

Comparative Evaluation of
Grafting Materials
for
Intra-oral Utilization

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The article evaluates the characteristics of different synthetic and biologically derived materials which are used in the preservation and augmentation of the alveolar ridge. The characteristics of the materials as reported in the literature are considered as they relate to normal bone healing and dental implant placement.

Key Words

Bone Grafting

Ridge Preservation

Bone Augmentation

Introduction

Alveolar bone is rapidly contoured after the loss of natural teeth. There is a 25% decrease in volume during the first year and a 40% to 60% decrease in width within the first three years after extraction. (Misch 1999) The preservation or restoration of the residual ridge following loss of teeth is critical to the surgical placement of dental implants such that they can be restored in an optimal esthetic and functional manner.

Numerous synthetic and biologically derived materials have been evaluated for use in the preservation or augmentation of bone. To date, the goal of complete, predictable regeneration has not been attained however; the literature has clearly demonstrated the clinical feasibility and histological possibility of bone regeneration. One or a combination of several synthetic or biologically derived materials has been utilized by clinicians within an extraction socket or osseous defect in an attempt to create osteo conductive scaffolding which would preserve the contours of the resulting ridge. The selection of one material or a combination of materials is often based on the clinician's instincts rather than clinical studies. The materials characteristics as they relate to normal bone healing, the location of the graft, the timing of the dental implant

insertion and the final clinical restorative goals should be considered when selecting a bone graft material which will be utilized intra orally.

Normal Osseous Healing

Osseous reconstruction or preservation becomes most clinically predictable when one considers the normal process of bone healing and how this process is impacted by the introduction of a graft material. The time line of the osseous healing phases must also be considered as they relate to the characteristics of the bone graft material and the timing of the implant insertion. Bone formation occurs by osteoblastic activity. Osteoblasts are fully differentiated cells and lack the capacity for migration and proliferation. Therefore, in order to allow bone formation to occur at a given site, undifferentiated mesenchymal progenitor cell (osteoprogenitor cells) must migrate to the site and proliferate to become osteoblasts. (Lang, Araujo & Karring 2003) Bone formation involves the proliferation and maturation of primitive precursor cells into functional osteoblasts. The bone cells allegedly originate from mesenchymal stem cells that commit to the osteogenic cell lineage becoming osteoprogenitor cells, pre-osteoblasts, osteoblasts, and osteocytes (Long, 2001)

Osteoprogenitor cells are divided into determined precursor cells and inducible precursor cells. Determined precursor cells are present in the endosteum of bone marrow and periosteum covering of bone. These cells possess an intrinsic capacity to proliferate and differentiate into osteoblasts. Inducible precursor cells represent mesenchymal cells that are present in other organs and tissues (i.e. muscle). These cells may become bone-forming cells when exposed to specific stimuli. The main osteoprogenitor cells are pericytes which are present in vascular tissue. The differentiation of osteoprogenitor cells to osteoblasts is dependent on BMP (bone morphogenic protein), IGF (insulin growth factor), PDGF (platelet derived growth factor), & FGF (fibroblast growth factor). (Zhang et al. 1997)

Bone healing can be looked at as occurring in four phases. These phases include the blood clotting phase, the wound cleansing phase, the bone formation phase and the bone modeling & remodeling phase.

In the first 24 hours following a tooth extraction the blood clotting phase takes place. Blood fills the socket and platelets aggregate and interact with fibrin to form a blood clot. The clot contains growth factors that induce and amplify the migration of mesenchymal cells and influence them to proliferate

and differentiate. Blood vessels are closed by thrombi and a fibrin network is formed. Within a few days, fibrinolysis occurs and the blood clot begins to break down.

Within 2-3 Days the wound cleansing phase begins. The blood clot begins contracting and inflammatory cells (i.e. neutrophils, monocytes) and fibroblasts begin to migrate along the fibrin network. As the neutrophils & macrophages migrate they begin to phagocytize damaged tissue. Macrophages release growth factors and cytokines further promote the migration, proliferation and differentiation of mesenchymal cells. Neutrophils eventually undergo apoptosis and macrophages subsequently withdraw from the socket. Portions of the traumatized bone will undergo necrosis and are removed by osteoclasts. The clot is then replaced by the formation of granulation tissue with blood vessels and collagen fibers. After 3 days, an increased density of fibroblasts is visible in the clot and the proliferation of epithelium from the wound margins is apparent. Remodeling of the socket begins with the presence of osteoclasts inducing bone resorption. One week after the extraction the socket is characterized by granulation tissue consisting of a vascular network, young connective tissue, and osteoid formation in the apical portion and epithelial coverage over the wound.

At the beginning of the bone formation phase mesenchymal, fibroblast-like cells start to proliferate and deposit a matrix in a process called fibroplasia. Granulation tissue will gradually replace blood clot. Newly formed vessels or angiogenesis begins & provisional connective tissue starts to establish. Pericytes migrate and differentiate into osteoblasts that produce a matrix of collagen fibers, which take on a woven pattern. By the first month the socket is characterized by a dense connective tissue overlying the residual sockets, which are now filled with granulation tissue. The process of mineralization is initiated and the formation of new bone (woven bone) has started. Wound coverage by epithelium is complete.

