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Tissue engineering for periodontal tissue regeneration

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INTRODUCTION

In 1993, Langer and colleagues proposed tissue engineering as a possible technique for regenerating lost tissue, and the restoration of various human tissues and organs is starting to become a reality. The concept of tissue engineering was introduced originally to address the chronic shortage of donated organs. This approach reconstructs natural target tissue by combining three elements: a scaffold or matrix, signaling molecules (for example, growth and differentiation factors and genes), and cells. Current approaches to tissue engineering can be divided roughly into two main types: ex vivo and in vivo. In the former, the target tissue is created in a laboratory by culturing cells on biodegradable scaffolds in the presence of specific trophic factors before their transplantation into the body. In the latter approach, the three elements mentioned above are placed into a tissue defect ‘‘in situ,’’ and the tissue is restored by maximizing the natural healing capacity of the body by creating a local environment that is favorable for regeneration.

Protein-based approaches

Growth and differentiation factors can regulate the adhesion, migration, proliferation, and differentiation of various types of cell by binding to appropriate receptors. Recent advances in genetic engineering technology have made it possible to obtain large quantities of human recombinant proteins. The use of growth and differentiation factors is the most popular tissue engineering approach for regenerating periodontal tissues. So far, several growth factors including transforming growth factor-b (TGF-b) superfamily members, such as bone morphogenetic protein-2 (BMP-2), BMP-6, BMP-7, BMP-12, TGF-b, basic fibroblast growth factor (bFGF), and platelet-derived growth factor (PDGF) have been used to regenerate periodontal tissues. Moreover, studies are underway currently to test the clinical potential of some of these factors.

Platelet-rich plasma (PRP) contains several platelet-released growth factors, including PDGF and TGF-b. PRP therefore represents an autologous growth factor cocktail that can be harvested from patients with minimal invasiveness and used for applications in oral and maxillofacial surgery. PRP stimulates the proliferation of human osteogenic cells and periodontal ligament cells. However, the benefits of this approach remain controversial: although some investigators have reported positive effects on bone formation others have failed to detect any improvement. This discrepancy in wound-healing outcomes might be explained partly by interindividual variation because PRP is an autologous resource. Moreover, the optimal concentrations of the calcium and thrombin, which are the inducers of platelet activation leading to growth factor release, are unclear.

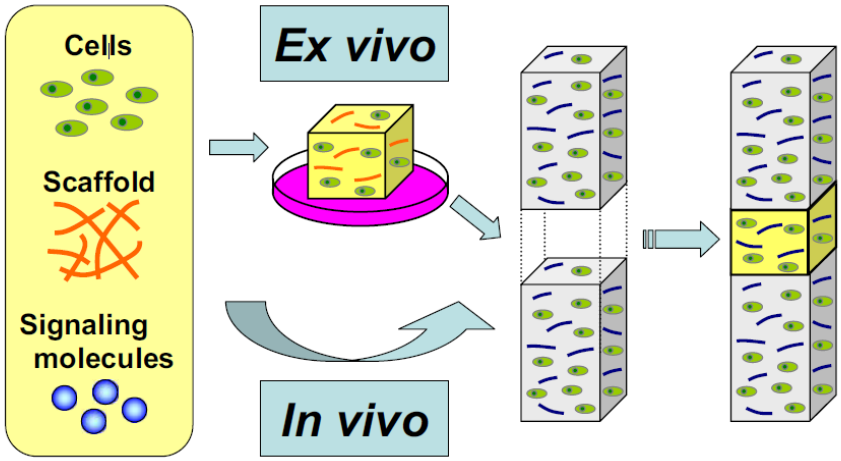


Fig. 1. Schematic representation of two major approaches to tissue engineering. First technique is to create tissue or organs in culture room by combining three elements (scaffold or matrix, signaling molecules, and cells) before transplanting tissue-engineered organ into patients (ex vivo approach). Second technique is to induce intrinsic healing activity at site of tissue defect using these three elements (in vivo approach).

If such a cell source and scaffold can be supplied at the site of the defect, it has been shown by case studies to induce bone formation successfully . Further information obtained from basic research at the cellular and molecular level could help this to become a reliable technique for assisting wound healing. The combination of a drug delivery system (DDS) with PRP may be effective, as the following paragraph shows. The development of synthetic or natural polymer-delivery vehicles for the sustained release of growth and differentiation factors will be crucial for their clinical utility. Locally-applied growth factors are defused away rapidly from the implantation site and have a short half-life as a result of a combination of physical and biologic degradation mechanisms. DDS using biomaterial vehicles has allowed the tissue-exposure time to be extended and protein stability to be maintained within the body. Nakahara and colleagues investigated the effectiveness of a tissue regeneration device combining a collagen sponge-scaffold material and gelatin microspheres, which prolonged the period of bFGF release in beagles with artificially prepared intrabony periodontal defects. This report was the first to demonstrate the usefulness of DDS and cell scaffolds for targeted periodontal regeneration using a full-scale animal experimental model.

