*Received: 18th June 2015 Accepted: 25th September 2015 Conflicts of Interest: None*

*Source of Support: Nil*

**Review Article**

# Tissue Engineering in today's dentistry: A Review

***M S Rama Rao[1], M Naveen Kumar[2], Janavathi[3], Anwesh Reddy[4], K Karthik[5], C N Vanajakshi[6]***

***Contributors:***

[1]Professor and Head, Department of Conservative Dentistry, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India; [2]Reader, Department of Oral Pathology, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India; [3]Senior Lecturer, Department of Conservative Dentistry, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India; [4]Senior Lecturer, Department of Periodontics, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India; [5]Lecturer, Department of Oral Pathology, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India; [6]Senior Lecturer, Department of Oral Pathology, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India.

***Correspondence:***

Dr. Kumar MN. Department of Oral Pathology, Sree Sai Dental College and Research Institute, Srikakulam, Andhra Pradesh, India. Email: Naveen_motupalli@yahoo.com

***How to cite the article:***

Rao MS, Kumar MN, Janavathi, Reddy A, Karthik K, Vanajakshi CN. Tissue engineering in periodontics: A review. J Int Oral Health 2015;7(11):136-140.

***Abstract:***

Periodontal disease is a major issue in day-to-day public health and due to advances in the development of the effective therapies to treat the disease and regenerate periodontium is an important goal of today's medicine. Recently, tissue engineering is promising, as well as the most advanced approach to reconstruct the periodontal apparatus. Periodontics has an ancient history of utilizing advances in science to expand and to improve periodontal therapies. In the above context, there has been a substantial and growing public and scientific awareness of a relatively new field of applied biological research called tissue engineering and is been increasingly viewed as having enormous clinical potential biology life sciences. Hence, this review provides an insight into the basic principles of tissue engineering and its applications in treating periodontal diseases.

***Key Words***: Biologic research, growth factors, periodontal disease, periodontal regeneration, tissue engineering

## Introduction

"Regeneration" is the word which since historic periods frustrates many physicians and dentists as we still don't have any highly predictable means of achieving it with consistent results. Now-a-days, there is an increasing demand for replacement material to repair defective tissues/diseased organs because of a gradual increase in human life expectancy. The management of periodontal defects, which includes destruction of the periodontal ligament, cementum and the formation of intrabony defects, is most challengeable work in clinical periodontics.

The term tissue engineering was initially defined by the first National Science Foundation in 1988 "application of the principles and methods of engineering and life sciences toward fundamental understanding of structure-function relationship in normal and pathological mammalian tissues and the development of biological substitutes for the repair or regeneration of tissue or organ function" was sponsored in a meeting. It will differ from standard therapies in that engineered tissues become integrated within the patient, affording a potentially permanent and specific treatment of the disease state.

Technology of tissue engineering applies the principles of biology, chemistry, physics and engineering for the development of substitutes that will replace, repair biological function of diseased/damaged human body parts, by manipulating/reproducing cells through extracellular microenvironment. This three dimensional extracellular architecture ("scaffold") can be fabricated in the shape of the tissue to restore, with the help of polymer hydrogel, self-assembly/non-woven matrix, nano-fibrous electrospun matrices, three-dimensional weaving/any other textile technology-based techniques, depending on their structural and functional requisites. The concept in periodontics has begun with guided tissue regeneration; it is a mechanical approach utilizing non-resorbable membranes to regenerate periodontal defects. In dental implantology, guided bone regeneration membranes, with/without mechanical support, are used for bone augmentation.[1]

Due to the regeneration of periodontal tissues, their original form/architecture and function are restored. To appreciate what is involved, the cross-talk among the components of the periodontium, together with the inherent regenerative capacity of this tissue, need to be considered. When considering periodontal regeneration, we believe that at least four criteria must be met in order for regeneration to occur. These include functional epithelial seal, new connective tissue fibers, new acellular extrinsic fiber cementum and alveolar bone height restored to within 2 mm of the cementoenamel junction.

