Periodontal Complex Regeneration: From Hard to Soft Tissue Interface

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Periodontal disease, as a prevalent public health problem, could leads to bone resorption and ultimate tooth loss. Due to the nature of interfacial hard to soft tissue structure in periodontal complex, its wound healing is more complicated than normal oral mucosa after surgical procedures. To improve the success rate of surgical regenerative therapies in periodontal tissue, having insight to the natural tissue dissimilarities and biomechanical concepts in periodontium is crucial. Optimal periodontal regeneration contains restoration of the all three components of periodontal ligament, cementum and alveolar bone. In recent decades, many researchers have developed and characterized several strategies according to interfacial tissue engineering for periodontal regeneration. In the current review, the bio-clinical principles and current strategies for periodontal regeneration and future perspective are discussed.

Keywords: Interfacial tissue engineering; Periodontium; Growth factors; Mesenchymal stem cells; Scaffold

Introduction

Periodontal disease is recognized as one of the important public health problems in the world. The US Department of Health and Human Services designating oral health, reported the periodontitis as one of the 42 health issue known among healthy population in 2020 (1, 2).

Periodontal diseases is considered as an inflammatory conditions that affect periodontium as the supportive structure of the teeth, which could result in tooth loss (3). The periodontium is composed of the integration of all the gingiva, periodontal ligament, cementum and alveolar bone working as a functional unit to provide support and retention of teeth within an arch, and prepare peripheral defense (4). These components come together to maintain a proper anchorage of the teeth to the peripheral bone (5). In the healthy periodontium several signals and cells are continuously remodeling alveolar bone with tuned balance between formation and resorption process. However, this normal hemostasis can be disrupted by excessive inflammatory response to local or systemic perturbation, leading to bone and connective tissue attachment loss, and apical migration of the junctional epithelium (6-8). Periodontal complex wound healing after any surgical procedure is a more complicated process than normal mucosal wound healing, and it comes from the nature of interfacial hard to soft tissue structure there. So, post-surgical wound integrity is considerably vulnerable to mechanical force as the mucogingival flap rests on the hard and avascular root surface (9).

To efficiently restore the periodontal complex structure and to enhance the success rate of surgical regenerative therapies, it is important to get an insight to the natural tissue dissimilarities and biomechanical concepts in periodontium. In the normal dentoalveolar system, the presence of periodontal soft tissue between two hard mineralized bodies facilitates comparative micromovement between the tooth and bone (10).

An optimal periodontal regeneration includes restoration of the all three components of alveolar bone, PDL, and dental cementum. The periodontal ligament (PDL) is a fibrous connective tissue, which attaches teeth to the alveolar bone through a direct contact with the cementum, forming the wall of the socket. The PDL layer is about 0.2 mm in width between alveolar bone and tooth, constituted by dense collagen fiber bundles that provide the tooth adequate resistance to the masticatory forces and also act as the peripheral defense against opportunistic bacteria (7).

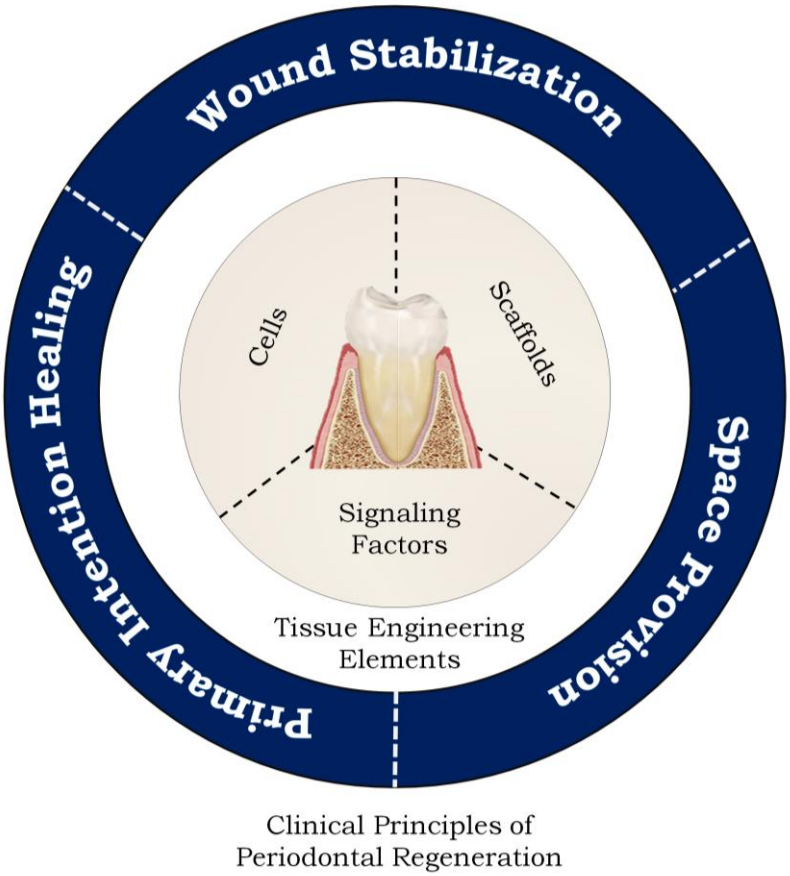

Figure 1. The tissue engineering elements and clinical principles for periodontal regeneration

Cementum is the site of adherence upon the superficial layer of the root of a tooth that connects to alveolar bone proper via the PDL (11). It is an avascular tissue that is histologically similar to bone. The cementum forming cells are cementoblasts, which are analogous to osteoblasts in bone—fully differentiated in lacunae arranged in a network of canaliculi (12). Unlike bone, these canaliculi do not channel blood vessels nor nerves; instead, they project to the alveolar space to provide nutrients to cementocytes via diffusion (13).

Alveolar bone is the foundation of the periodontium; therefore, it is vital to restore its integrity. Without bone, surrounding tissues cannot be reconstructed (12). The alveolar bone is composed of a thin, superficial layer of cortical bone fenestrated by many foramina that enable the passage of nervous tissue and blood vessels from the bone marrow to the PDL, providing vasculature to the periodontium complex (12). Approximately 50% of the bone proper is composed of inorganic hydroxyapatite, and matrix proteins and organic collagen constitute the residual 50% (12, 14).

The conventional regenerative periodontal treatments aim to decrease the fast progression of the disease, with the removal of debris and bacteria from the site of the wound, and create a proper alveolar bone architecture to maintain the periodontal health (15); However, the complete healing and functional recovery of the damaged periodontium are still not achieved (16). This review presents the bio-clinical principles and current strategies for periodontal regeneration (Figure 1).

Bio-clinical principles for periodontal tissue regeneration

The regenerative clinical strategies with the goal of restoring the loss of bone and periodontal attachments are comprising dental root biomodification, application of biomaterials and autologous bone grafts, guided tissue regeneration, as well as applying bone derivatives and wound biomodification factors (17). However, the surgical outcomes of periodontal regeneration have not been predictable yet (18).

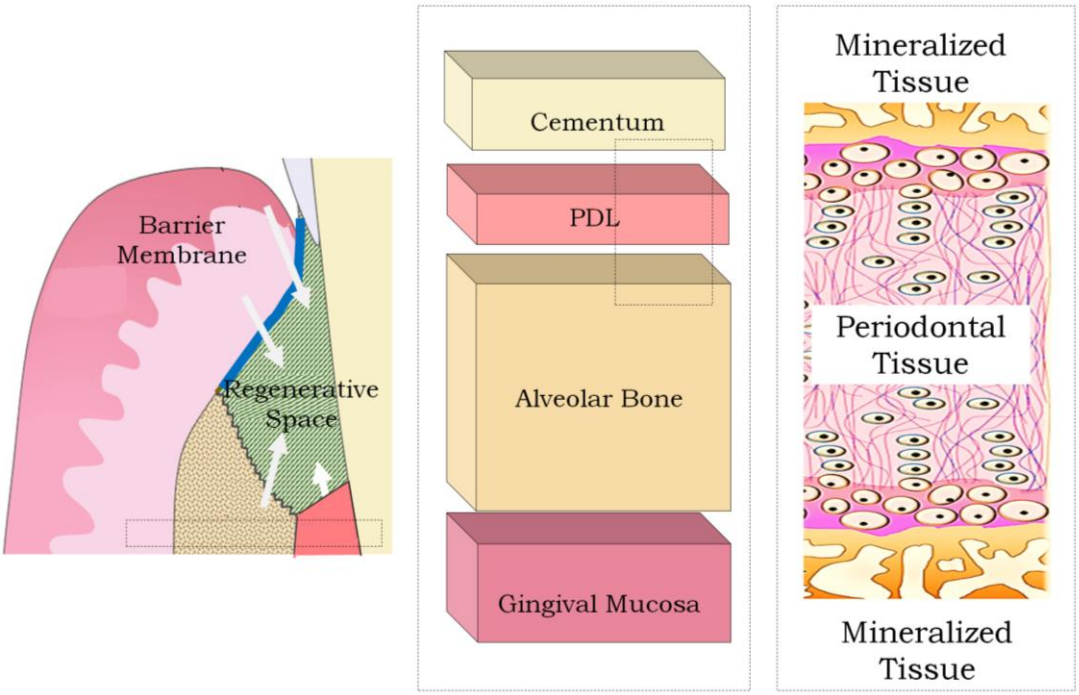

Figure 2. Hard to soft tissue interfaces in periodontal complex

Besides the limitation of current clinical regenerative techniques, it has been well documented that improper surgical management are responsible for many of limited and variable periodontal regeneration success. Wound stabilization, space provision and primary intention healing are three main critical factors which play the pivotal role in the consequence of periodontal regeneration attempt and can easily be underscored (9, 17).