In this controlled delivery system, positively charged bFGF molecules formed an ionic complex with acidic gelatin and were released gradually as a result of the degradation of the vehicle in vivo over an extended period. In the bFGF-treated group, 4 weeks after implantation, numerous capillary vessels were observed within the regenerated tissue around the residual gelatin vehicles containing bFGF. This observation indicated that the powerful angiogenic activity of bFGF still was present at this stage. These findings imply that a rich vascular supply is essential throughout the healing process to facilitate periodontal regeneration.

Currently, most delivery approaches involve a single growth factor. These techniques might be unable therefore to induce well-developed vascular networks leading to tissue regeneration, as angiogenesis results from a complicated series of cellular and molecular interactions. However, recent advances in polymeric technologies have allowed the delivery of multiple angiogenic growth factors with distinct kinetics by a single scaffold. For example, Richardson and colleagues reported the development of a new porous polymer scaffold that can deliver vascular endothelial growth factor (VEGF) and PDGF-BB. In this system, VEGF, which stimulates the outgrowth of immature vessels consisting of naked endothelial cells, is mixed with the polymer scaffold, resulting in its rapid release from the vehicle during the first days or weeks after implantation. Meanwhile, PDGF-BB, which stimulates the maturation and stabilization of nascent vessels by way of the recruitment of smooth-muscle cells, is pre-encapsulated within microspheres in the polymer scaffold, from which it is released by degradation in a delayed fashion. The controlled delivery of VEGF and PDGF-BB significantly increases the maturity of the resultant vessel networks. Similarly, Cao and colleagues reported that a combination of PDGF-BB and FGF-2 synergistically induced stable vascular networks, whereas single growth factors were unable to maintain the newly formed blood vessels. Such dual growth factor-delivery technologies could be useful in periodontal tissue engineering.

Cell-based approaches

Cell transplantation is currently a hot topic in the medical field and cellbased therapy using autologous cells is expected to play a central clinical role in the future . Several preclinical studies using mesenchymal stem cells (MSCs) have shown efficient reconstruction of bone defects larger than those that would spontaneously heal (that is, critical size defects). Dental cell-seeding studies have attempted to regenerate periodontal tissues since the 1990s, although clinical applications have become realistic only in recent years. Kawaguchi and colleagues used bone-marrow-derived MSCs in combination with atelocollagen to regenerate periodontal tissues in experimental class III furcation defects in dogs. After cell expansion for 2 weeks, autologous MSCs mixed with collagen gel were transplanted into the defects. In the MSC-treated groups, significant periodontal tissue structures were observed 1 month after implantation compared with the collagen gel group.

However, at the experimental cell concentrations, no significant difference was observed between the extent of regeneration of the bone and cementum tissues. The investigators concluded that additional studies using different scaffold materials and a various range of cell concentrations would be required to obtain conclusive results. Akizuki and colleagues used autologous periodontal ligament cells obtained from extracted tooth roots to fabricate cell sheets using a temperature-responsive cell-culture approach based on cell-sheet engineering.

A special culture dish, in which the dish surface is hydrophobic under normal culture conditions at, allowing cells to attach themselves to it and grow but becomes hydrophilic at 20_C so that cells detach themselves spontaneously, was used. This process enabled the collection of the confluent cell cultures as a single sheet in which the deposited extracellular matrix and cell-cell junction proteins remained intact, in contrast to traditional enzymatic treatments for cell detachment, which damage the cultured cells. Autologous periodontal ligament-cell sheets along with a reinforced hyaluronic-acid carrier were implanted into experimental dehiscence defects in dog molars. After 8 weeks, significantly improved periodontal tissue regeneration was observed compared with control cases that received the hyaluronic-acid cell carrier alone. Although further studies are needed to confirm the reproducibility of these regenerative effects, cell-sheet engineering has shown great potential as a new cell-based periodontal therapy. Cell transplantation has been shown to promote periodontal regeneration compared with the carrier alone as the control. However, it remains unclear whether the transplanted cells differentiate into osteoblasts, cementoblasts, and fibroblasts to form bone, cementum, and periodontal ligament, respectively or whether they recruit surrounding host cells to facilitate the regeneration of the periodontal tissues.