### *Periodontal tissue engineering*

Once considered a distant dream and only experimental in nature, tissue engineering is now clinically applicable with two

commercially available systems that involve the use of platelet derived growth factor-BB tricalcium phosphate (GEM 21) and bone morphogenic protein - Type 1 collagen sponge (INFUSE). The development of a third promising system utilizing basic fibroblast growth factor-2 is under trials.[2]

## The Concept of Tissue Engineering (Figure 1)

### *Signaling molecules*

Research is based mainly on two main approaches involving preparations containing biological mediators to selectively enhance the cells that populate the periodontal wound. The first approach utilized semi-purified preparations such as enamel matrix derivative and autologous platelet-rich plasma preparations. The second approach utilized recombinant growth factors such as recombinant platelet-derived growth factor-BB, human basic fibroblast growth factor, and bone morphogenic protein.[3]

### *Platelet rich plasma preparation*

Platelets are enriched by 338% and several growth factors like platelet-derived growth factor, transforming growth factor beta-1, fibroblastic growth factor-2, epidermal growth factor, vascular endothelial growth factor are seen.[4] This mixture of growth factors putatively stimulates the proliferation of fibroblasts and periodontal ligament cells, extracellular matrix formation and neovascularization. Platelet-rich plasma is used to stabilize graft materials for implant site augmentation and appears to increase early soft tissue healing.[5,6]

### *Enamel matrix derivative*

Enamel matrix derivative harvested from developing porcine teeth has recently been reported to induce periodontal regeneration as in a clinical case report. It contains a mixture of low molecular weight proteins that stimulate cell growth and the differentiation of mesenchymal cells including osteoblasts.[7]

### *Growth factors for biomimicry*

They are peptides which transmit signals between cells and thereby modulate their activity. They promote regeneration through a variety of cell-tissue interactions, promoting: Cell migration, attachment and subsequent spreading, cell proliferation, cell differentiation, and matrix synthesis. Several growth factors have been identified some of which are found in bone matrix, e.g. platelet-derived growth factor and fibroblast growth factor.[8]

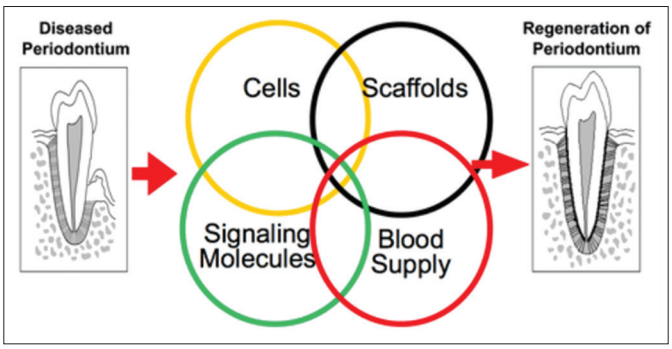


**Figure 1:** The four research areas that hold the key to the future of periodontal tissue engineering and regenerative medicine.

### *Bone morphogenetic proteins (BMP)*

Research by Urist and others has shown that bone demineralized in hydrochloric acid, lyophilized, and implanted in ectopic sites, has the potential to induce bone formation. This phenomenon has been termed the bone induction principle.[2]

### *The bone morphogenetic family*

The BMPs were discovered based on their presence in purified bone inductive extracts derived from bone. An extensive purification (more than 300,000-fold) was required to provide protein of sufficient purity. This suggests that the osteoinductive proteins are minor components of bone matrix, and present at lower levels than many other growth factors. BMP"s have been isolated from bovine and human sources out of which bone morphogenic protein-2 (osteopontin-2 [OP-2]), bone morphogenic protein-3 (osteogenin), and bone morphogenic protein-7 (OP-1) are of interest in periodontal regeneration.[9]

### *Gene therapy*

One of the major limitations associated with the use of growth and differentiation factors are their short biological half-lives. Gene therapy can be used for extended local delivery of this factors.[10] Recently, gene delivery of platelet-derived growth factor was accomplished by the successful transfer of the growth factor gene into the cementoblast and other periodontal cell types. Gene therapy studies utilizing bone morphogenic proteins have also been performed.[11,12]

### *Scaffold or supporting matrices*

Major roles for the Scaffold/supporting matrices are:

Structural reinforcement - To prevent collapse/damage of the surrounding tissue into the wound site. The barrier is used to restrict cellular migration in a selective manner. Scaffold - for cellular migration and proliferation and serve as a time release mechanism for signaling molecules.