The importance of the wound stability was illustrated by comprising the adhesion of the fibrin clot to the dental root surface with heparin solution (19, 20). The blood clot is formed between the root surface and the mucogingival flap post surgically not only to seal the wound area but also provide a preliminary matrix for cellular migration. The histological evaluation depicted that the un-attached fragile fibrin clot allowed progressive migration of the gingival epithelial cells into the point that the intact fibrin-clot inhibit the progression. This experimental treatment led to the formation of a long junctional epithelium, whereas the epithelium was stopped on the cemento–enamel junction in control defects (20). So, to harness the regenerative potential of the available regenerative techniques, the surgical procedures should comprise proper wound stabilization to decrease possible tensile forces, well post-surgical protection to avoid disruption by the mechanical insult and improve hard-to soft adhesion by root surface biomodification (21, 22). (Figure 2)

A common procedure used in bone regeneration process is bone grafting (23-25). It may be performed when the residual bony walls of the defect are still preserved and can provide vasculature and mechanical support for the graft. Because epithelial tissue regenerates rapidly, barriers are implemented to preserve space for regeneration of bone, known as Guided Bone Regeneration (GBR) (26). Both the barrier and bone graft may derive from a previously harvested, non-living source (allogenic) or from a donor site (autogenic). Autograft materials are more invasive by nature, predicated upon a second surgical site. Allograft materials are less invasive but also less potent. Either way, GBR yields regeneration with histological success but with unpredictable results (26).

Bioactive mediators including stem cells, growth factors and gene therapy agents are also implemented, having demonstrated a limited range of utility in clinical scenarios, as well as limited regenerative capacity. Results are similar to those obtained with bone grafting (16). Conventional clinical techniques have yet to produce results with complete regeneration in most cases.

Table 1. Scaffolds

Monophasic Scaffolds/Materials	Stem Cells	Animals Models or Clinical Trials	Target Tissue	REF
PGA	HPLCs (healthy tooth extracted)	Mice (subcutaneous bilateral implantation)	PDL	(27)
PLGA	hADSCs	Rats (critical size bone defects)	Bone	(28)
PCL	BMSCs	Nude Rat	Bone	(29)
	HPLCs sheets	In vitro study	Bone, PDL Regeneration	(30)
Alginate	PDLSCs and GMSCs	Nude mice	Cartilage Regeneration	(31)
Collagen/ PSEC	Gingival fibroblasts and PDL fibroblasts (third molars)	In vitro study (PDL healing properties of collagen barrier membranes)	PDL	(32)
Collagen hydrogel/tacrolimus	No stem cells were used	Rat	Bone	(33)
Gelatin/CHI/BaG	Osteoblast-like cells (MG-63)	In vitro studies	Alveolar Bone	(34)
CHI	HPLCs	In vitro study	PDL	(35)
	Rat BMSCs	Rats (periodontal defects)	PDL	(36)
Hyaluronic Acid	HPLCs (healthy tooth extracted)	In vitro studies	PDL	(37)
Hyaluronic Acid/ECM/ IL-1-Hydrogel	Gingival margin-derived stem cells	Pigs (periodontal defects)	Alveolar Bone, PDL	(38)
TCP	HPLCs	In vivo	Alveolar Bone, Cementum	(35)
BaG	BMSCs	Rats (bone defects)	Bone	(29)

Biphasic Scaffolds/Materials	Stem Cells	Animal Model	Target Tissues	REF
HA/ Collagen	Dog BMSCs	Dog (alveolar bone defect)	Alveolar Bone, PDL	(39)
PCL/Collagen	PDLSCs	In vitro studies	PDL	(40)
PCL/CaP	Cell sheets (PDL cells, gingival margin derived cells and alveolar bone derived cells)	Rats (periodontal defects)	Alveolar Bone, PDL, Cementum	(41)
PCL/BaG	Sheep BMSCs	Rats (bone defects)	Bone	(29)
	No stem cells was used	Rats (bone defects)	Bone	(42)
PCL (Periodontal Compartment)	Periodontal ligament cells (Merino sheep)	Rats (subcutaneous)	PDL	(43)
PCL/TCP (Bone Compartment)	Osteoblasts (Merino sheep)	Rats (subcutaneous)	Alveolar Bone	(43)
PLGA/TiO2 nanotube	No cells	Rats (bone defect)	Bone	(44)
PLGA/TCP	Gingival Fibroblasts transduced with BMP-7	Dogs (Class II furcation defects)	Alveolar Bone, PDL, Cementum	(45)
PCL/Gelatin	Cell sheets (PDL cells, gingival margin derived cells and alveolar bone derived cells)	Rats (periodontal defects)	Alveolar Bone, PDL, Cementum	(46)
PCL/Collagen/HP	Periodontal ligament stem cells (PDLSCs)	In vitro studies	Alveolar Bone, PDL, Cementum	(40)

PLGA/Hyaluronic Acid	No cells; used as a periodontal barrier	Rats (critical size bone defects)	Bone	(47)
PCL-PGE (PCE nanofibers)/CHI	Rat RBMSCs	Rats (periodontal defects)	Alveolar Bone, PDL, Cementum	(36)
HA/Collagen	Human and mouse MSCs	In vitro	Alveolar Bone	(48)
TCP/CHI	HPLCs	In vivo	Alveolar Bone, PDL, Cementum	(49)
Collagen/L-PRF	No stem cells used	Rabbit	Bone	(50)
CHI/Hydrogel/p-DNA-BMP-2	No stem cells used	Dogs (periodontitis model)	Alveolar Bone	(51)
CHI/Hydrogel/p-DNA-BMP-2	No stem cells used	Rats (calvaria defects)	Bone	(51)

Multiphasic Nanocomposite Scaffolds	**Stem Cells**	**Animals Models or Clinical Trials**	**Target Tissue**	**REF**
PLGA-Chitin/ nBGC/Cementum Protein 1	hDFCs	Rabbits (maxillary periodontal defects)	Cementum	(52)
PLGA-Chitin/ nBGC/FGF-2	hDFCs	Rabbits (maxillary periodontal defects)	PDL	(52)
PLGA-Chitin/ nBGC/ PRP	hDFCs	Rabbits (maxillary periodontal defects)	Alveolar Bone	(52)
PLGA/Collagen/HP	Osteoblast precursor cell line (MC3T3) from a mouse	In vitro studies	Alveolar Bone, PDL, Cementum	(53)
TCP/gelatin+ nanoCHI/rhBMP2	BFPSCs	In vitro studies	Bone	(23)
PCL/PLLA/CP/CHI	hADSCs	In vitro studies	Bone	(54)

PGA: Polyglycolic acid, hADSCs: Human adipose-derived stem cells, PLGA: Poly Lactic Glycolic Acid, PCL: Polycaprolactone, BMSCs: bone marrow stromal cells, PDLSCs: periodontal ligament stem cells, GMSCs: gingival mesenchymal stem cells, PSEC: platelet secretome, CHI: chitosan, HPLCs: human periodontal ligament cells, BaG: Bioglass, ECM: Extracellular Matrix, TCP: tri-calcium phosphate, HA: hydroxyapatite, PDLSCs: Periodontal ligament stem cells, MSCs: mesenchymal stem cells, hDFCs: human dental follicle stem cells, nBGC: nano bioactive glass ceramic, PRP: platelet-rich plasma, BFPSCs: buccal fat pad-derived stem cells, PLLA: Poly (L-Lactic Acid)

Tissue Engineering Approaches for Periodontal Regeneration

Although the conventional periodontal surgical approaches using bone substitutes, or biologics continue to offer reliable methods to restore the periodontium, reconstruction of large periodontal defects still remains challenging for the clinicians. Further, the limited access and resource of bone substitute and autologous grafts have led to attempts of developing novel tissue engineering techniques to achieve promising outcomes in periodontal regeneration.

Tissue engineering was introduced by Langer and Vacanti in 1993 as an interdisciplinary field that applies the principles of engineering and life sciences toward the development of biological substitutes that restore, maintain, or improve tissue function or a whole organ(55).

Periodontal tissue engineering consists of a combination of three important principles which include providing stability for the new tissue to growth, preventing the invasion of soft gingival tissue to the wound (56), obstruct the zone from the faster-proliferating gingival fibroblasts (57, 58), and offer proper biological signals to stimulate the repair of the periodontal tissue (59).

The cells of the epithelial and gingival tissue grow faster than periodontium cells. Therefore, these tissue cells frequently repopulate the void space of the periodontal wound, competing for the space that would ideally be occupied by mesenchymal

cells. The epithelial cells weakly attach to the dental root, forming a condition known as long junctional epithelium (LJE) (60). LJE is prone to relapse of periodontitis, thus, there is an increasing interest in studies of tissue engineering approaches for periodontal regeneration (57).

A wide variety of periodontal tissue regenerative therapies—like guided bone regeneration, application of bone substitutes (61-66) and platelet-rich plasma and fibrin (67-73) have been frequently used. Recently, induced pluripotent stem cells (iPSCS) were successfully applied for periodontal regeneration (74). These cells represented a promising source of stem cell with convincing regenerative outcomes (75, 76).

Scaffolds in Periodontal Complex Regeneration

Scaffolds are custom-made grafts used to facilitate tissue regeneration by providing physical support for cell adherence, growth and movement. (77, 78). Table 1 summaries several studies using different types of scaffolds for periodontal regeneration.

The porosity of scaffolds provides the stability and attachment required for the new cells to grow, migrate and differentiate, allowing the proper regeneration of the new tissue. Scaffolds can be categorized as monophasic (one compartment), biphasic (two compartments) and multiphasic (more than two compartments). The quantity of compartments determines the number of structures that can be regenerated at the same time.

Several biocompatible materials (bioceramics and polymers) have been reported in the literature to be successful in the creation of scaffolds (44, 79, 80). The materials chosen to create scaffolds for regeneration of periodontal tissue directly depend on the tissue to be regenerated. For instance, bioceramics and synthetic polymers are related with hard tissue (alveolar bone and cementum) regeneration, while natural polymers are associated to soft tissue (PDL and gingival tissue) regeneration (81).

Since the periodontium consists of both mineralized tissue, including alveolar bone and cementum, and soft tissues, including connective tissue, PDL and marginal gingiva, its regeneration can be performed using a multiphasic scaffold with three compartments (82). This multiphasic approach demonstrated success in an animal study by Sowmya S *et al.*, whom created a porous tri-layered nanocomposite hydrogel scaffold, and implanted it with or without growth factors into maxillary periodontal defects of rabbit (52). Compared to other groups, the tri-layered scaffold with growth factors exhibited complete defect closure and healing with new cementum, fibrous PDL and alveolar bone with well-defined bony trabeculae. Furthermore, Nabavi *et al.* fabricated a collagen hydrogel released tacrolimus, and demonstrated favorable outcomes for bone regeneration(33). The collagen hydrogel can release drugs or growth factors with the potential of periodontal tissue regeneration.