Gene delivery-based approaches

Numerous tissue regeneration studies have investigated various gene-delivery techniques . These techniques involve a gene encoding a therapeutic protein being introduced into cells, which can then express the target protein. This technique avoids the problems associated with the protein-delivery method by maintaining constant protein levels at the site of the defect. Genetic engineering approaches generally consist of two modalities: in vivo and ex vivo gene delivery. In the former, gene constructs, such as expression plasmid DNA or a viral particle, are physically entrapped within a scaffold or matrix. When the scaffold containing the gene constructs is implanted into the tissue defect, the host cells migrate into the implant, take up the gene constructs and start to produce the encoded protein. By contrast, in the latter approach, cultured cells are transfected (in nonviral delivery systems) or transduced (in viral delivery systems) with gene constructs in vitro before they are transplanted into the tissue defect. Jin and colleagues investigated gene therapy by incorporating BMP-7 and PDGF-B genes into adenovirus vectors. Rat syngeneic dermal fibroblasts were transduced ex vivo with adenoviruses encoding BMP-7 (Ad-BMP-7). These cells were then seeded onto gelatin sponges and placed into periodontal osseous defects. Ad-PDGF-B was used for in vivo direct gene transfer. This vector was initially mixed with a collagen matrix before implantation into rat periodontal alveolar bone defects. These adenoviral gene-transfer approaches stimulated regenerative activities in the periodontal tissues, including osteogenesis, cementogenesis, and periodontal ligament formation.

Remarks and future directions

Which is more important target for tissue engineering, teeth or the tissues supporting them? In previous decades, numerous studies have investigated the regeneration of periodontal tissues as introduced in the present review. Recently, the focus has shifted, with two studies attempting to regenerate teeth. However, although these groups successfully reconstituted the individual structural elements that make up a tooth crown (that is, the dentin, enamel, and dental pulp), neither managed to regulate the morphogenesis of the crown or to regenerate the tooth root. Further improvements and innovative approaches will be required to reconstruct ‘‘complete’’ teeth using tissue engineering. Despite the recent interest in tooth regeneration, the view remains that, in a clinical setting, the fundamental goal is actually the regeneration of periodontal tissues, particularly centering on the periodontal ligament. If the periodontal ligament could be regenerated, an artificial tooth implant would suffice, yielding almost natural occlusion and mastication accompanied by real sensation while chewing.

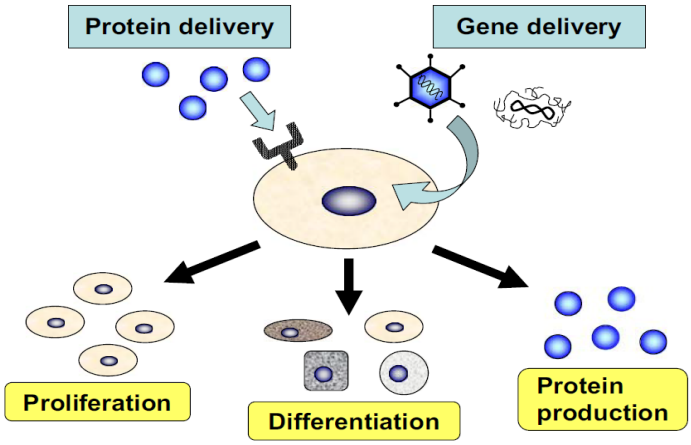


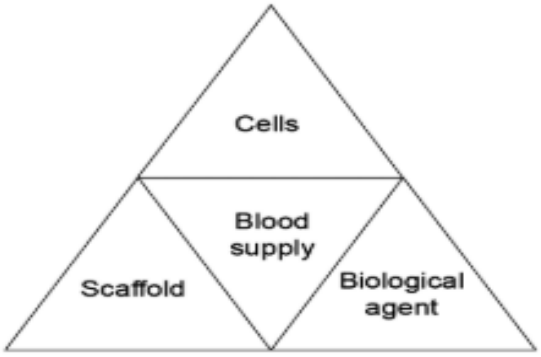
Fig. 2. Schematic representation of activation of stem cells through tissue-engineering approaches. Locally applied therapeutic proteins bind to appropriate receptors displayed at cell surface. Subsequently, cells are activated and undergo proliferation or differentiation. After taking up gene constructs through in vivo or ex vivo gene transfer using a plasmid or viral vector, genetically modified cells can either be seeded or migrate into scaffold, where they continuously secrete transgene-encoded therapeutic proteins into surrounding tissues.

It is difficult to judge which theory is correct, but talking to patients about realizing the dream of regenerating teeth has revealed the huge impact that such technology would have on many of their lives. By extension, patient demand clearly defines the ultimate goals for the development of advanced regenerative dental techniques. Thus, the regeneration of teeth is likely to propose an important research topic, which is intimately related to the natural reconstruction of periodontium. Guided tissue regeneration (GTR) was clinically applied in dental regenerative therapy before any other medical field, and the enamel matrix derivative, Emdogain, is the first periodontal therapy based on a biologic approach.

These techniques are examples of first-generation regenerative therapies. The various attempts at tissue regeneration, which come under the general heading of ‘‘tissue engineering,’’ introduced in the present review should probably be classed as ‘‘second-generation’’ regenerative medicine. This second-generation approach is making surprisingly rapid progress and its remit has expanded from its original application in the medical field to various related disciplines in which the introduction of biologic and engineering knowledge and skills are allowing the development of new approaches.