During the tissue modeling & remodeling phase the woven bone is gradually replaced by lamellar bone and bone marrow through the process of modeling and remodeling. It will take several months until a woven bone in the extraction socket is replaced by bone marrow and lamellar bone. Two months following extraction bone formation in the socket is complete. The bony height of the original sockets has not yet been reached and the trabecular pattern is still undergoing remodeling.

Mechanisms of Osseous Regeneration

Osseous regeneration is mediated through several individual processes or combination of processes. **Osteogenesis** is a mechanism of forming bone directly from osteoblasts. This occurs when osteoblasts are transplanted with grafting material into a defect and establishes centers for bone formation. (Hoexter 2002) Osteogenesis requires the presence of autogenous bone.

Osteoinduction describes a process whereby new bone is produced in an area where there was no bone before or where one tissue or its derivative causes another undifferentiated tissue to differentiate into bone. In other words, the ability of a graft to induce non-differentiated stems cells or osteoprogenitor cells to differentiate into osteoblasts. The phenomenon of osteoinduction was first described in the classic works of Urist (Urist & McLean 1952, Urist 1965, Urist et al 1977).

Osteoconduction describes bone formation by the process of ingrowth of capillaries and osteoprogenitor cells from the recipient bed into, around and through a graft. Therefore the graft or bioimplant acts as a scaffold for new bone formation (Buchardt 1983). Unlike osteoinduction, this process occurs in an already bone containing environment.

Autogenous Bone

Traditionally, the "Gold Standard" for osseous regeneration has been autogenous bone. It is resorbable, eventually replaced by new bone and is non-allergenic. However, due to limited intraoral sources and need for secondary surgical sites which can lead to increased morbidity, much interest has been regarded in other options.

Allogenic Bone

Allogeneic bone is non-vital osseous tissue taken from one individual and transferred to another individual of the same species. Allografts have been the favorite of periodontists for many years.

There are three forms of allogeneic bone:

Fresh frozen- rarely used today for the purposes of bony reconstruction because of the concerns related to the transmission of viral diseases (Buchardt 1983).

Freeze-dried- the process of freeze-drying removes moisture from the bone resulting in a graft with mechanical strength that can be used to onlay areas or as a crib to retain autogenous bone (Marx 1993). This graft is osteo conductive and has no osteogenic or osteoinductive capabilities and consequently requires a source of osteo competent cells. Therefore, are usually placed in conjunction with autogeneic grafts. **Freeze**

Dried Bone Allograft has had the collagen / protein removed. It is a source of calcium, it is osteo conductive and provides a three dimensional scaffold for bone guidance and apposition. The most effective mineralized bone is cancellous bone. Cancellous bone is porous which allows for cellular invasion and resorption. Cortical allografts are considered to be non-resorbable and although a filler, they are dense, and non-resorbable.

Demineralized Freeze-Dried Bone Allograft (DFDBA) has had the mineral component removed. It is an excellent source of collagen. Collagen is used as a spacer between particles of bone replacement graft materials (such as xenografts). Spacing is essential for cell transport and vascularization throughout the graft. DFDBA is a one dimensional source of collagen that provides this spacing. Claims of osteoinduction are made but vary considerably from tissue bank to tissue bank and from donor to donor. Demineralized Freeze-Dried Bone Allograft (DFDBA) has been considered the most effective materials for bone regeneration in periodontal defects but it has gained in popularity with all clinicians that are depending on guided bone regeneration as a pre-implant treatment for their patients.

There is a big difference in processing methods of allografts. Some are freeze-dried, some are frozen, and some are irradiated. The most effective methods are those that maintain the porous structure and anatomy of the mineralized bone with scrutinized sterilization that adheres to the AATB guidelines.

Puros Allograft (Zimmer Dental)

Puros is a human cancellous bone processed by a unique, proprietary preservation/sterilization process called solvent-dehydration or Tutoplast process. In contrast to simple dehydration by freeze-drying this process completely removes all bone marrow, fat and noncollagenous proteins leaving the pure extracellular matrix in unchanged condition. The process removes all antigenicity and is highly effective against bacteria, viruses and prions. Puros comes sterile with a shelf life of 5 years at room temperature. It quickly hydrates in normal saline turning into a paste like consistency that is easy to manipulate and transfer to the recipient site. It can be nicely condensed without shattering thereby providing perfect preconditions for rapid remodeling. The preserved collagen activates blood clotting that prevents particle migration. Puros shows excellent biocompatibility (1) and remodels completely in non-critical (2) as well as in critical size defects (3). Puros is used in orthopaedics since 15 years with excellent results

comparable to autograft bone (4). It also has proven effective clinically in oral surgery. Implants could be placed after 4 month in grafted extraction sockets (5) and after 5 month after crest augmentation (6). The authors reported minimal loss of volume and the feeling of strong, solid bone at reentry.