Growth Factors in Periodontal Complex Regeneration

Clinical studies showed that the innate regenerative potential in three walls bony defects is quite substantial. The defect architecture provides proper wound stability, and the presence of cellular and vascular resources facilitate healing process. Whereas, the regeneration of supra-alveolar defects, two- and one-wall intrabony defects, is naturally reduced. So, to enhance the success rate of regenerative techniques in such clinical scenarios, bioactive stimulation using growth and differentiation factors are considered (83-85).

A growth factor is any substance that induces the proliferation, differentiation and/or hypertrophy of a cell; usually, this substance is a type of protein or steroid that induces activation or inhibition on transcription factors downstream to induce the aforementioned effects (86). Growth factors have effects on not only the process of regeneration but also the development of the specialized tissues that make up the periodontium. They also act as important triggers for cell events, having an implicit role in the restoration of function of the tissue in question (4). Their action has the largest effect on precursor cells that are either predisposed to respond to a growth factor or have high potency due to being relatively undifferentiated (4).

Growth factor delivery to target cells, physiologically, is very specific and highly controlled; thus, clinical translation of growth factor therapies have been relatively poor (26). Tissue engineering involves the innovation of design strategies to deliver growth factors in a stable condition to the site of interest (86). The growth factors involved in periodontal tissue regeneration are platelet-derived growth factor (PDGF), insulin growth factor (IGF), fibroblast growth factor (FGF), transforming growth factor-beta (TGF-β), bone morphogenic proteins (BMPs) and enamel matrix derivates (EMD) (13).Combination of these growth factors is used to stimulate the cell proliferation, cell activity, tissue formation and extracellular matrix synthesis of human PDL cells, which are crucial processes for periodontal regeneration (57, 87, 88).

PDGF is a dimeric molecule that bears factor-BB homodimer, which is a potent mediator of periodontal tissue repair and regeneration. Accelerated wound healing and proliferation activity has been reported in periodontal ligament cells exposed to PDGF (89-93).

Table 2. Growth Factors

Growth Factor(s)	Carrier Materials/Matrix	Animals Models or Clinical Trials	Target Tissue	REF
BMP-2	CPC/Ceredex	Dog (supra-alveolar defects)	Cementum	(94)
	Calcium Phosphate/ Injectable Cement	Wistar rats (30 periodontal defects)	Alveolar Bone	(95)
	Collagen/Sponge	Baboon (intrabony defects)	Cementum	(96)
	Collagen/Sponge	Human (buccal wall defects)	Alveolar Bone	(97)
	Gelatin/Hydrogel	Monkey (6 mm diameter defect)	Alveolar Bone	(98)
	Dex-GMA Gelatin/ Pellet Scaffold Composite	Dog (periodontal furcation defect)	Alveolar Bone, PDL, Cementum	(99)
BMP-6	Collagen/Sponge	Dog (supra-alveolar periodontal defects)	Alveolar Bone, Cementum	(47)
BMP-7	Collagen/Sponge	Human (insufficient bone height)	Alveolar Bone	(100)
BMP-12	Collagen/Sponge	Dog (supra-alveolar periodontal defects)	PDL	(101)
PDGF	Bone Allograft/ Granules	Human (infrabony defect/molar class II furcation)	Alveolar Bone, PDL, Cementum	(102)
	PDGF-BB/Scaffold (b-TCP)	Human (localized periodontal defect)	Alveolar Bone, PDL, Cementum	(103)
	-	Human (localized periodontal defect)	Alveolar Bone, PDL	(104)
	Acellular Dermal Matrix	Human (localized periodontal defect)	Alveolar Bone, PDL	(105)
IGF	Dextran-Co-Gelatin/Pellets	Dog (periodontal furcation defect)	Alveolar Bone, PDL, Cementum	(106)
PDGF+IGF (synergism)	Methylcellulose/ Gel	Dog and/or Monkeys (ligature-induced periodontitis)	Alveolar Bone	(107, 108)
TGF-b	Calcium Carbonate/ Granule-Lump	Dog (supra-alveolar periodontal defects)	Alveolar Bone, Cementum	(109)
FGF-2	Collagen/ Membrane	Dog (class III defects)	Alveolar Bone, PDL, Cementum	(110)
	Hydroxypropyl Cellulose	Dog (3-wall periodontal defects)	Alveolar bone	(28)
	Calcium Phosphate/ Cement	Wistar rats (30 periodontal defects)	PDL	(95)
	b-TCP	Human (deep infrabony defects)	Alveolar Bone, PDL	(111)
EMD	EMD-gel	Human (deep infrabony defects)	Alveolar Bone, PDL, Cementum	(112)
	EMD/ Collagen Sponge	Monkeys (open flap debridement)	PDL, Alveolar bone	(113)
	Osteogain/ Collagen Sponge	Monkeys (open flap debridement)	Alveolar Bone, PDL, Cementum	(113)
	EMD/ Natural Bone	Human (deep infrabony defects)	Alveolar Bone, PDL, Cementum	(114)
	EMD/b-TCP	Human (deep infrabony defects)	Alveolar Bone, PDL, Cementum	(114)

FGF are heparin binding proteins that stimulate angiogenesis to defective area and fibroblast proliferation (13). Animal trials have demonstrated considerably enhanced periodontal regeneration in dogs (115, 116).

BMPs are disulfide-linked homodimers that induce osteoblast differentiation. Although favorable periodontal regeneration has been reported when applying these growth factors to periodontal defects, tooth resorption has been noted as a side-effect; thus, it has not been deemed suitable for periodontal therapy (117-119).

Human cementoblastoma-derived protein is highly expressed in cementoblast and is speculated to regulate cementoblast differentiation and mineralization of cementum matrix. Inflammatory mediators also have a considerable effect on periodontal tissue regeneration, inducing early growth response gene-1 (egr1), which is speculated to mediate anabolism in a murine cementoblastic lineage (120).

Platelet-rich plasma (PRP) is a strategy often implemented in regenerative medicine that exhibits potent regenerative properties. Many growth factors including PDGF, transforming growth factor-beta and insulin-like growth factor—are found in PRP preparation (13, 121). Use of PRP clinically has demonstrated decreased probing pocket depths in regenerated defect sites. Reports upon application of PRP in various defect sites have demonstrated variable results, suggesting efficient utility of PRP is not yet fully understood (122). Table 2 listed several growth factors used for periodontal regeneration.

Dental Stem Cells in Periodontal Complex Regeneration

Mesenchymal stem cells (MSCs) are defined as pluripotent progenitor cell derived from embryonic mesoderm. MSCs have the potential of multilineage differentiation, forming different tissues including bone (osteoblasts), cartilage (chondrocyte), tendon, ligament, marrow stroma, adipocyte, dermis, muscle and connective tissue (123, 124). MSCs can be found in the bone marrow, spleen and thymus. MSCs derived from bone marrow (BMMSCs) can differentiate into different cell lineages such as adipogenic, osteogenic, chondrogenic, neurogenic and myogenic cells (125).

The increasing interest regarding tissue regeneration technology has led to researchers demonstrating different dental tissues contain several types of MSCs. Stem cells from dental tissues were discovered at first by Gronthos *et al.* In fact, in their study, progenitor cells were isolated from human dental pulp with the term of 'post-natal dental pulp stem cells' (DPSCs) (126). Successively, stem cells have been isolated and characterized from exfoliated deciduous teeth (SHED) (127), PDL (PDLSCs) (123), dental follicle (PCs) (128) and apical papilla (SCAP) (129, 130).

Dental pulp stem cells (DPSCs), multipotent post-natal stem cells, not only have the ability of self-renewal, but also can differentiate into various cells responsible for the formation and maintenance of dentin organ and dental pulp. Gronthos *et al.* were the first to isolate DPSCs from human dental pulp (126); they presented the potential of multilineage differentiation and self-renewal of DPSCs, along with its capacity to regenerate a dentin-pulp-like complex both in vitro and in vivo (131). Hernandez-Monjaraz *et al.* (132) and Aimetti *et al.* (133) reported promising results of autologous and allogenous DPSCs for alveolar bone regeneration in intrabony defects. Moreover, according to a randomized controlled clinical trial, the application of DPSCs can significantly improve level of clinical attachment and bone fill after 1 year of follow-up (134).

Similar to DPSCs, stem cells from human exfoliated deciduous teeth (SHED) derived from the dental mesenchyme, termed ectomesenchyme. Miura M *et al.* discovered a population of cells with colonogenic and highly proliferation ability which can differentiate into different cell types such as odontoblasts, adipocytes, and neural cells in deciduous teeth. Also, they demonstrated the capability of SHED for bone and dentin formation in vivo, as well as survival in mouse brain, and expression of neural markers (127, 135). They proved that a different type stem cells could be isolated from primary teeth. Gao *et al.* showed a specific immunomodulatory role of SHED stem cells that induces M2 macrophage polarization and reduces inflammatory cells infiltration (136).

Periodontal ligament stem cells (PDLSCs) are multipotent stem cells obtained from the dental mesenchyme, or ectomesenchyme, and obtained from the PDL tissue. Seo BM *et al.* isolated PDLSCs from PDL tissue of human third molars, surgically extracted. They demonstrated the differentiation of PDLSCs into cementoblast-like cells, adipocytes and collagen-forming cells in vitro. In addition, PDLSCs has the ability to form a cementum/PDL-like structure for periodontal tissue repair when were transplanted into immunocompromised rodents (137). Liu Y

et al. transplanted autologous PDLCs obtained from miniature pigs into previously created periodontal defects, and presented PDLCs were able to regenerate the periodontal tissue (138). Dangaria *et al.* (139) showed that PDL progenitor cells that loaded on tooth root thoroughly formed PDL in rats' alveolar bone. In addition, transplantation of PDLSCs can be a regenerative approach for treatment of periodontal disease due to its anti-inflammatory effect (140).