In the near future, third-generation periodontal therapies will involve nanoscale science and moldless manufacturing technology commonly known as rapid prototyping (RP) or solid free-form fabrication (SFF). These scientific and technologic innovations will make it possible to fabricate complex scaffolds that mimic the different structures and physiologic functions of natural fibro-osseous tissues, including those, such as periodontium, which consist of hard and soft tissues. The advancement of such technology might also make it possible to produce patient-specific cell-scaffold constructs with optimal distribution of cells and high vascular permeability.

Requirement for tissue engineering



The engineered tissues have to have sufficient biomechanical strength, architectural properties and space-maintaining ability. The engineered tissue has to maintain space for in-growth of alveolar bone, but it also has to be exclusionary with respect to the epithelial tissues to prevent the formation of a long junctional epithelium. In addition, biological functions have to be appropriate to allow cellular recruitment and proliferation, vascularization and the delivery of the appropriate factors for regeneration .

Two types of tissue engineering based on roles of cells

Passive
Active

- Passive engineering : The cells are not actively participating in the cellular changes.

  Eg. GTR-based therapies; barrier membranes
  Acellular dermal matrix (ADM)

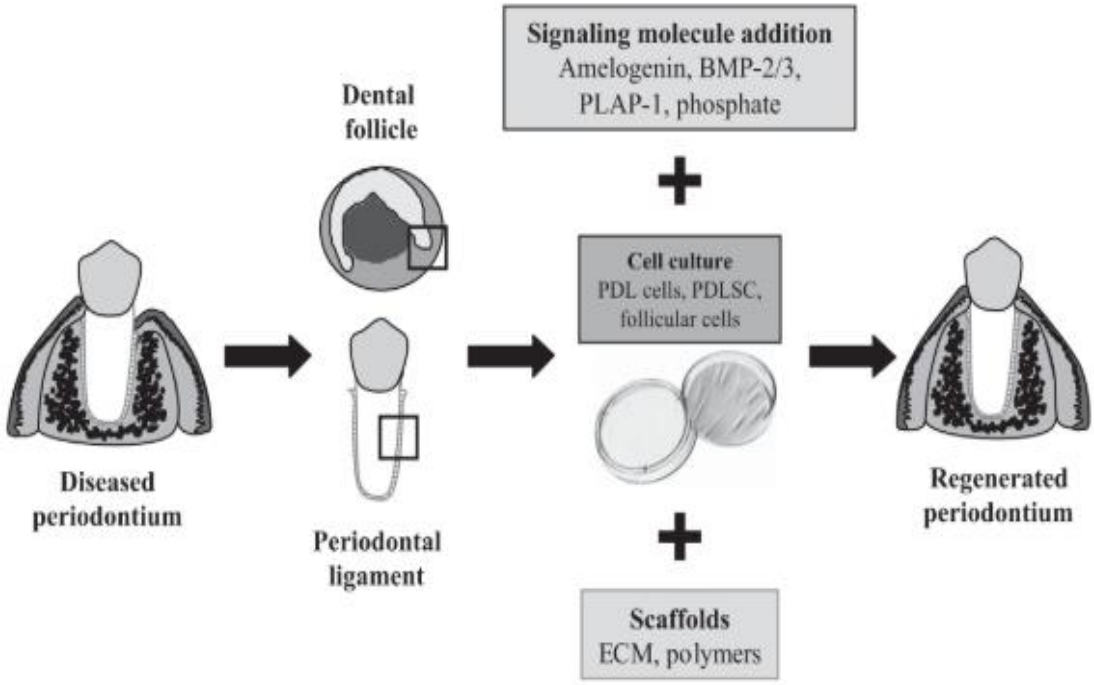
- Active tissue engineering : The cells are biologically active in the regeneration process.

  Eg. Enamel matrix derivative (EMD)

Growth factors: recombinant human platelet-derived growth factor-BB (rhPDGF-BB) plus β-TCP plus collagen wound dressing.

Cell therapy
Autologous fibroblast: Isolagen
Bilayered cell therapy (BLCT): Celltx
Human fibroblast-derived dermal substitute (HFDDS): Dermagraft

Cells obtained from the dental follicle and periodontal ligament are cultured in vitro, and signaling molecules that enhance cell differentiation into periodontal tissues may be added to the culture. Once identified, these molecules need to be delivered to the periodontal wound, in appropriate scaffolds.



Scaffold or Supporting Matrices

The use of scaffolding matrices to deliver growth factors to promote periodontal tissue regeneration.

Supporting matrices for engineering bone and soft tissue includes
Processed bone allografts,
Synthetic and natural polymers,

Synthetic ceramics,
Bovine type I collagen, and
Calcium sulfate .

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 Roles for the supporting matrices

1. Provide physical support for the healing area so that no collapse of the surrounding tissue into the wound site occurs.

eg. Bone allografts and synthetic ceramics such as TCP.

