formation and development.
* Calcium sulfate was found to be a suitable carrier for rhPDGF-BB with a longer release kinetic profile (~16 days) as compared to β-TCP.
* Because both materials are resorbable, the current debate centers on whether a longer sustained release of rhPDGF-BB would be more advantageous to periodontal and bone regeneration.

Other Carriers

* Bioresorbable polymers of poly lactic-co-glycolic acid and polyglycolic acid have been considered as scaffolding agents for tissue engineering due to their biodegradable and tissue compatibility properties.
* Although these agents were promising as carriers for osteogenic factors in animals, variable tissue responses have made clinical application of these materials problematic. These tissue responses include inflammation, foreign body reaction, and local acid accumulation during polymer degradation.