The dental follicle is an ectomesenchymal tissue surrounding the teeth, containing multipotent stem cells or precursor cells (PCs), capable of differentiating into osteoblasts, PDL cells and cementoblasts, PDL cells and osteoblasts. Morsczeck *et al.* isolated PCs from the dental follicle of extracted human third molars teeth. PCs have the ability of differentiation, formation of a mesothelium-like cellular structure, and encapsulating a connective tissue-like matrix in vitro. When PCs with hydroxyapatite powder were transplanted into immunocompromised mice, they exhibit no signs of bone or cementum formation. Consequently, PCs are undifferentiated and unique cells that exist in the periodontium during or prior to tooth eruption (128). Moreover, in vitro analysis revealed that even after 30 passages, PCs can form a cementum and PDL-like tissue (141).

Stem cells from apical papilla (SCAP) are multipotent stem cells, derived from the dental mesenchyme, or ectomesenchyme. Sonoyama W *et al.* transplanted ex vivo expanded-SCAP with a hydroxyapatite/tricalcium phosphate (HP/TCP) scaffold into immunocompromised mice (129). Results indicated regeneration of a typical dentin structure, with a layer of dentin tissue and connective tissue forming on the surface of the HA/TCP; in fact, SCAP has the potential to differentiate into functional dentinogenic cells. The in vitro results of odontogenic differentiation indicate that SCAP not only are a unique population of postnatal stem cells but also distinct from DPSCs. In addition, to accomplished functional root regeneration, they implanted a root-shaped HA/TPC/gelfoam scaffold containing SCAP and PDLCs into a mini-pig socket. Due to mimicking a bio-physiological root/periodontal environment, they used human SCAP with PDLSCs to form dentin and PDL in vivo. Eight weeks post-transplantation, cementum was formed on the surface of HAe/TCP scaffold after 8 weeks, and Sharpey's fibers anchored into the cementum were observed. According to this data, utilizing a combination of mesenchymal stem cell like SCAP/PDLSCs allows for regeneration of root/periodontal tissue (129). Moreover, Sonoyama W *et al.* studied the properties of SCAP by histologic immunohistochemical and immunocytofluorescent analyses. They demonstrated that the potential of SCAP for osteo/dentinogenic differentiation similar to both DPSCs and bone marrow-derived MSCs (130). Moreover, local injection of SCAPs effectively reconstructed periodontal defects following induced periodontitis in a minipig (142).

Park JC *et al.* demonstrated successfully isolation of inflamed PDLSCs (iPDLSCs) from inflamed PDL tissue and intra-bony defects, and compared it with healthy PDLSCs in periodontal regeneration (143). Proliferative and migratory capacities were evaluated and compared using histologic immunohistochemical, colony-forming unit assay and immunocytofluorescent analyses. iPDLSCs demonstrated differentiation similar to healthy PDLSCs into either osteogenic, cementogenic or adipogenic microenvironments. In addition, both groups presented new cementum-like tissue, PDL formation and the same proliferative potential. This is in contrast with the migratory potential, which showed a significant increase in iPDLSCs.

Liao J *et al.* used immunohistochemistry in collected periapical inflammatory tissues to detect mesenchymal stem cell (MSC) marker expression. In vitro, although the cells demonstrated a strong osteogenic potential, showed weak adipogenic capacity. Transplantation of the cells in vivo , possibility of mineralized tissues formation in immunocompromised mice was observed. The researchers concluded that human periapical inflammatory tissues proposed the presence of MSCs due to expression of MSC markers. The isolated cells not only have the capacity for mineralized matrix formation in vitro and in vivo, but also displayed typical mesenchymal cell immunophenotype (144).

Alongi *et al.* demonstrated the isolation of DPSCs from inflamed pulps (IPs) and their ability for tissue regeneration in vivo. They isolated DPSCs from normal pulp (NPs) tissue and IPs (145) tissue using histologic immunohistochemical, immunocytofluorescent and population doubling analyses to compare the potential of tissue regeneration. The immunohistochemical analysis demonstrated higher levels of mesenchymal stem cell marker expression in DPSCs-IPs. Total population doubling of DPSCs-IPs was lower compared to DPSCs, and DPSCs-IPs appeared to have a decreased osteo/dentinogenic potential compared with DPSCs-NPs based on the mineral deposition in cultures. However, DPSCs-IPs formed pulp/dentin complexes similar to DPSCs-NPs when transplanted into immunocompromised

mice. The researchers concluded that DPSCs-IPs can be isolated, and their mesenchymal stem cell marker profiles are similar to those from NPs. Although some stem cell properties of DPSCs-IPs were altered, cells from some samples remained potent in tissue regeneration in vivo (146).

On the other hand, Tang HN *et al.* (147) discovered that stem cells derived from inflamed PDL (i-PDLSCs) are dysfunctional and compromised, due to impaired immunomodulation, in comparison with stem cells derived from healthy PDL (h-PDLSCs). They concluded that despite the enhanced extracellular matrix proteins of i-PDLSCs, they are impaired in tissue regeneration compared to h-PDLSCs.

Cell Sheet Engineering Approaches for Periodontal Regeneration

Cell sheet engineering is the process by which a confluent culture of a cell or progenitor cell of interest is produced and harvested in vitro for implantation in vivo (148). In this approach, a cell is cultured upon a substrate that is assimilated to reflect the ECM of the target tissue.

The substratum upon which these cells are cultured reflect the ECM of the target tissue to be repaired; thus, it is important to detach the sheet from culture while maintaining the integrity of the substrate. For this, studies continue to investigate optimal methods for detachment from growth plate (60). This differs from conventional cell culturing, in that a detachment would otherwise be facilitated by proteolytic enzymatic treatment. This will relieve the cells of the substratum, making it unideal for impregnation unto a target (60). Enzymatic treatment also damages cell membranes due to hydrolyzation of membrane-bound proteins, inhibiting the regenerative potential of the cell sheet.

For this reason, cell sheet engineering with the aim of tissue regeneration employs a thermo-responsive polymer [e.g. poly (N-isopropylacrylamide) (NIPAAm)] for harvesting of cell sheets. The surface NIPAAm becomes hydrophobic at 37 °C, facilitating the attachment, spread and proliferation of cells. At 32 °C, the temperature responsive properties of NIPAAm make its superficial surface hydrated and hydrophilic, causing the cells to detach and making it possible to harvest the cells from the culture surfaces without the use of enzymes. The cell sheet can then be implanted in the periodontal defect to promote regeneration continuously (149).

The regenerative capacity of human PDL cell sheets alone were tested on the root surface of periodontal defects in athymic rats (150). The results demonstrated cementogenesis and attachment of newly formed collagen fibers to the newly regenerated cementum layer. This was compared to a control that showed no clear cementum layer. Consequently, according to this study, cell sheet engineering alone has tissue regenerative capacity.

The use of autologous tissues for cell sheet culture has the potential to overcome immune reaction to the cell sheet graft. Furthermore, regarding periodontal defects, PDL cells show plasticity in their regeneration, forming both hard and soft tissues (151). This was demonstrated in transplantation of an autologous, three-layered PDL cell sheet to dogs on the dental root surface of a three-wall periodontal defect, with the bony defect filled with porous beta-tricalcium phosphate (60). Bone and cementum were regenerated with well oriented collagen fibers.

Cell sheet technology has been effectively used in several studies for periodontal regeneration in preclinical trials (60, 150, 152, 153). Moreover, other researchers have conducted both in vitro and in vivo studies combining the cell sheet technology with porous scaffolds, which successfully accomplished periodontal regeneration (46, 154).

Combinatorial Techniques for Periodontal Regeneration

Prospective studies in periodontal regeneration via tissue engineering integrates biomaterial scaffolds, growth factors and progenitor cells to simultaneously regenerate a variety of oral and dental tissues (26). This approach aims for a less invasive, yet effective, personalized and integrated regeneration of the targeted periodontal defects (26). The goal of employing combinatorial scaffold fabrication techniques is to generate a scaffold that compensates for differential growth rates of different histological layers, induces growth factor delivery at predictable rates and is metabolized at a rate appropriate for the layers regenerative capacity, and also maintaining an optimal environment to facilitate the growth and orientation of specific cell types (155). Such scaffolds describe multiphasic constructs—templates for tissue regeneration that guide the growth of new tissues in a manner that considers different cell types and their requirements for growth and maturation to restore form and function of histologically variable tissue, such as the periodontium (156).

An ideal design of a multiphasic scaffold is fabricated with

respect to morphology and degradation kinetics and physicochemical properties, that improve cell growth and function (129). Tomography, internal microstructure and interconnectivity of the pores and channels of the scaffold are just some of the properties that affect cellular behavior upon the scaffold, including migration, adhesion, proliferation and differentiation (155). Because of this, combinatorial techniques explore various scaffold fabrication techniques to form an optimal, multiphasic scaffold.

Structural properties, like pore size and geometry, may be refined with the utility of multiple scaffold fabrication techniques (26). These fabrication techniques include porogen leaching, gas foaming, freeze-drying, electrospinning and 3D-printing (26, 157, 158). Any combination of these techniques may be employed to gather the specifications of the scaffold of interest. Establishing a pore network ideal for cellular migration, attachment, proliferation and differentiation, as well as circulation of nutrients and metabolic wastes, provides a more optimized environment for cell culturing and intracellular communication for integration of the multiple phases (155). The scaffold is also ideally biodegradable at a rate in tandem with the complementary cell/tissue growth and maturation to provide supplementary physical support while being slowly replaced by the living tissue (159).

It has been demonstrated that impregnating scaffolds with cells derived from cell sheet engineering produces improved results in vivo (160). Mimicking periodontal structure in the design of a scaffold effectively promoted ligamentogenesis in the study by W. Jiant *et al.*, demonstrating that biomimetics is an ideal strategy in the fabrication of a scaffold. It also provides a greater capacity for topographic cues and regenerative orientation of periodontal tissues in vivo (160). Moreover, an in vitro study investigated the regenerative capacity of an osteoconductive biphasic scaffold. The scaffold design by Costa *et al.*, (123) which was enhanced by calcium phosphate combined with cell sheet method, was offered for simultaneous regeneration of alveolar bone/PDL complex.

Demineralized freeze-dried bone allografts (DFDBA) in conjunction with platelet-rich fibrin (PRF), platelet-rich plasma (PRP), amnion membrane (AM), and enamel matrix derivative (EMD) have been studied with the aim of periodontal regeneration (161-163). In a recent systematic review and meta-analysis, the evaluated studies showed that only PRF has a positive impact on recession reduction. Only PRP depicted a statistically significant enhancement in bone formation. AM just gained more CAL, while EMD did not ameliorate any clinical outcome (164).