2. Serve as a barrier to restrict cellular migration selectively.

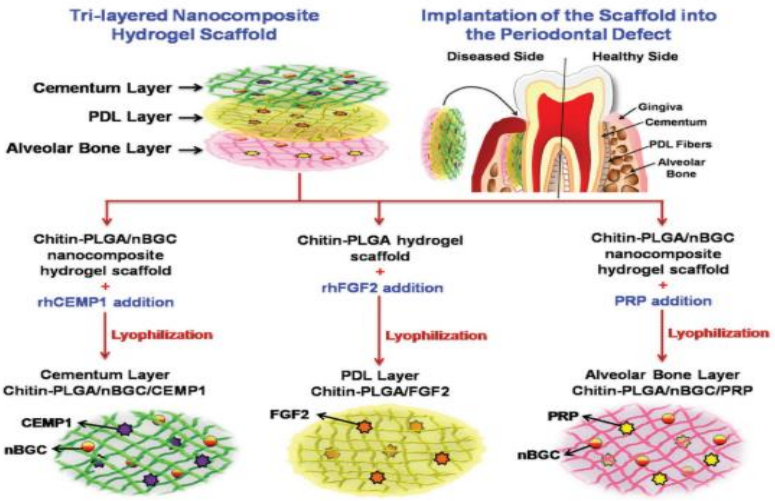
eg. GTR and guided bone regeneration (GBR) where nonresorbable PTFE and resorbable polylactate, polyglycolic acid, and calcium sulfate are used.

3. Serve as a scaffold for cellular migration and proliferation. eg. collagen matrix

A recent development in scaff old design for periodontal tissue engineering has been the use of multiphasic scaff olds

A multiphasic scaff old can be defined as a construct that incorporates variations in architectural organization (porosity, pore organization, etc.) and/or chemical composition, which aims to recapitulate the structural organization of the target tissue.

Multilayered scaffold



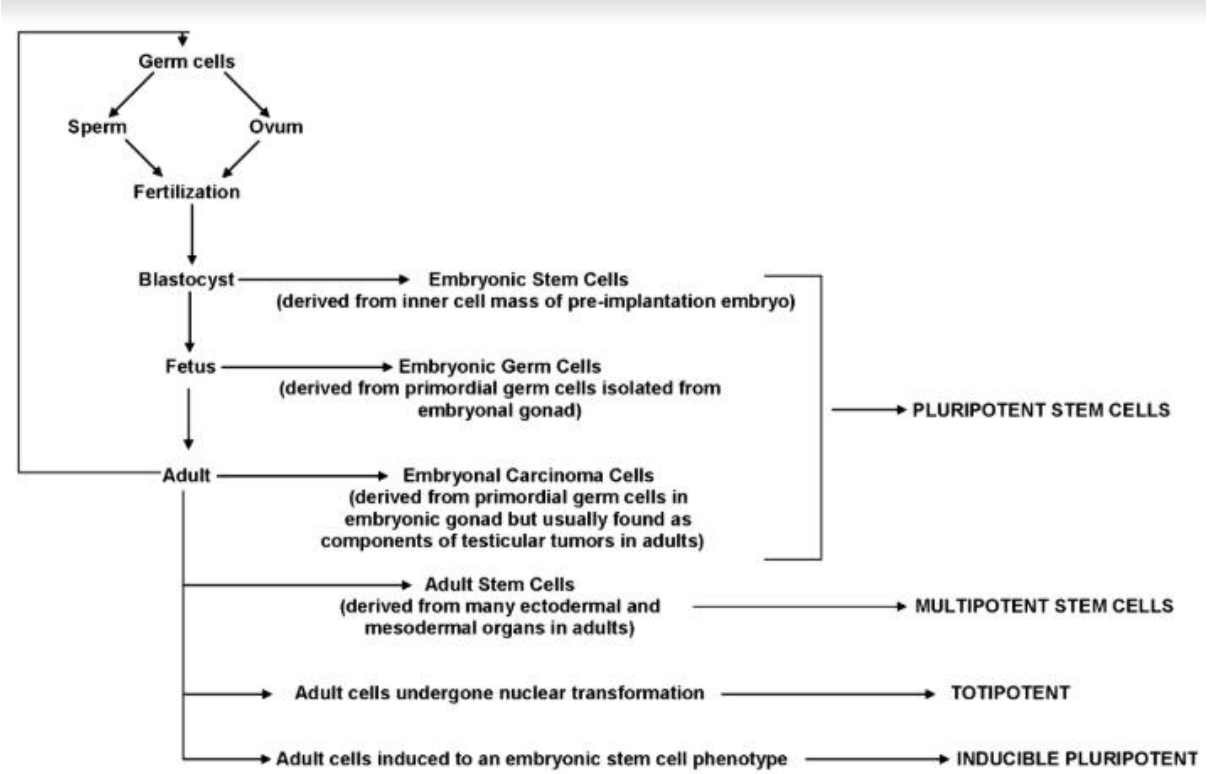
Stem cells are divided into two broad categories, embryonic stem cells and adult stem cells . Embryonic stem cells have two unique properties

(i) virtually unlimited proliferative potential in an undiff erentiated state
(ii) their pluripotency, which is the capability of diff erentiating into cells from all three germ layers

However, human embryonic stem cell research has been associated with major ethical concerns
Adult stem cells are found in the majority of fetal and adult tissues and are thought to play roles in long-term tissue maintenance and/or repair by replacing cells that are either injured or lost. They are generally multipotent stem cells that can form a limited number of cell types.

eg. Hematopoietic and MSC.