(Guenther et al 1996, Dalkyz et al 2000, Fornaro et al 2000, Rocci et al 2000, Block et al 2002, Trentz et al 2003)

IRRADIATED ALLOGENIC CANCELLOUS BONE & MARROW

(Rocky Mountain Bone Bank)

Irradiated Allogenic Cancellous Bone & Marrow was developed exclusively for the dental industry. These grafts have been distributed in the United States and abroad since 1984.

Indications for use are Sinus Augmentation, Sinus Lift, On-lay grafts and periodontal defects. The Irradiated Cancellous Bone & Marrow is uniquely processed from human vertebra, the richest source of bone marrow in the human body thorough out an individual lifetime. The grafts are morselized to give maximum scaffolding, frozen at -70 degrees centigrade and then irradiated with 2.5 to 3.8 Megarads of irradiation from a Cobalt 60 source for sterilization. The irradiation eliminates antigenicity, preserves the graft and demonstrates a 10⁻¹⁰ Cidal effect on bacteria, virus, and fungus. The sterile grafts have a 2-year shelf life and are maintained at room temperature. The

grafts present moist eliminating the need for rehydration. It is reported by clinicians to give results closest to autogenous grafts allowing implants to be place 4 months after transplantation.

Demineralized bone matrix (DBM)- The current widespread use of DFDBA is based on the supposed osteoinductive ability of demineralized bone graft preparations. These desired osteoinductive factors (Transforming growth factor-beta, osteogenic, insulin-like growth factor, fibroblast growth factor) and bone morphogenetic proteins (BMP) are removed from mineralized tissue by using a demineralization agent such as hydrochloric acid. BMP is a hydrophobic glycoprotein within the bone matrix and thought to be responsible for an osteoinductive effect by eliciting the differentiation of host mesenchymal cells into osteoblasts. This process leads to osteoblastic invasion, new bone formation, and overall remodeling within about 3 weeks. (Urist 1965, Zhang et al 1997)

The issue of safety and risk of disease transmission when using allografts is always a concern. Allografts can be treated by various methods such as:

Freeze-drying

Gamma irradiation

Electron beam radiation

Ethylene oxide

It is assumed that the process of demineralization of the freeze-dried graft and DBM eliminates or inactivates the p24 core protein and reverse transcriptase (HIV). Some have estimated that the risk of HIV transmission alone with allograft bone is 1 in 1.6 million (SEOPF, 1988). Buck et al 1990, confirmed the presence of the HIV in bones of infected patients however, they estimated the risk of transmission of the virus after freeze drying bone to be 1 in 8 millions. They multiplied 1.6 million given by Boyce, by 1/5. This dilution factor was because out of HIV positive five bone samples, only one of them retained the virus after freeze drying . Another study showed that the risk of transmitting HIV with properly screened has been calculated to be in 1 in 2.8 billion (Russo & Scarborough 1995). One case of Hepatitis B and 3 cases of Hepatitis C transmission have been reported with the latest case occurred in 1992 (Laurencin, 2003). Although rigorous donor screenings and tissue treatments have greatly reduced the incidence of HIV and Hepatitis transmission, other diseases have been passed on as recently as April 2000 in orthopedic medicine where 2 different patients received bone-tendon-bone allografts for anterior cruciate ligament reconstruction from a common donor and developed septic arthritis; and in November 2001 where a patient

underwent reconstructive knee surgery and within 4 days died from clostridium infection (Laurencin, 2003). Although many methods can reduce the risk of disease transmission, other concerns have developed. For example, treatments used to sterilize the tissues, removes proteins and factors, reducing or eliminating the osteoinductive tissue. In addition, the nature of organic material and the processing of high heat or radiation may cause these materials to crystallize and harbor endotoxins or dormant pathogens. (Valen & Ganz 2002)

Xenografts consist of skeletal tissue that is harvested from one species and transferred to the recipient site of another species (Van den Bogaerde & White 1997, Hammer et al 1998). These grafts can be derived from mammalian bones and coral exoskeletons. Bovine derived bone has been commonly used (Block & Posner 1995, Jensen et al 1996), even though other sources are such as porcine or murine bone is available. Xenogeneic bone was popular in the 1960's but fell into disfavor due to reports of patients developing autoimmune diseases following bovine bone transplants (Pierson et al 1968, Buchardt 1983). The introduction of these products in the 1990's comes after the development of methods to remove the protein from the bone particles (Iwamoto et al. 1997). This processing reduces the antigenicity making these grafts more tolerable to host tissue (Basle et al 1998). The

result is that the organic component of bone is almost completely removed. This inorganic bone matrix then has the structure of bone, making its osteoinductive abilities imparted by the organic elements. Eventually xenogeneic bone should be replaced by host tissue, which would make it useful for defect or extraction sites. Resorption of bovine derived bone has been observed in animal studies (Merckx et al 1997) but not consistently in human clinical trials (Hallman et al 2001, Valentin 1998, Skoglund 1997). Bio-Oss®, PepGen and Osteograf®/N all have published histologic studies demonstrating bony encapsulation osseous bridging between the graft particles and enhancement of osseointegration around immediate implants but none show complete resorption of the graft material (Skoglund et al, 1997, Tehemar et al, 2003)

Bovine bone has been used successfully in dentistry for many years. The two main manufacturers / distributors of bovine bone are OsteoHealth (Bio-Oss®) and Dentsply Friadent CeraMed (OsteoGraf®/N and PepGen -P-15™). Although both companies meet the FDA guidelines for processing xenografts, there is a marked difference in their methods.