Adaptability to the defect is a major barrier to overcome in the fabrication of the scaffold. Scaffolds that are rigid must be personalized to the defect targeted. A fabrication technique produced as a non-specific, calcium phosphate block with an electrospun mesh is not easily adapted to a defect space. Alternative fabrication techniques that either produce a scaffold specific to the defect in which it is to be grafted or utilize a medium with less rigidity are some ways to overcome this barrier (52, 155). For instance, in an animal model, the tissue specific tri-layered nanocomposite hydrogel scaffold approach demonstrated complete defect closure and regeneration of the periodontium (52). Each layer was made up of a chitin hydrogel impregnated with varying degrees of poly (lactic-co-glycolic acid) (PLGA) due to limiting the degradation rate, that had a similar composition to extracellular matrix of many tissues; thus, it is easily processed in vivo. The chitin-PLGA polymeric blend lacks enough biomineralization capacity on its own; thus, an array of dental ceramics and nano-bioactive glass ceramic (nBGC) were utilized to enhance regeneration of hard tissue (52). In addition, PRP, recombinant human cementum protein 1 (rhCEMP1) and recombinant human fibroblast growth factor 2 (rhFGF2) were added to the alveolar, cementum and PDL layers, respectively, to facilitate the migration and differentiation potential of dental follicle stem cells (52).

The restoration of function requires a focus on the restoration of physiological loading and homeostasis of the defect area, especially considering the PDL and tooth root interface (4, 160). This necessitates functional cementum formation with restoration of a properly aligned periodontal ligament. This need to fulfill functional orientation demonstrates the importance of a scaffold design that facilitates the alignment of the regenerating PDL phase (165, 166).

Scaffolds fabricated with 3D-printing have the advantage of precise control over the scaffold's microarchitecture in various regions, which is an ideal method for constructing a scaffold of multiple phases. 3D-printing fabrications utilizes a layer-by-layer method and can provide simultaneous impregnation of growth-factor delivery onto the scaffold. In a study by Lee C.H. *et al.*, a scaffold was 3D-printed using hydroxyapatite (HA) impregnated with PLGA microspheres containing growth factors. PLGA impregnation was designed in such a way for spatiotemporal release specific to each phase. The microchannels of the scaffold were made up of polycaprolactone fibers (CL) and were integrated

perpendicular to the HA scaffold (PCL/HA), with three phases specified for alveolar bone, PDL and dentin/cementum. Dental progenitor stem cells (DPSC's) were cultured on all three phases of the scaffold. The three phases were labeled A, B and C, corresponding to the three tissue layers of dentin/cementum, PDL and alveolar bone, respectively. A pilot study was performed to assert optimal topographical and porous design for the various phases to facilitate specific tissue layer regeneration. Cell differentiation was influenced by encapsulating growth factors into the different phases of the scaffold: (A) received amelogenin, while (B) received connective tissue growth factor (CTGF). In vivo studies on immunocompromised mice revealed that scaffolds with spatiotemporal delivery of growth factors demonstrated multiphasic tissue layering consisting of a PDL-like collagen fibrous layer in phase (B) that interfaced phase (A) with phase (C). Furthermore, it is suggested that de novo formation of dentin-/cementum-like structures occurred with three-dimensional architecture (5). While the results yielded positive results, clinical application is questionable, as autologous embryonic tooth germ cells are not readily available nor are the aesthetic results fully understood yet (5).

Concluding Remarks and Future Perspective

Combinatorial techniques for the fabrication of multiphasic scaffolds have the capacity to prepare less invasive regeneration of tissue defects with more complete and aesthetic results.

Methods have successfully induced differentiation of stem cells in multiphasic scaffolds with relative ease; treatment with growth factors results in cells differentiated for the purpose of function. When placed in biological system, these cells thrive, interacting within each layer to form interfaces that may be indicative of restored function. This has not been asserted as of yet further investigation of the regenerated defect is necessary. Furthermore, many studies allude to unanticipated results, and greater emphasis on how biological systems respond to different elements of these scaffold designs could provide an understanding of an optimal design. Fabrication design predominantly concentrates on the facilitation of cell growth and migration.

Future approaches should continue to dominate the conventional construct limitations, most notably, stiffness, handling properties and adaptability. Of the techniques discussed, a design that facilitates collagen orientation requires investigation.

Periodontal defects present themselves clinically in a case by case basis; thus, fabrication techniques must compensate for personalization of the defect in question. Ideal fabrication techniques include those that are easily manipulatable and/or plastic. Because multiple phases attempt to mimic histological layering, orientation of the scaffold may lend itself to adaptability issues in the clinical setting. This issue demonstrates the needs for finding a method in order to personalize each scaffold per patient defect area parameters. 3D-printing fabrication is a candidate to fulfill this requirement.

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References

1. Dye BA, Li X, Thornton-Evans G. Oral health disparities as determined by selected healthy people 2020 oral health objectives for the United States, 2009-2010: US Department of Health and Human Services, Centers for Disease Control and …; 2012.
2. Eke PI, Borgnakke WS, Genco RJJP. Recent epidemiologic trends in periodontitis in the USA. 2020;82(1):257-67.
3. Kinane DF, Stathopoulou PG, Papapanou PNJNRDP. Periodontal diseases. 2017;3(1):1-14.
4. Bosshardt DD, Stadlinger B, Terheyden H. Cell-to-cell communication–periodontal regeneration. Clinical oral implants research. 2015;26(3):229-39.
5. Lee CH, Hajibandeh J, Suzuki T, Fan A, Shang P, Mao JJ. Three-dimensional printed multiphase scaffolds for regeneration of periodontium complex. Tissue Engineering Part A. 2014;20(7-8):1342-51.
6. Larsson L, Decker A, Nibali L, Pilipchuk S, Berglundh T, Giannobile WJJodr. Regenerative medicine for periodontal and peri-implant diseases. 2016;95(3):255-66.
7. Nanci A, Bosshardt DD. Structure of periodontal tissues in health and disease. Periodontology 2000. 2006;40(1):11-28.
8. Bhatia A, Sharma RK, Tewari S, Narula SC, Khurana HJJoISoP. Periodontal status in chronic periodontitis depressed patients on desvenlafaxine: An observational study. 2018;22(5):442.
9. Susin C, Fiorini T, Lee J, De Stefano JA, Dickinson DP, Wikesjö UMJP. Wound healing following surgical and regenerative periodontal therapy. 2015;68(1):83-98.
10. Ho SP, Kurylo MP, Fong TK, Lee SS, Wagner HD, Ryder MI, et al. The biomechanical characteristics of the bone-periodontal ligament-cementum complex. 2010;31(25):6635-46.
11. Jiang W, Li L, Zhang D, Huang S, Jing Z, Wu Y, et al. Incorporation of aligned PCL–PEG nanofibers into porous chitosan scaffolds improved the orientation of collagen fibers in regenerated periodontium. Acta biomaterialia. 2015;25:240-52.
12. Han J, Menicanin D, Gronthos S, Bartold P. Stem cells, tissue