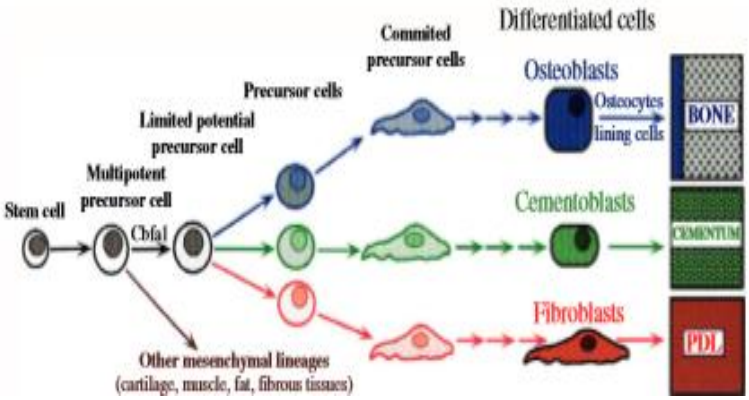
periodontium is mesenchymal in origin



Stem cell

A tissue engineering strategy for periodontal regeneration that exploits the regenerative capacity of stem cells residing within the periodontium. The literature supports the principle that cultured periodontal ligament cells applied in a scaffold or gel can induce periodontal regeneration around teeth. MSCs reside within the periodontal ligament and are responsible for tissue homeostasis serving as a source of renewal cells for cementoblasts, osteoblasts and fibroblasts throughout adult life. Cells present in the periodontal ligament, including cementoblasts, osteoblasts, fibroblasts, myofibroblasts, endothelial cells, nerve cells,epithelial cells and progenitor cell. These progenitor cells are enriched in locations adjacent to blood vessels .

Cellular differentiation of periodontal tissue



Signaling molecules

Two commercially available tissue engineering system for periodontal regeneration are:

Enamel matrix derivative(EMD)
Platelet derived growth factor(PDGF)- beta tricalcium phosphate(β- TCP). others
BMP type -1collagen sponge.
FGF-2

EMD (Enamel matrix derivative)

Effective in the treatment of infrabony defects. When EMD treatment was compared with GTR using bioresorbable membranes, the clinical results were comparable and stable over as much 10 yr. period. In a study comparing EMD, GTR, EMD in combination of GTR With OFD. All three had results are superior to those of OFD. No additional improvement when EMD used with GTR.When EMD used with autogenous bone graft ,DFDBA, xenograft, and bioactive glass. Additional improvement in clinical parameters were observed as compared with the use of either EMD or DFDBA alone. A recent review (Bosshardt 2008) concluded that

(1) EMDs increase the cell proliferation of periodontal ligament and gingival fibroblasts and cells of osteoblast and chondrocyte lineage;

(2) EMDs have biologic effects on cells of the osteoblast lineage, including up- regulation of markers of bone formation;

(3) specific small amelogenin polypeptides (5 kDa) have osteoinductive properties when tested in an ectopic bone- forming model;

(4) the evidence does not demonstrate an inductive role for EMDs on cementogenesis

Recombinant human platelet derived growth factor

- PDGF is 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.
- Dohan and colleagues showed a significant stimulation of human bone mesenchymal stem cells when in contact with L-PRF.
- This effect was dose-dependent during the first weeks in normal conditions and during the whole experiment in differentiation conditions. The cultures without L-PRF in differentiation conditions did not rise above the degree of differentiation of the cultures in normal conditions with L-PRF up to the 14th and 28th day, respectively.
- Recombinant human PDGF was used in conjunction with allogenic bone to correct class 2 furcations and interproximal infrabony defect on hopeless teeth.
- 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 clinical trial.
- Commercially available (GEM 21S , osteohealth) - combination of rhPDGF with a β-TCP

Advantages

Easy to use .
Requires no barrier membranes

BMP

- A group of regulatory glycoproteins that are members of TGF- β family 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 BMP7 (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.

Recombinant Human Fibroblast Growth Factor(rhFGF)

- 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.

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.