Osteo Health uses a chemical bath of the bone, numerous washings then a slow ramp-up of temperature to dry the bone prior to

pulverizing. The dry heat temperature used is always below 600° C at the highest point. Using chemicals and low temperature to remove the bovine proteins from the bone, allows for a "softer" end product. BioOss® has a total % (after processing) of approximately 60%. This porous structure allows for cellular invasion and a breakdown of the particulate during the resorptive phase. The porosity also offers a more user friendly material. BioOss® is porous therefore, it absorbs the hydrating fluid (sterile saline, sterile water, blood etc) and forms a semi-cohesive mass which is fairly easy to handle and manipulate.

Dentsply Friadent CeraMed deproteinates the bovine bone used in OsteoGraf®/N and PepGen P-15 with a non-chemical, high temperature method. There are no chemicals involved. Numerous water baths prepare the bone for the extreme high temperature that reaches 1100° C. At 1100°, it is impossible for any protein or organic to survive. This includes prions which are linked to Bovine Spongiform Encephalopathy (BSE). However, this extreme temperature does affect the natural porosity of the bone. OsteoGraf®/N and PepGen P-15 are approximately 28% porous. This reduction in both % of porosity and size of the pores, results in a denser, less resorbable material. Less

porosity makes it less absorptive, less cohesive and more difficult to handle and place.

PepGen P-15, Dentsply Friadent CeraMed's enhanced bovine bone product, is the same (fairly) dense material as OsteoGraf®/N, but with a synthetic, short chain peptide attached. The cell binding domain of Type I collagen is synthetically reproduced and attached to the bovine bone. The first step in regeneration is cell binding. Therefore, the synthetic peptide (P-15) allows for a more rapid cell attachment to the particle which begins the regenerative process. However, without the porosity, osteoclastic activity is minimized.

Bovine bone has been used successfully in dentistry for many, many years. An excellent filler and provides a scaffold for new bone apposition. However, histology's have clearly illustrated the new bone formation is restricted to around the particles and some bridging of new bone formation between the particulate material. The full resorption of bovine bone has not been seen.

Resorption time is ALWAYS patient and site dependant resorption time is affected by porosity. The more porous, the faster it will resorb all cells (with the exception of the red blood cell) need to be stable to begin their normal physiological function

which results in resorption. The amino acid sequence found in P-15 enhances the cell binding / attachment beginning the resorption cycle. Bovine bone is too dense to completely resorb in a normal pH balanced site higher temperatures (above 600° C) will destroy residual protein and prions. However, these high temperatures will reduce the pore size and volume.

While bovine xenografts may reduce morbidity by eliminating the donor site, their disadvantage is the concern with the possibility of future bovine spongiform encephalopathy due to potential slow virus transmission in bovine-derived products (Bons et al 2002, Hunter 2002). One interesting xenogeneic transplant, Biocoral, is derived directly from the exoskeletons of corals from the Group Madrepora of the genus *acropora* (Guillemin et al 1987). These corals are harvested from the relatively unpolluted waters of the reefs off New Caledonia, a point of importance since coral from contaminated waters can contain petrochemical impurities. These are composed largely of calcium carbonate and are osteo conductive. The enzyme carbonic anhydrase, liberated by osteoclasts, is responsible for the breakdown of this material with the total time being approximately 18 months (Roux et al 1988).

Allografts and Xenografts can vary strongly in porosity, water, mineral and organic content. This is caused by extremely variable compositions depending on donor site, living condition and age of the donor (Tadic & Epple 2004, Grynypas 1995).

C-Graft (Clinician's Preference) is an apatite that has been hydro thermally processed from marine algae in a way that does not destroy the microstructure (Ewers & Simons 1992). The highly porous interconnecting honeycombed structure allows for new bone ingrowth and total replacement of this graft material in a period of time that equates to the remodeling time of new, vital bone. Bone ingrowth requires a specific pore volume and pore size. The processed marine algae, C-Graft, is approximately 68% porous with micropores that allow bone tissue to grow both on and into the particulate material. (Kasperk et al, 1988) C-Graft became available in the United States within the last year. Prior to this U.S. introduction, it has enjoyed wide use and documented clinical efficacy in numerous other countries for almost 15 years. Several documented cases and clinical studies have compared the histological and histomorphometric analysis of C-Graft to bovine derived materials resulting in evidence of superior bone quality and quantity. (Ewers et al, 2004) Although the bovine HA is microporous as well as inert and biologically compatible, it

provides only a scaffold for new bone to develop around the particulate material and bridge the (essentially) non-resorbable particles together. C-Graft's honeycombed chamber has shown to remodel to bone while providing a hospitable habitat within the actual porosity of the particle. (Schopper et al, 2003)