engineering and periodontal regeneration. Australian dental journal. 2014;59:117-30.
13. Ishikawa I, Iwata T, Washio K, Okano T, Nagasawa T, Iwasaki K, et al. Cell sheet engineering and other novel cell-based approaches to periodontal regeneration. Periodontology 2000. 2009;51(1):220-38.
14. Mojahediana M, Fahimipourb F, Larsena KL, Kalantarc M, Bastamid F, Omatalie N. Ethanol-based sol-gel synthesis of nano-crystalline hydroxyapatite with different calcium phosphorus ratios (Ca/P). Journal of Ceramic Processing Research. 2016;17(11):1138-42.
15. Ramachandra SS, Rana R, Reetika S, Jithendra K. Options to avoid the second surgical site: a review of literature. Cell and tissue banking. 2014;15(3):297-305.
16. Bartold P, Gronthos S, Ivanovski S, Fisher A, Hutmacher D. Tissue engineered periodontal products. Journal of periodontal research. 2016;51(1):1-15.
17. Wikesjö UM, Selvig KAJP. Periodontal wound healing and regeneration. 1999;19(1):21-39.
18. Narvekar A, Luan KW, Gholami F. Decision Trees in Periodontal Surgery: Resective Versus Regenerative Periodontal Surgery. Advances in Periodontal Surgery: Springer; 2020. p. 23-41.
19. Wikesj UM, Nilvéus RJJop. Periodontal repair in dogs: effect of wound stabilization on healing. 1990;61(12):719-24.
20. Wikesjö UM, Claffey N, Egelberg JJJocp. Periodontal repair in dogs Effect of heparin treatment of the root surface. 1991;18(1):60-4.
21. Shewale AH, Gattani D, Mahajan R, Saravanan S, Bhatia NJJoAiM, Research M. Root Surface Biomodification: Current Status and a Literature Review on Available Agents for Periodontal Regeneration. 2016:1-14.
22. Karam P, Sant'Ana A, de Rezende M, Greghi S, Damante C, Zangrando MJJopr. Root surface modifiers and subepithelial connective tissue graft for treatment of gingival recessions: a systematic review. 2016;51(2):175-85.
23. Bastami F, Paknejad Z, Jafari M, Salehi M, Rad MR, Khojasteh A. Fabrication of a three-dimensional β-tricalcium-phosphate/gelatin containing chitosan-based nanoparticles for sustained release of bone morphogenetic protein-2: Implication for bone tissue engineering. Materials Science and Engineering: C. 2017;72:481-91.
24. Hills DJP. Evaluation of Demineralized Freeze-Dried Bone Allograft in Combination with Chorion Membrane in the Treatment of Grade II Furcation Defects: A Randomized Controlled Trial. 2019;39:659-67.
25. Khojasteh A, Kheiri L, Behnia H, Tehranchi A, Nazeman P, Nadjmi N, et al. Lateral ramus cortical bone plate in alveolar cleft osteoplasty with concomitant use of buccal fat pad derived cells and autogenous bone: phase I clinical trial. 2017;2017.
26. Ivanovski S, Vaquette C, Gronthos S, Hutmacher D, Bartold P. Multiphasic scaffolds for periodontal tissue engineering. Journal of dental research. 2014;93(12):1212-21.
27. Langer R, Vacanti JJTETUOB, ENGINEERING. Tissue engineering. Science 260: 920-926. 1993;98.
28. Asa'ad F, Monje A, Larsson L. Scaffolds for periodontal tissue engineering. Handbook of Tissue Engineering Scaffolds: Volume One: Elsevier; 2019. p. 479-504.
29. Babo PS, Reis RL, Gomes ME. Periodontal tissue engineering: current strategies and the role of platelet rich hemoderivatives. Journal of Materials Chemistry B. 2017;5(20):3617-28.
30. Khojasteh A, Fahimipour F, Eslaminejad MB, Jafarian M, Jahangir S, Bastami F, et al. Development of PLGA-coated β-TCP scaffolds containing VEGF for bone tissue engineering. Materials Science and Engineering: C. 2016;69:780-8.
31. Ouchi T, Nakagawa TJRT. Mesenchymal stem cell-based tissue regeneration therapies for periodontitis. 2020;14:72-8.
32. Iwata T, Washio K, Yoshida T, Ishikawa I, Ando T, Yamato M, et al. Cell sheet engineering and its application for periodontal regeneration. Journal of tissue engineering and regenerative medicine. 2015;9(4):343-56.
33. Hosseinpour S, Ahsaie MG, Rad MR, taghi Baghani M, Motamedian SR, Khojasteh A. Application of selected scaffolds for bone tissue engineering: a systematic review. Oral and maxillofacial surgery. 2017;21(2):109-29.
34. Shahin-Shamsabadi A, Hashemi A, Tahriri M, Bastami F, Salehi M, Abbas FM. Mechanical, material, and biological study of a PCL/bioactive glass bone scaffold: Importance of viscoelasticity. Materials Science and Engineering: C. 2018.
35. Eslami H, Lisar HA, Kashi TSJ, Tahriri M, Ansari M, Rafiei T, et al. Poly (lactic-co-glycolic acid)(PLGA)/TiO2 nanotube bioactive composite as a novel scaffold for bone tissue engineering: In vitro and in vivo studies. Biologicals. 2018.
36. De Bruyckere T, Cosyn J, Younes F, Hellyn J, Bekx J, Cleymaet R, et al. A randomized controlled study comparing guided bone regeneration with connective tissue graft to re-establish buccal convexity: one-year aesthetic and patient-reported outcomes. 2020.
37. Waller T, Herzog M, Thoma DS, Hüsler J, Hämmerle CH, Jung REJCOIR. Long-term clinical and radiographic results after treatment or no treatment of small buccal bone dehiscences at posterior dental implants-A randomized, controlled clinical trial. 2020.
38. Temmerman A, Cortellini S, Van Dessel J, De Greef A, Jacobs R, Dhondt R, et al. Bovine-derived xenograft in combination with autogenous bone chips versus xenograft alone for the augmentation of bony dehiscences around oral implants: A randomized, controlled, split-mouth clinical trial. 2020;47(1):110-9.
39. Preeja C, Arun S. Platelet-rich fibrin: Its role in periodontal regeneration. The Saudi Journal for Dental Research. 2014;5(2):117-22.
40. Verma UP, Yadav RK, Dixit M, Gupta A. Platelet-rich fibrin: A paradigm in periodontal therapy–A systematic review. Journal of International Society of Preventive & Community Dentistry. 2017;7(5):227.
41. Bastami F, Khojasteh A. Use of Leukocyte-and Platelet-Rich Fibrin

for Bone Regeneration: A Systematic Review. Regeneration, Reconstruction & Restoration. 2016;1(2):47-68.
42. Rezaei M, Jamshidi S, Saffarpour A, Ashouri M, Rahbarghazi R, Rokn AR, et al. Transplantation of Bone Marrow" Derived Mesenchymal Stem Cells, Platelet-Rich Plasma, and Fibrin Glue for Periodontal Regeneration. 2019;39(1).
43. Arbildo H, Gamarra L, Rojas S, Infantes E, Lamas C, Vásquez HJJoOR. Clinical effect of platelet rich fibrin in the treatment of periodontal intrabony defects. Systematic review and meta-analysis. 2019;6(5):127-35.
44. Caruana A, Savina D, Macedo JP, Soares SCJEjod. From Platelet-Rich Plasma to Advanced Platelet-Rich Fibrin: Biological Achievements and Clinical Advances in Modern Surgery. 2019.
45. Navarro LB, Barchiki F, Junior WN, Carneiro E, da Silva Neto UX, Westphalen VPDJSR. Assessment of platelet-rich fibrin in the maintenance and recovery of cell viability of the periodontal ligament. 2019;9(1):1-9.
46. Bastami F, Nazeman P, Moslemi H, Rezai Rad M, Sharifi K, Khojasteh A. Induced pluripotent stem cells as a new getaway for bone tissue engineering: a systematic review. Cell proliferation. 2016.
47. Cho YD, Kim KH, Ryoo HM, Lee YM, Ku Y, Seol YJ. Recent advances of useful cell sources in the periodontal regeneration. Current stem cell research & therapy. 2018.
48. Bastami F, Nazeman P, Moslemi H, Rezai Rad M, Sharifi K, Khojasteh AJCP. Induced pluripotent stem cells as a new getaway for bone tissue engineering: a systematic review. 2017;50(2):e12321.
49. Park D, Spencer JA, Koh BI, Kobayashi T, Fujisaki J, Clemens TL, et al. Endogenous bone marrow MSCs are dynamic, fate-restricted participants in bone maintenance and regeneration. Cell stem cell. 2012;10(3):259-72.
50. Fahimipour F, Rasoulianboroujeni M, Dashtimoghadam E, Khoshroo K, Tahriri M, Bastami F, et al. 3D printed TCP-based scaffold incorporating VEGF-loaded PLGA microspheres for craniofacial tissue engineering. Dental Materials. 2017.
51. Eslami H, Lisar HA, Kashi TSJ, Tahriri M, Ansari M, Rafiei T, et al. Poly (lactic-co-glycolic acid)(PLGA)/TiO2 nanotube bioactive composite as a novel scaffold for bone tissue engineering: In vitro and in vivo studies. Biologicals. 2018;53:51-62.
52. Fahimipour F, Rasoulianboroujeni M, Dashtimoghadam E, Khoshroo K, Tahriri M, Bastami F, et al. 3D printed TCP-based scaffold incorporating VEGF-loaded PLGA microspheres for craniofacial tissue engineering. Dent Mater. 2017;33:1205-16.
53. Salehi M, Ehtrami A, Bastami F, Farzamfar S, Hosseinpour S, Vahedi H, et al. Polyurethane/Gelatin Nanofiber Neural Guidance Conduit in Combination with Resveratrol and Schwann Cells for Sciatic Nerve Regeneration in the Rat Model. 2019;20(3):490-500.
54. Tavelli L, McGuire MK, Zucchelli G, Rasperini G, Feinberg SE, Wang HL, et al. Extracellular matrix-based scaffolding technologies for periodontal and peri-implant soft tissue regeneration. 2020;91(1):17-25.
55. Millan C, Vivanco JF, Benjumeda-Wijnhoven IM, Bjelica S, Santibanez JF. Mesenchymal Stem Cells and Calcium Phosphate Bioceramics: Implications in Periodontal Bone Regeneration. Advances in experimental medicine and biology. 2018.
56. Sowmya S, Mony U, Jayachandran P, Reshma S, Kumar RA, Arzate H, et al. Tri-Layered Nanocomposite Hydrogel Scaffold for the Concurrent Regeneration of Cementum, Periodontal Ligament, and Alveolar Bone. Advanced healthcare materials. 2017;6(7):1601251.
57. Nabavi MH, Salehi M, Ehterami A, Bastami F, Semyari H, Tehranchi M, et al. A collagen-based hydrogel containing tacrolimus for bone tissue engineering. Drug Deliv Transl Res. 2019;10(1):108-21.
58. Donos N, Dereka X, Calciolari EJJocp. The use of bioactive factors to enhance bone regeneration: A narrative review. 2019;46:124-61.
59. Wikesjö UM, Xiropaidis AV, Thomson RC, Cook AD, Selvig KA, Hardwick WRJJocp. Periodontal repair in dogs: rhBMP-2 significantly enhances bone formation under provisions for guided tissue regeneration. 2003;30(8):705-14.
60. Wikesjö UM, Xiropaidis AV, Thomson RC, Cook AD, Selvig KA, Hardwick WRJJocp. Periodontal repair in dogs: space-providing ePTFE devices increase rhBMP-2/ACS-induced bone formation. 2003;30(8):715-25.
61. Mitchell AC, Briquez PS, Hubbell JA, Cochran JR. Engineering growth factors for regenerative medicine applications. Acta biomaterialia. 2016;30:1-12.
62. Tatakis DN, Wikesjö UM, Razi SS, Sigurdsson TJ, Lee MB, Nguyen T, et al. Periodontal repair in dogs: effect of transforming growth factor-β1 on alveolar bone and cementum regeneration. 2000;27(9):698-704.
63. Wikesjö UM, Razi SS, Sigurdsson TJ, Tatakis DN, Lee MB, Ongpipattanakul B, et al. Periodontal repair in dogs: effect of recombinant human transfornning growth factor-β1 on guided tissue regeneration. 1998;25(6):475-81.
64. Li F, Yu F, Xu X, Li C, Huang D, Zhou X, et al. Evaluation of recombinant human FGF-2 and PDGF-BB in periodontal regeneration: a systematic review and meta-analysis. Scientific reports. 2017;7(1):65.
65. Giannobile WV, Hernandez RA, Finkelman RD, Ryarr S, Kiritsy CP, D'Andrea M, et al. Comparative effects of plateletderived growth factor-BB and insulin-like growth factor-I, individually and in combination, on periodontal regeneration in Macaca fascicularis. 1996;31(5):301-12.
66. Howell TH, Fiorellini JP, Paquette DW, Offenbacher S, Giannobile WV, Lynch SEJJop. A phase I/II clinical trial to evaluate a combination of recombinant human platelet-derived growth factor-BB and recombinant human insulin-like growth factor-I in patients with periodontal disease. 1997;68(12):1186-93.
67. Ridgway HK, Mellonig JT, Cochran DLJIJoP, Dentistry R. Human histologic and clinical evaluation of recombinant human platelet-derived growth factor and beta-tricalcium phosphate for the