Advantage

- 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.
- MSC cellular allograft bone represents a unique, nonimmune material rich in MSCs, osteoblasts, and osteocytes.
- These cells displayed an “osteoinductive” molecular signature and the presence of MSCs surface markers that were more than 100-fold of what are found in iliac crest bone.
- cellular allograft has been used successfully in regenerative treatment of periodontal defects in both a single-rooted tooth and a multirooted tooth.

Periodontal ligament stem cells

A small population of multipotent stem cell populations termed periodontal ligament stem cells (PDLSCs).When cultured, these cells produce adherent clonogenic clusters that resemble fibroblasts and are capable of differentiating into adipocytes, osteoblast- and cementoblast-like cells in vitro, and demonstrate the capacity to produce cementum- and periodontal ligament like tissues in vivo. These cells have been identified within the periodontal ligament using a number of MSC-associated markers including STRO-1, CD146 and CD44. This led to their discovery in healthy and diseased periodontium and showed that they were mainly located in the perivascular regions and more widely distributed in diseased periodontium stem cell have properties of self-renewal and multilineage differentiation, PDLSCs offer significant potential for periodontal regenerative therapies.

Cell sheets and periodontal tissue engineering

A novel technique for using biologically driven approaches for regeneration involves the use of cell sheets . This requires the identification and isolation of cells required for periodontal regeneration and then growing these cells on a temperature sensitive sheet in culture plates. Cell sheet construction involves the use of a temperature-sensitive polymer biomaterial, poly-N-isopropylacrylamide (PIPA Am), in the cell culturing process. Once a mature cell sheet is formed, it is harvested by decreasing the temperature, which leads to the detachment from the temperature-sensitive substrate. This allows harvesting of a complete sheet of cellular material with an intact extracellular matrix and cell–cell junctions.

Computer Aided Design (CAD)/ COMPUTER Aided Manufacture(CAM)

CAD/CAM for tissue engineering was first introduced into the clinic in the 1990s, enabled surgeons to reconstruct trauma or tumor defects in an accurate manner . Medical technologies including procedures such as computed tomography, magnetic resonance imaging and 3D laser scanning has enabled the acquisition of site-specific digital 3D models. An impressive array of CAM techniques to create physical models. These models are very useful in the preoperative planning stages of advanced surgical regenerative procedures. In dentistry, CAD/CAM has found applications in dental implant placement performed using customized drill guides and this has significantly improved the accuracy of implant positioning and placement over conventional surgical guides. Furthermore, using medical imaging and CAD/CAM technology presurgical planning can be enhanced through the production of moulds and study models, which can be used for real-time implant placement in a controlled environment replicating the surgical site. In addition to the indirect use of such

customized models, which permit direct in vivo applications, such CAD/CAM technology is increasingly being utilized to manufacture anatomically precise scaff olds with flexible properties .

Nanotechnology

Nanotechnology is defined as the development and use of specific phenomena and direct manipulation of materials at the nanoscale. Nanotechnology is a multidisciplinary field and encompasses a diverse range of technologies derived from biological sciences, physics, chemistry, biochemistry, engineering and materials science. The development and introduction of biomaterials with nanoscale features with the ability to form ideal interactions with tissues has been a very significant development in the field of tissue engineering. Future developments in this field may well be further enhanced through the development of nanodevices and nanostructures for regeneration of damaged periodontal tissues.

Nano-sized hydroxyapatite has been developed to for periodontal regeneration and shown to have slightly improved clinical outcomes compared to traditional b-tricalcium phosphate for the management of intrabony periodontal defects. The recent development particularly on nanoparticles and nanotubes for periodontal management, the materials developed from them such as the hollow nanospheres, core–shell structures, nanocomposites, nanoporous materials and nanomembranes will play a growing role in materials development for the dental industry. Synthetic biologic systems that are self-assembling are based on known biological processes, which occur in tissues during the development and maintenance of tissue homeostasis. Through such approaches, systems can be constructed at the nano-, micro- or even macro-scale. An important feature of nano-constructed self-assembling materials is their ability to be constructed into nano-scaled domains and blocks to allow purpose built control and delivery system. Platelet-Rich Fibrin New techniques were investigated to overcome the disadvantages of PRP . Second generation .This resulted in the introduction of PRF by Choukroun and coworkers in 2001. PRF can be seen as an autologous biomaterial made of a fibrin matrix that contains the highest concentration of platelets , the highest concentration of growth factors, including platelet derived growth factor (PDGF), vascular endothelial growth factor (VEGF), and transforming growth factor (TGF)

Classification

- Based on the leukocyte content and fibrin structure platelet concentrates can be classified into four main categories
- Pure platelet-rich plasma (P-PRP) without leukocytes and with a low-density fibrin network after activation
- Leukocyte- and platelet-rich plasma (L-PRP) with leukocytes and with a low-density fibrin network after activation
- Pure platelet-rich fibrin (P-PRF) without leukocytes and with a high-density fibrin network .