Alloplastic bone substitutes are synthetic substances that have been processed for clinical use in osseous regeneration. (They) Non-resorbable alloplastic materials have been shown to act as filler materials because of their sintering density at high temperatures. Some complications seen with alloplasts are fibrous tissue encapsulation. In addition, studies have shown that "non-resorbable" materials may acts as a barrier to bone forming cells and behave as irritants because of their chemical and mechanical densities which result in exfoliation, fragmentation and migration of such particulates to other organs results in functional interferences with systemic complications. (Valen & Ganz 2002) Resorbable alloplastic materials are today available in many different modifications depending on the medical indications. In the last 30 years many research and development activities were done to identify the reasons of sudden particular decomposition, scar tissue formation and fibrous tissue encapsulation. By using these results, resorbable alloplastic materials have been developed that do not show these

complications today. There are 3 types of alloplastic substances in clinical use today: Hydroxyapatite (HA, $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$) is a tribasic calcium phosphate ceramic which was sometimes called tricalcium phosphate incorrectly. HA typically has a calcium to phosphate ratio of 1.67 (Han et al, 1984). It can be divided into 2 groups depending on its ability to resorb (Jarcho 1986, Alexander et al 1987, Ricci et al 1989, Brown & Constanz 1994). Non-resorbable alloplastic materials include Osteograft® D, Calcitite, and HTR. Resorbable alloplasts include Osteograft® LD, and Osteogen. The porous form allows rapid fibrovascular tissue ingrowth, which may stabilize the graft and help resist micromotion. Past studies have shown to be fibro-encapsulated and surrounded by macrophages and multinucleated giant cells. (Valen & Ganz 2002) (Examples: OsteoGraft®- Dentsply Friadent Ceramed, Inc.- HA as blocks and particulates) Other ceramics:

Tricalcium phosphate (TCP, $\text{Ca}_3(\text{PO}_4)_2$)- with a different stoichiometric profile has a calcium to phosphate ratio of 1.5 [(it is a mineral whitlockite and not HA)]. Bone has calcium to phosphate ratio of 1.3 to 1.7 so that TCP ceramics are in between these dimensions (Grynblas 1995, Morse & Kaminski 1975, Han et al, 1984, Hollinger et al 1989). Two different modifications of tricalcium phosphate are existing, alpha TCP

and beta TCP. These two modifications are different in the crystalline state: both have the chemical formula $\text{Ca}_3(\text{PO}_4)_2$ but the atomic positions are different what changes the properties. Therefore the alpha-TCP is thermodynamically more instable than beta-TCP what can be seen in a higher solubility/faster dissolution rate. By dissolution alpha-TCP crystallizes to non-resorbable hydroxyapatite. Caused by these circumstances alpha-TCP is hardly resorbable and can clinically be seen years after implantation (Merten et al, 2001.) Phasing pure beta-TCP (without traces of alpha-TCP or HA) show good and predictable results (Horch, 2004). Norian and Bone Source are Calcium Phosphate Cements (CPCs) which form Hydroxyapatite or apatite products while hardening. So they are HA materials! (Vitoss-Orthovia- available as solid piece, putties, or paste)

Cerasorb[®] is a pure phase tricalcium phosphate material (Tadic & Epple 2004). The principles were already developed in the early 70ies at the Battelle Institute. It consists of a pure phase beta-tricalcium phosphate and is available in different porosities for reaching different resorption properties: round shaped Cerasorb[®] granulate with a porosity of 35% for safe applications in sensitive defects (sinus floor elevations), polygonal-broken Cerasorb[®] PerioSet with a porosity of 25% for periodontal use were the defect stability in early stages is

more important than a high resorption rate, Cerasorb[®] MaxiPore with a high porosity of up to 65% for fast resorption results. The porosity in combination with a stable structure allows predictable resorption results without decomposition into small particles. The primary particle size of all Cerasorb[®] modifications is above 7-10 μm . As it was shown smaller particles can stimulate inflammatory reactions of the surrounding tissue through phagocytosis of by macrophages (Shimizu 1988). Thus small particles in the neighboring lymph nodes, as detected by DeGroot in the 1980ies, are avoided (DeGroot 1988). The sum of the following properties: synthetic material, heated higher than 1000°C, phase purity, particle size, stable structure and porosity leads to risk free, predictable and fast resorption results (Peters & Reif 2004).