treatment of periodontal intraosseous defects. 2008;28(2).
68. Nevins M, Giannobile WV, McGuire MK, Kao RT, Mellonig JT, Hinrichs JE, et al. Platelet-derived growth factor stimulates bone fill and rate of attachment level gain: Results of a large multicenter randomized controlled trial. 2005;76(12):2205-15.
69. Sato Y, Kikuchi M, Ohata N, Tamura M, Kuboki YJJop. Enhanced cementum formation in experimentally induced cementum defects of the root surface with the application of recombinant basic fibroblast growth factor in collagen gel in vivo. 2004;75(2):243-8.
70. Rossa Jr C, Marcantonio Jr E, Cirelli JA, Marcantonio RA, Spolidorio LC, Fogo JCJJop. Regeneration of Class III furcation defects with basic fibroblast growth factor (b-FGF) associated with GTR. A descriptive and histometric study in dogs. 2000;71(5):775-84.
71.Choi S-H, Kim C-K, Cho K-S, Huh J-S, Sorensen RG, Wozney JM, et al. Effect of recombinant human bone morphogenetic protein-2/absorbable collagen sponge (rhBMP-2/ACS) on healing in 3-wall intrabony defects in dogs. Journal of periodontology. 2002;73(1):63-72.
72.Saito E, Saito A, Kawanami MJJop. Favorable Healing Following Space Creation in rhBMP-2–Induced Periodontal Regeneration of Horizontal Circumferential Defects in Dogs with Experimental Periodontitis. 2003;74(12):1808-15.
73.Sigurdsson TJ, Lee MB, Kubota K, Turek TJ, Wozney JM, Wikesjö UMJJop. Periodontal repair in dogs: recombinant human bone morphogenetic protein-2 significantly enhances periodontal regeneration. 1995;66(2):131-8.
74.Carmona-Rodríguez B, Alvarez-Pérez MA, Narayanan AS, Zeichner-David M, Reyes-Gasga J, Molina-Guarneros J, et al. Human Cementum Protein 1 induces expression of bone and cementum proteins by human gingival fibroblasts. Biochemical and biophysical research communications. 2007;358(3):763-9.
75.Bastami F, Vares P, Khojasteh A. Healing Effects of Platelet-Rich Plasma on Peripheral Nerve Injuries. J Craniofac Surg 2017;28(1):e49-e57.
76.Plachokova AS, Nikolidakis D, Mulder J, Jansen JA, Creugers NH. Effect of platelet-rich plasma on bone regeneration in dentistry: a systematic review. Clinical oral implants research. 2008;19(6):539-45.
77.Caplan AI. Mesenchymal stem cells. Journal of orthopaedic research. 1991;9(5):641-50.
78.Semyari H, Rajipour M, Bastami F, Semyari H. Isolation and Culture of Mesenchymal Stem Cells From Rabbit Scapular Subcutaneous Adipose Tissue and Their Ability to Differentiate Into Osteoblasts. Avicenna Journal of Dental Research. 2015;7(2).
79.Huang G-J, Gronthos S, Shi S. Mesenchymal stem cells derived from dental tissues vs. those from other sources: their biology and role in regenerative medicine. Journal of dental research. 2009;88(9):792-806.
80.Gronthos S, Mankani M, Brahim J, Robey PG, Shi S. Postnatal human dental pulp stem cells (DPSCs) in vitro and in vivo. Proceedings of the National Academy of Sciences. 2000;97(25):13625-30.
81.Miura M, Gronthos S, Zhao M, Lu B, Fisher LW, Robey PG, et al. SHED: stem cells from human exfoliated deciduous teeth. Proceedings of the National Academy of Sciences. 2003;100(10):5807-12.
82.Morsczeck C, Götz W, Schierholz J, Zeilhofer F, Kühn U, Möhl C, et al. Isolation of precursor cells (PCs) from human dental follicle of wisdom teeth. Matrix Biology. 2005;24(2):155-65.
83.Sonoyama W, Liu Y, Yamaza T, Tuan RS, Wang S, Shi S, et al. Characterization of the apical papilla and its residing stem cells from human immature permanent teeth: a pilot study. Journal of endodontics. 2008;34(2):166-71.
84.Sonoyama W, Liu Y, Fang D, Yamaza T, Seo B-M, Zhang C, et al. Mesenchymal stem cell-mediated functional tooth regeneration in swine. PloS one. 2006;1(1):e79.
85.Gronthos S, Brahim J, Li W, Fisher L, Cherman N, Boyde A, et al. Stem cell properties of human dental pulp stem cells. Journal of dental research. 2002;81(8):531-5.
86.Hernandez-Monjaraz B, Santiago-Osorio E, Ledesma-Martinez E, Alcauter-Zavala A, Mendoza-Nunez VM. Retrieval of a periodontally compromised tooth by allogeneic grafting of mesenchymal stem cells from dental pulp: A case report. The Journal of international medical research. 2018;46(7):2983-93.
87.Aimetti M, Ferrarotti F, Gamba MN, Giraudi M, Romano F. Regenerative Treatment of Periodontal Intrabony Defects Using Autologous Dental Pulp Stem Cells: A 1-Year Follow-Up Case Series. The International journal of periodontics & restorative dentistry. 2018;38(1):51-8.
88.Ferrarotti F, Romano F, Gamba MN, Quirico A, Giraudi M, Audagna M, et al. Human intrabony defect regeneration with micrografts containing dental pulp stem cells: A randomized controlled clinical trial. Journal of clinical periodontology. 2018;45(7):841-50.
89.Hu L, Liu Y, Wang S. Stem cell-based tooth and periodontal regeneration. Oral diseases. 2018;24(5):696-705.
90.Gao X, Shen Z, Guan M, Huang Q, Chen L, Qin W, et al. Immunomodulatory Role of Stem Cells from Human Exfoliated Deciduous Teeth on Periodontal Regeneration. Tissue engineering Part A. 2018.
91.Seo B-M, Miura M, Gronthos S, Bartold PM, Batouli S, Brahim J, et al. Investigation of multipotent postnatal stem cells from human periodontal ligament. The Lancet. 2004;364(9429):149-55.
92.Liu Y, Zheng Y, Ding G, Fang D, Zhang C, Bartold PM, et al. Periodontal ligament stem cell-mediated treatment for periodontitis in miniature swine. Stem cells. 2008;26(4):1065-73.
93. Dangaria SJ, Ito Y, Luan X, Diekwisch TG. Successful periodontal ligament regeneration by periodontal progenitor preseeding on natural tooth root surfaces. Stem cells and development. 2011;20(10):1659-68.
94. Nagata M, Iwasaki K, Akazawa K, Komaki M, Yokoyama N, Izumi Y, et al. Conditioned medium from periodontal ligament stem cells enhances periodontal regeneration. Tissue Engineering Part A. 2017;23(9-10):367-77.
95. Zhai Q, Dong Z, Wang W, Li B, Jin Y. Dental stem cell and dental tissue regeneration. Frontiers of medicine. 2018:1-8.
96. Li G, Han N, Zhang X, Yang H, Cao Y, Wang S, et al. Local Injection of Allogeneic Stem Cells from Apical Papilla Enhanced Periodontal Tissue Regeneration in Minipig Model of