Leukocyte- and platelet-rich fibrin (L-PRF) with leukocytes and a high-density fibrin network

Advantages L-PRF

It creates a bioactive construct that stimulates the local environment for differentiation and proliferation of stem and progenitor cells and it acts as an immune regulation node with inflammation control abilities, such as slow continuous release of growth factors over a period of 7 to 14 days.

L-PRF introduces the concepts of natural tissue regeneration (NTR) and natural bone regeneration (NBR). NTR is based on promoting regeneration of various periodontal tissues required for a normal tooth function (cementum, bone, periodontal ligament) with L-PRF membranes as filling material.

Autologous membranes, which have a dense fibrin network, are strong (one membrane can withstand a load of approximately 400 g before rupture) and have excellent biologic properties (rich in platelets, growth factors, and cytokines), opening many new clinical avenues.

PRGF may be used as a treatment modality for osteoarthritis, treatment of ulcers, tissue en platelet-rich growth factor (PRGF), which was first described by Anitua and coworkers.

L-PRF

Platelet concentrates used as a sole filling material or as a scaffold have emerged as a potential regenerative technique .They accelerate neoangiogenesis and stimulate the grafted cells enhancing bone regeneration. Leucocyte- and platelet-rich fibrin (L-PRF) is produced from a small peripheral blood sample that is immediately centrifuged without any anticoagulant. Coagulation starts during the centrifugation. Three layers are obtained:

Red blood corpuscles at the bottom of the tube .
Platelet-poor plasma on the top
Intermediate layer called “buffy coat” where most platelets and leucocytes are concentrated.

leukocytes in platelet concentrates is of great importance. They have potential antibacterial characteristics but can also regulate cell proliferation and cell differentiation. In addition, they are the basic cells responsible for the wound healing process and the first cells to start neoangiogenesis. In fact, they contain VEGF, which acts as a potent vascular growth factor. The leukocytes are also a source of production of the growth factors. During bone injury, monocytes and macrophages modulate the acute inflammatory response, produce growth factors such as bone morphogenetic protein 2 (BMP-2) and PDGF-BB, and induce osteogenesis in mesenchymal stem cells.

CONCLUSIONS

Complete and predictable regeneration of periodontal tissues lost due to trauma or disease presents a major challenge. Periodontal regeneration requires consideration of many factors that parallel periodontal development, including the use of optimal progenitor cell population, signalling molecules and matrix scaffold in an orderly temporal and spatial sequence. This requires better understanding of: (a) the mechanisms of self-renewal in order to sufficiently regulate adult stem cell growth in vitro to generate required cell numbers needed for different applications; (b) the regulation of stem cells during differentiation and maturation into tissue-specific cell types, as well as during wound healing; (c) the interactions between stem cells and the immune system, in particular, regarding use of allogeneic cell populations; and (d) mechanisms needed to control and prevent ex vivo-expanded mesenchymal stem cells from transformation.

REFERENCES

[1]. Xiao Y, Qian H, Young WG, et al. Tissue engineering for bone regeneration using differentiated alveolar bone cells in collagen scaffolds. *Tissue Eng*. 2003;9:1167.
[2]. Yang C, Hillas PF, Baez JA. The application of recombinant human collagen in tissue engineering. BioDrugs. 2004;18:103.
[3]. Takayama S, Murakami S, Shimabukuro Y, et al. Periodontal regeneration by FGF-2 (bFGF) in primate models. J Dent Res. 2001;80:2075.
[4]. Gault P, Black A, Romette JL, et al. Tissue-engineered ligament: implant constructs for tooth replacement. J Clin Periodontol 2010;37:750–758.
[5]. Hutmacher DW, Cool S. Concepts of scaffold-based tissue engineering–the rationale to use solid free-form fabrication techniques. J Cell Mol Med 2007;1:654–669.
[6]. Slavkin HC, Bartold PM. Challenges and potential in tissue engineering. Periodontol 2000 2006;41:41.
[7]. Jiang Y, Jahagirdar BN, Reinhardt RL, et al. Pluripotency of mesenchymal stem cells derived from adult marrow. Nature 2002;418:41–49
[8]. Srisuwan T, Tilkorn DJ, Wilson JL, et al. Molecular aspects of tissue engineering in the dental field. Periodontol 2000 2006;41:88–108.
[9]. Aukhil I. Biology of wound healing. Periodontol 2000 2000;22:44–50
[10]. Bartold PM, McCulloch CA, Narayanan AS, Pitaru S. Tissue engineering: a new paradigm for periodontal regeneration based on molecular and cell biology. Periodontol 2000 2000;24:2532–2569
[11]. Thomson JA, Itskovitz-Eldor J, Shapiro SS, et al. Embryonic stem cell lines derived from human blastocysts. Science 1998;282:1145–1147.
[12]. Hughes FJ, Ghuman M, Talal A. Periodontal regeneration: a challenge for the tissue engineer? Proc Inst Mech Eng H 2010;224:1345–1358.