Pro Osteon- Interpore Cross International, Inc. unique product based on sea coral which is converted from calcium carbonate to calcium hydroxyapatite. The advantage of this material is that the structure of the coral, which is similar to trabecular bone, is maintained. However, it is brittle and not suitable to load-bearing sites. This material has not currently been approved by the FDA for intra-oral use. Coral is a macro porous material not a microporous. The depths of the particles are basically non-

porous. The macro porous structure lends itself well to vascular ingrowth, but not as well for cellular invasion.

Interpore-200 which is a porous hydroxyapatite has been used in ridge augmentation, periodontics, and orthognathic surgery. It has a highly interconnected, uniform and consistent three-dimensional porosity. It is approximately 55 to 65 % porous with pore diameters of 200 micron. The porosity is filled with woven and lamellar bone within 3 months with biomechanical properties similar to those of a cancellous bone graft. In animals, Interpore-200 exhibits 0 to 5 per cent biodegradation per year. **(White & Shors 1986)**

Bioglass (Bioactive glass) - a biologically active silicate-based glass. Silica-phosphate chains have been used in dentistry as restorative materials such as glass ionomer cement. Its high modulus and brittle nature make its application limited, but it has been used in combination with to form bioactive bone cement and with metal implants as a coating to form a calcium-deficient carbonated calcium phosphate layer. This layer facilitates the chemical bonding of the implant to surrounding bone (Ziffe et al 1991, Merkx et al 2003). They may have osteo-conductive properties tested in animal trials (Turunen et al 1997) and have been used in treatment of periodontal bony defects (Nasr et al

2000, Yukna et al 2001) but have been shown to have incomplete resorption.

(Examples: Biogran -3i, Inc.; PerioGlas -US Biomaterial, Inc.)

The body does not recognize silicate and never resorbs the material. Histologically one finds osteoid formation around the silicate, Calcium sulfate- also known as plaster of Paris. It is biocompatible, bioactive, and resorbable after 30-60 days. The time of resorption is faster the new bone can be formed.

Significant loss of its mechanical properties occurs upon its degradation; therefore, it is a questionable choice for load-bearing applications.

(Examples: Capset- LifeCore,

SurgiPlaster- Bio-lok. Osteo set-Wright Medical Technology,

Inc.- tablet for defect packing; Allomatrix- Osteoset combined with DBM in putty or injectable paste. Osteo set is currently

not utilized for intra-oral use) Polymers including

polymethylmethacrylate and hydroxyethyl methacrylate. (Example-

HTR) are non-resorbable. They serve as great dimensional

stabilizers, which can be utilized very effectively to plump out tissue and preserve ridge contours

Studies:

(Pansegrau et al 1998). Compared the rate and degree of

osseointegration of dental implants when placed into autogenous corticocancellous bone vs. freeze-dried corticocancellous bone

grafts. 30 experimental and 15 control implants were placed in 15 dogs. Implants were harvested at 1, 2, and 3 months and evaluated by light microscopy, microradiography, and histomorphometry. Results concluded that at both 2 months and 3 months, implants placed in autogenous bone osseointegrated to a significantly greater degree than freeze-dried bone.

(Becker et al. 1996) FDBA allografts are similar to synthetics and do not induce bone growth, but allow bone to fill around the non-vital particles by osteoconduction. MFDBA does not appear to be an ideal graft material. Non-decalcified bone matrix elicits extensive inflammatory reactions with macrophage and giant cell infiltration and minimal to an absence of bone formation. It is not resorbed by osteoclasts but rather giant cells. Giant cells apparently do not have the potential to initiate the coupling mechanism, which is necessary for activation of osteoblastic activity. Therefore, MFDBA does not have the potential to initiate bone induction and elicits an inflammatory reaction, which may interfere with bone formation.

(Forum 2002) Combination Techniques:

In a field test combining results of many practitioners, it was reported that the addition of autogenous bone to FDBA significantly improved clinical results. 63% of sites treated

with FDBA alone had more than 50% bone regeneration. The composite of autogenous bone plus FDBA showed 80% of treated sites with more than 50% bone fill.

(Rummelhart et al. 1989) Study conducted to clinically compare MFDBA to DFDBA. 22 defects in 9 patients were grafted and evaluated based on standardized radiographs, presurgical and postsurgical soft tissue measurements and osseous measurements at time of reentry in 6 months. Findings revealed no significant difference between the two materials in term of osseous repair or clinical attachment gain.

(Becker et al. 1994) Bone-forming capacity of DFDBA and autologous bone in extraction sockets were evaluated histologically. 7 paired sites with grafted with DFDBA or autologous bone and reentered in 3 to 13 months. DFDBA sites although clinically looked similar to adjacent bone and sites were dense on probing, histologically revealed the presence of dead particles of DFDBA with no evidence of bone formation or osteoclastic resorption of the particles. Autologous sites revealed vascular channels with woven and lamellar bone.