Periodontitis. BioMed research international. 2018;2018.
97. Park JC, Kim JM, Jung IH, Kim JC, Choi SH, Cho KS, et al. Isolation and characterization of human periodontal ligament (PDL) stem cells (PDLSCs) from the inflamed PDL tissue: in vitro and in vivo evaluations. Journal of clinical periodontology. 2011;38(8):721-31.
98. Liao J, Al Shahrani M, Al-Habib M, Tanaka T, Huang GT-J. Cells isolated from inflamed periapical tissue express mesenchymal stem cell markers and are highly osteogenic. Journal of endodontics. 2011;37(9):1217-24.
99.Bouwens DG, Cevidanes L, Ludlow JB, Phillips C. Comparison of mesiodistal root angulation with posttreatment panoramic radiographs and cone-beam computed tomography. American journal of orthodontics and dentofacial orthopedics : official publication of the American Association of Orthodontists, its constituent societies, and the American Board of Orthodontics. 2011;139(1):126-32.
100.Alongi DJ, Yamaza T, Song Y, Fouad AF, Romberg EE, Shi S, et al. Stem/progenitor cells from inflamed human dental pulp retain tissue regeneration potential. Regenerative medicine. 2010;5(4):617-31.
101.Tang HN, Xia Y, Yu Y, Wu RX, Gao LN, Chen FM. Stem cells derived from "inflamed" and healthy periodontal ligament tissues and their sheet functionalities: a patient-matched comparison. Journal of clinical periodontology. 2016;43(1):72-84.
102.Raju R, Oshima M, Inoue M, Morita T, Huijiao Y, Waskitho A, et al. Three-dimensional periodontal tissue regeneration using a bone-ligament complex cell sheet. 2020;10(1):1-16.
103.Carter S-SD, Costa PF, Vaquette C, Ivanovski S, Hutmacher DW, Malda J. Additive biomanufacturing: an advanced approach for periodontal tissue regeneration. Annals of biomedical engineering. 2017;45(1):12-22.
104.Flores MG, Yashiro R, Washio K, Yamato M, Okano T, Ishikawa I. Periodontal ligament cell sheet promotes periodontal regeneration in athymic rats. Journal of clinical periodontology. 2008;35(12):1066-72.
105.Leon E, Iwasaki K, Komaki M, Kojima T, Ishikawa I. Osteogenic effect of interleukin-11 and synergism with ascorbic acid in human periodontal ligament cells. Journal of periodontal research. 2007;42(6):527-35.
106.Washio K, Iwata T, Mizutani M, Ando T, Yamato M, Okano T, et al. Assessment of cell sheets derived from human periodontal ligament cells: a pre-clinical study. Cell and tissue research. 2010;341(3):397-404.
107.Hasegawa M, Yamato M, Kikuchi A, Okano T, Ishikawa I. Human periodontal ligament cell sheets can regenerate periodontal ligament tissue in an athymic rat model. Tissue engineering. 2005;11(3-4):469-78.
108.Dan H, Vaquette C, Fisher AG, Hamlet SM, Xiao Y, Hutmacher DW, et al. The influence of cellular source on periodontal regeneration using calcium phosphate coated polycaprolactone scaffold supported cell sheets. Biomaterials. 2014;35(1):113-22.
109.Cai X, Yang F, Yan X, Yang W, Yu N, Oortgiesen DA, et al. Influence of bone marrow-derived mesenchymal stem cells pre-implantation differentiation approach on periodontal regeneration in vivo. Journal of clinical periodontology. 2015;42(4):380-9.
110.Rasperini G, Pilipchuk S, Flanagan C, Park C, Pagni G, Hollister S, et al. 3D-printed bioresorbable scaffold for periodontal repair. Journal of dental research. 2015;94(9_suppl):153S-7S.
111.Fahmy MD, Jazayeri HE, Razavi M, Masri R, Tayebi L. Three-Dimensional Bioprinting Materials with Potential Application in Preprosthetic Surgery. Journal of Prosthodontics. 2016;25(4):310-8.
112.Mohammadi Z, Mesgar AS-M, Rasouli-Disfani F. Reinforcement of freeze-dried chitosan scaffolds with multiphasic calcium phosphate short fibers. Journal of the mechanical behavior of biomedical materials. 2016;61:590-9.
113.Chen X, Liu Y, Miao L, Wang Y, Ren S, Yang X, et al. Controlled release of recombinant human cementum protein 1 from electrospun multiphasic scaffold for cementum regeneration. International journal of nanomedicine. 2016;11:3145.
114.Dorati R, DeTrizio A, Modena T, Conti B, Benazzo F, Gastaldi G, et al. Biodegradable scaffolds for bone regeneration combined with drug-delivery systems in osteomyelitis therapy. Pharmaceuticals. 2017;10(4):96.
115.Costa PF, Vaquette C, Zhang Q, Reis RL, Ivanovski S, Hutmacher DW. Advanced tissue engineering scaffold design for regeneration of the complex hierarchical periodontal structure. Journal of clinical periodontology. 2014;41(3):283-94.
116.Sali DD, Pauline George J. Demineralized Freeze Dried Bone Allograft With Amniotic Membrane in the Treatment of Periodontal Intrabony Defects-12 Month Randomized Controlled Clinical Trial. Journal of periodontology. 2016(0):1-18.
117.Agarwal A, Gupta ND, Jain A. Platelet rich fibrin combined with decalcified freeze-dried bone allograft for the treatment of human intrabony periodontal defects: a randomized split mouth clinical trail. Acta Odontologica Scandinavica. 2016;74(1):36-43.
118.Kukreja BJ, Dodwad V, Kukreja P, Ahuja S, Mehra P. A comparative evaluation of platelet-rich plasma in combination with demineralized freeze-dried bone allograft and DFDBA alone in the treatment of periodontal intrabony defects: a clinicoradiographic study. Journal of Indian Society of Periodontology. 2014;18(5):618.
119.Zhou S, Sun C, Huang S, Wu X, Zhao Y, Pan C, et al. Efficacy of Adjunctive Bioactive Materials in the Treatment of Periodontal Intrabony Defects: A Systematic Review and Meta-Analysis. BioMed Research International. 2018;2018.
120.Fretwurst T, Larsson L, Yu SH, Pilipchuk SP, Kaigler D, Giannobile WV. Periodontal Tissue Bioengineering: Is the Future Now? Compendium of continuing education in dentistry (Jamesburg, NJ : 1995). 2018;39(4):218-23; quiz 24.
121.Siaili M, Chatzopoulou D, Gillam DG. An overview of periodontal regenerative procedures for the general dental practitioner. The Saudi Dental Journal. 2018;30(1):26-37.
122.Sorensen RG, Wikesjö UM, Kinoshita A, Wozney JM. Periodontal repair in dogs: evaluation of a bioresorbable calcium phosphate cement (CeredexTM) as a carrier for rhBMP-2. Journal of clinical periodontology. 2004;31(9):796-804.
123.Oortgiesen DA, Walboomers XF, Bronckers AL, Meijer GJ, Jansen

JA. Periodontal regeneration using an injectable bone cement combined with BMP-2 or FGF-2. Journal of tissue engineering and regenerative medicine. 2014;8(3):202-9.
124.Blumenthal NM, Koh-Kunst G, Alves ME, Miranda D, Sorensen RG, Wozney JM, et al. Effect of surgical implantation of recombinant human bone morphogenetic protein-2 in a bioabsorbable collagen sponge or calcium phosphate putty carrier in intrabony periodontal defects in the baboon. Journal of periodontology. 2002;73(12):1494-506.
125.Wikesjö UM, Polimeni G, Qahash M. Tissue engineering with recombinant human bone morphogenetic protein-2 for alveolar augmentation and oral implant osseointegration: Experimental observations and clinical perspectives. Clinical implant dentistry and related research. 2005;7(2):112-9.
126.Takahashi Y, Yamamoto M, Yamada K, Kawakami O, Tabata Y. Skull bone regeneration in nonhuman primates by controlled release of bone morphogenetic protein-2 from a biodegradable hydrogel. Tissue engineering. 2007;13(2):293-300.
127.Chen F-M, Zhao Y-M, Zhang R, Jin T, Sun H-H, Wu Z-F, et al. Periodontal regeneration using novel glycidyl methacrylated dextran (Dex-GMA)/gelatin scaffolds containing microspheres loaded with bone morphogenetic proteins. Journal of controlled release. 2007;121(1-2):81-90.
128.Park JK, Yeom J, Oh EJ, Reddy M, Kim JY, Cho DW, et al. Guided bone regeneration by poly(lactic-co-glycolic acid) grafted hyaluronic acid bi-layer films for periodontal barrier applications. Acta Biomater. 2009;5(9):3394-403.
129.Van den Bergh J, Ten Bruggenkate C, Groeneveld H, Burger E, Tuinzing D. Recombinant human bone morphogenetic protein-7 in maxillary sinus floor elevation surgery in 3 patients compared to autogenous bone grafts. Journal of clinical periodontology. 2000;27(9):627-36.
130.Sorensen RG, Polimeni G, Kinoshita A, Wozney JM, Wikesjö UM. Effect of recombinant human bone morphogenetic protein-12 (rhBMP-12) on regeneration of periodontal attachment following tooth replantation in dogs: A pilot study. Journal of clinical periodontology. 2004;31(8):654-61.
131.Nevins M, Camelo M, Nevins ML, Schenk RK, Lynch SE. Periodontal regeneration in humans using recombinant human platelet-derived growth factor-BB (rhPDGF-BB) and allogenic bone. Journal of periodontology. 2003;74(9):1282-92.
132.Nevins M, Kao RT, McGuire MK, McClain PK, Hinrichs JE, McAllister BS, et al. Platelet-derived growth factor promotes periodontal regeneration in localized osseous defects: 36-month extension results from a randomized, controlled, double-masked clinical trial. Journal of periodontology. 2013;84(4):456-64.
133.Mishra A, Avula H, Pathakota KR, Avula J. Efficacy of modified minimally invasive surgical technique in the treatment of human intrabony defects with or without use of rhPDGF-BB gel–a randomized controlled trial. Journal of clinical periodontology. 2013;40(2):172-9.
134.Carney CM, Rossmann JA, Kerns DG, Cipher DJ, Rees TD, Solomon ES, et al. A comparative study of root defect coverage using an acellular dermal matrix with and without a recombinant human platelet-derived growth factor. Journal of periodontology. 2012;83(7):893-901.
135.Chen F-m, Zhao Y-m, Wu H, Deng Z-h, Wang Q-t, Zhou W, et al. Enhancement of periodontal tissue regeneration by locally controlled delivery of insulin-like growth factor-I from dextran–co-gelatin microspheres. Journal of Controlled Release. 2006;114(2):209-22.
136.Giannobile WV, Hernandez RA, Finkelman RD, Ryarr S, Kiritsy CP, D'Andrea M, et al. Comparative effects of plateletderived growth factor-BB and insulin-like growth factor-I, individually and in combination, on periodontal regeneration in Macaca fascicularis. Journal of periodontal research. 1996;31(5):301-12.
137.Howell TH, Fiorellini JP, Paquette DW, Offenbacher S, Giannobile WV, Lynch SE. A phase I/II clinical trial to evaluate a combination of recombinant human platelet-derived growth factor-BB and recombinant human insulin-like growth factor-I in patients with periodontal disease. Journal of periodontology. 1997;68(12):1186-93.
138.Tatakis DN, Wikesjö UM, Razi SS, Sigurdsson TJ, Lee MB, Nguyen T, et al. Periodontal repair in dogs: effect of transforming growth factor-β1 on alveolar bone and cementum regeneration. Journal of clinical periodontology. 2000;27(9):698-704.
139.Rossa Jr C, Marcantonio Jr E, Cirelli JA, Marcantonio RA, Spolidorio LC, Fogo JC. Regeneration of Class III furcation defects with basic fibroblast growth factor (b-FGF) associated with GTR. A descriptive and histometric study in dogs. Journal of periodontology. 2000;71(5):775-84.
140.Nagayasu-Tanaka T, Anzai J, Takaki S, Shiraishi N, Terashima A, Asano T, et al. Action mechanism of fibroblast growth factor-2 (FGF-2) in the promotion of periodontal regeneration in beagle dogs. PLoS One. 2015;10(6):e0131870.
141.Cochran D, Oh T-J, Mills M, Clem D, McClain P, Schallhorn R, et al. A randomized clinical trial evaluating rh-FGF-2/β-TCP in periodontal defects. Journal of dental research. 2016;95(5):523-30.
142.Sculean A, Schwarz F, Chiantella GC, Miliauskaite A, Arweiler NB, Becker J. Nine-year results following treatment of intrabony periodontal defects with an enamel matrix derivative: report of 26 cases. International Journal of Periodontics & Restorative Dentistry. 2007;27(3).
143.Shirakata Y, Miron RJ, Nakamura T, Sena K, Shinohara Y, Horai N, et al. Effects of EMD liquid (Osteogain) on periodontal healing in class III furcation defects in monkeys. Journal of clinical periodontology. 2017;44(3):298-307.
144.Döri F, Arweiler NB, Szántó E, Ágics A, Gera I, Sculean A. Ten-year results following treatment of intrabony defects with an enamel matrix protein derivative combined with either a natural bone mineral or a β-tricalcium phosphate. Journal of periodontology. 2013;84(6):749-57.

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