(Schwartz et al. 1998) The variability in clinical outcome could be a function of differences in either DFDBA processing

techniques and/or donor characteristics. Wide variations in commercial bone bank preparations of DFDBA exist, even within the same bank. Several possible explanations could account for the varied clinical results including age and gender of donor. 27 batches of DFDBA from different donors and same bone bank were studied. Each batch was implanted bilaterally in the thigh muscle of nude mice. After 56 days, implants were excised and examined histologically. 70% induced new bone formation. The ability of DFDBA to induce new bone formation is suggested to be age-dependent but not gender-dependent.

(Guillemin, Mellonig, & Brunsvold 1993) Clinically evaluated the use of DFDBA in conjunction with expanded polytetrafluorethylene (ePTFE) membrane for the treatment of intraosseous defects. 15 advanced periodontal patients with 2 bilateral interproximal probing depths of equal or greater than 6 mm pockets. Defects from each pair were randomly treated with DFDBA alone or DFDBA with ePTFE. Each site was surgically reentered and measured at six months. Both group showed significant difference when compared to baseline but no difference between the groups in respect to bone fill or defect resolution.

(NYSDA 2002) Use of ABX:

There is an inconsistency of literature concerning the effect of the use of antibiotics on regenerative outcomes on humans. One study utilizing FDBA alone or in combination with autogenous bone showed that the use of antibiotics resulted in greater graft success. The advantage of using antibiotics, however, is not unequivocal. Another series of studies showed antibiotics use did not improve clinical outcomes at one year.

(Becker et al. 1996) While case studies (Mellonig) present clinical documentation of a bone-like material with in treated defects, there's an absence of histological evidence that commercially-available allografts or intra-oral autologous bone chips induce bone formation or promote bone to implant integration. The rationale for using allograft is its osteoinductive potential (Urist 1965, 1983). However, histologically, there's apparently no evidence that commercially-available allografts initiate the cascade of events, which lead to significant amounts of bone formation.

Study:

4 types of grafts were evaluated:

A cortico-cancellous autologous graft and DFDBA

Autologous bone harvested from edentulous ridges adjacent to extraction sites or bone powder retained in drill flutes

DFDBA

FDBA

(5 grafts used with DFDBA and ePTFE membrane)

Grafts were placed into 21 extraction sites of 15 patients:

6 autologous bone

7 DFDBA (5 with membrane)

7 FDBA

1 onlay graft + DFDBA + membrane

Bone scoring system used:

0 = sections consisted of entirely dead implanted bone within connective tissue and granulation.

1 = mature, vital, host bone present, dead implanted bone particles not attached to host bone.

2 = dead implanted bone chips present, implanted bone attached to host bone by cement lines, granulation tissue present.

3 = dead implanted bone, mature host bone, blood vessels, cartilage, osteoblasts, new bone formation present.

4 = absence of implant, total restitution of original defect by host bone.

Biopsies at 4-13 months after grafting:

Autologous bone (2.33)- surrounded by minimally inflamed CT, blood vessels evident, vascularized channels and osteoblastic activity observed, mature host bone present.

DFDBA (0.98)- non-vital bone particles enmeshed in minimally inflamed CT.

FDBA (0.18)- combination of vital and non-vial bone, granulation tissue, no osteoblastic activity present.

Onlay Graft (0)- non-vital cortical bone, no revascularization, no osteoblastic or osteoclastic activity, granulation tissue present.

Presence of non-vital grafted spicules embedded in connective tissue was the overriding histological characteristic observed from all DFDBA and FDBA specimens. The non-vital particles were present regardless of graft type, presence or absence of barrier membrane, defect type, or time interval between graft placement and biopsy.

DFDBA sites demonstrated the presence of non-vital particles at all evaluation periods. Retention of DFDBA particles is related to the inability of osteoclasts to resorb demineralized bone. Mellado et al. (1995) at 1 year reported greater bone fill in periodontal defects augmented with ePTFE barrier only when compared with sites, which received DFDBA and barrier membrane. Findings suggest that augmentation of extraction sockets with grafting materials may actually interfere with the normal

healing process. There is a possibility that barrier membranes may actually delay initial vascularization of the grafted sites.

The variability in bone induction with DFDBA is probably associated with the varying amounts of human bone morphogenetic protein (hBMP) in commercially purchased DFDBA. Urist (1983) has demonstrated that large amounts of human bone are required in order to extract small amounts of human BMP. Bone is presumably formed by osteoconduction. Despite the presence of BMP in DFDBA, direct clinical comparison of treatment success using MFDBA (osteo conductive) and DFDBA (osteoinductive) yielded similar results.

(Yukna, D. Callan, J. Krauser et al. 1998) A synthetic cell-binding peptide (P-15) combined with anorganic bovine-derived hydroxyapatite bone matrix (ABM) was compared to DFDBA in osseous defects in 31 patients. The peptide component of P-15 is a synthetic clone of the 15 amino acid sequence of Type I collagen that is involved in binding of fibroblasts and osteoblasts. Relative defect fills clinically showed 87% with ABM/P-15 and 58% with DFDBA. Three patients were evaluated histologically. Only perio defects were studied.

(Young et al. 1999) Investigated the possible use and ultimate fate of anorganic xenogeneic bone (BioOss®) compared with autogenous bone with regards to the response of surrounding CT and possible resorption by multinucleated cells. 19 New Zealand white rabbits had four bone defect sites prepared and filled with autogenous, BioOss® + autogenous, BioOss® alone, and control. Results after 12 weeks showed that autogenous bone was actively resorbed and new bone was formed in close apposition to the particles. In contrast, the xenogeneic bone was degraded to lesser extent and new bone was apparent adjacent to bone particles without signs of resorption. Therefore, it is questionable if xenogeneic materials are resorbable and if any side effects occur as a result of the material's tendency to linger in the recipients' bed.

(MacNeill et al. 1999) raises questions about appropriateness of using the rabbit model for evaluation of xenografts and allografts that require extended periods of time for resorption and replacement by bone, since rabbits show rapid healing.

(Froum, Gomez, Breault. 2002) In a recent comparative study of the treatment of intraosseous defects, a bioactive glass showed no significant difference to DFDBA in comparison of bone fill

(61.8% vs. 62.5%) and defect resolution (73.3% vs. 80.9%) respectively.

(Hallman et al. 2001, Merks et al. 2003) Although xenogeneic and allogenic materials can reduce the volume of autogenous bone needed, data from clinical histology indicates that not all xenogeneic and allogenic materials will be resorbed and replaced by autogenous bone with time. This may leave the augmented bone with a composite rather than a homogenous structure, which could influence future dental implant survival. In fact, autogenous bone without anorganic additives seems to result in the greatest amount of bone in sinus floor augmentation after a four to six month healing period. Bovine bone material and HA seem to result in the lowest amount of bone formed.

(Becker et al. 1998) Compared DFDBA, bovine, autologous and human BMP. 8 patients with 13 extraction sites and 6 implants placed concluded that only hBMP promotes extraction socket healing without interfering with normal healing. In addition, only sockets grafted with hBMP histologically contained vital woven and lamellar bone. Becker attributes differences in findings of other bone induction studies to the fact that controlled laboratory experiments in mice do not have the same histological outcome compared to commercially-prepared allogenic

bone in humans. Other studies rarely use histological findings in multiple human biopsies.

To date the goal of complete, predictable regeneration has not been attained. The 1996 World Workshop in Periodontics addressed osseous regeneration of periodontal defects. They established criteria for proof of regeneration that included in ascending order of importance:

Animal histology

Human controlled clinical trials

Human histology

For any treatment or material to be considered truly regenerative and make the "A list" of proven materials, it must have histologic proof of positive effect in humans.

Currently, only five treatment materials are acknowledged to fulfill the two most important criteria of human trials and human histology:

Intraoral autogenous bone

DFDBA

DFDBA + BMP

Expanded polytetrafluorethylene guided tissue barrier

Extensive root conditioning with citrus acid

All other treatment/materials are missing at least one of the three parts of the proof triad.

(Yukna et al. 2002)

Despite the many advances in bone graft substitutes, new materials and approaches to bone healing continue to be investigated. Due to the limitations of current materials and risks involved with autogenous grafts, current techniques are in development toward tissue engineering. One company, Laurencin Laboratories, have created a porous bio mimetic matrix that provides an osteo conductive surface for osteoblast attachment and an interconnected pore system to allow cellular proliferation and migration.

Another approach is directing differentiation of mesenchymal stem cells to Therapeutics, Inc. They currently have several products under development based on stem cells with varied applications including regeneration of new bone, cartilage, tendon, cardiac muscle, and adipose tissue.

Conclusion

Published clinical studies have clearly demonstrated the clinical feasibility and histological possibility of bone regeneration as well as enhanced preservation of the alveolar ridge following dental extraction utilizing numerous synthetic and biologically

derived materials. Regeneration or preservation of the alveolar ridge following tooth loss is most predictable if the characteristics of the different grafting materials are related to the physiologic bone healing process. The selection of an appropriate bone graft material should be based on documented clinical studies as well as the characteristic and qualities of the material relative to the time of implant insertion. The material must create scaffold, preserving the desired alveolar volume, conduct or induct new vital bone and preferably resorb as it is replace with vital bone. The graft material must have adequate density to stabilize the implant after insertion and then remodel in such a way that an osseous integration can occur between the implant and the vital bone. Following implant insertion the implant should internally stimulate the bone and thus assist in preserving the alveolar volume and contours. The rate of resorption and the preservation of the scaffold should be considered as it relates to the proposed time of implant insertion. The longer the time for implant insertion the slower should be the resorption characteristics of the bone grafting material. This will allow for preservation of the scaffold and the alveolar volume without the benefits of the internal stimulation produced by the endosseous implant. The shorter the predicted time of implant insertion the faster the resorption characteristics of the material can be allowing for faster

replacement with new vital bone while being internally stimulated by the endosseous implant. Combination of different grafting materials may possibly be mixed so as to alter the resorption time and spacing between particles, but this article addressed the material qualities of each bone grafting material in it's pure form.

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