### *Matrices (porous structures) absorbable*

Synthetic polymers namely polylactic acid and polyglycolic acid. Natural polymers such as collagen (Types I, II, III, IV), collagen-glycosaminoglycan copolymer, fibrin, and chitosan. Natural mineral, such as an anorganic bone, is available. Alternatively, a variety of xenograft, alloplastic, and allogenic grafting materials are available as scaffolding agents for tissue engineering.[2]

### *Substrate modification to enhance cell selection*

This type of scaffold contains specific surface peptides that selectively permit cell binding, osteoblastic phenotypic expression, and differentiation. These constructs can be used to

**Table 1: Cells and molecules participating in periodontal regeneration.**

| | |
|---|---|
| Cells | Epithetlial, fibroblasts, osteoblastic cells, junctional epithelium, gingival fibroblasts, periodontal ligament fibroblasts, osteoblasts, alveolar bone cells, cementoblasts |
| Molecules | Growth factors: Fibroblast growth factor 1 and 2, BMPs, insulin-like growth factor-I and II, adhesion molecules: Fibronectin, laminin, osteopontin, collagens, cementum attachment protein, structural proteins: Types I, III, V, XII and XIV collagens, proteoglycans, osteocalcin, tenasin, enamel matrix proteins |

BMP: Bone morphogenetic proteins

direct the regenerative response. They are still in experimental phase and require further investigation for clinical application.[13]

***Cells***

The cells used are autologous parenchymal cells, allogenic parenchymal cells, and marrow stromal stem cells (Table 1).

***Cell therapy***

It has been recently employed in periodontal surgery. The most common application involves a cell expansion strategy in an *ex vivo* environment followed by transplantation back into the defect area. Tissue-banked human fibroblasts or patient's connective tissue from attached gingiva of the retromolar area is harvested on a collagen/silicone bilayer membrane which later could be used as a donor tissue.[14]

**Applications of Tissue Engineering Principles: Periodontal Regeneration**

***Enamel matrix derivative***

It has been effective in the treatment of infrabony defects and has been shown to be safe for clinical use. The concern remains whether commercial batches will be consistent and provide comparable clinical results in all cases.[7]

***Recombinant human platelet-derived growth factor***

It is commercially available in combination with a tricalcium phosphate carrier. Preliminary studies suggest that it is easy to use, requires no barrier membranes and produced results similar or superior to other regenerative graft materials.[15]

***Recombinant human fibroblast growth factor-2***

Topical application of fibroblast growth factor-2 into intraosseous defects in alveolar bone induces significant periodontal tissue regeneration.[16]

**Application at Implant Site Preparation**

The challenge lies in regenerating adequate volume of hard and possibly soft tissue. Recombinant human platelet-derived growth factor, and recombinant human bone morphogenic protein-2 may be used for implant site preparation. Animal studies suggest that regenerated bone would have a similar bone-implant interface compared with native bone.[17]

A unique property of these periosteal stem cells and progenitor cells is that in all age groups, they retain their ability of differentiation into a variety of cell lineages. However, it has been found that their capacity to differentiate in the direction of chondrogenic and adipogenic lineages reduces with age.[18-20] Pericytes have also been recognized as a distinct cell population in the periosteum. They have a function of re-vascularization and promotion of bone formation, but their role in periosteal bone formation is deemed minimal at present.[21]

With the advancements in tissue engineering, Mizuno *et al.* illustrated a technique to regenerate periodontal defects using autologous membranous cultured periosteum (CP) or human CP (HCP) sheets. They reported considerable regeneration of the bone defects. Regarding the technique, a prior surgery is required before actual regenerative surgery to harvest a piece of the periosteum (usually from the posterior mandibular body). The periosteum specimen is then placed directly onto a culture dish in a pre-defined culture medium: 10% fetal bovine serum, 25 mg ascorbic acid, antibiotics (penicillin [100 IU/ml], and streptomycin [100 mg/ml]), and an antifungal agent (amphotericin-B [250 ng/ml]) and is incubated at 37°C; 10% $CO_2$. Culture medium has to be changed every 2-3 days. The periosteum samples are incubated until the cells form a sheet-like structure (4 weeks approximately).[22] Several reports have showed the efficacy of HCP sheets in the treatment of periodontal intrabony defects. All the cases showed significant improvements in clinical and radiographic parameters of the defects.[23-25]

**Role Pharmacological Agents for Periodontal Regeneration**

***Non-steroidal anti-inflammatory drugs (NSAID's)***

NSAID's are a class of drugs that will provide analgesic, antipyretic and anti-inflammatory effects. The basis of anti-inflammatory drugs in periodontal disease treatment is related to the control of prostaglandin E2 (PGE2) through the inhibition of cyclooxygenase-2 (COX-2) enzyme. Higher levels of PGE2 are associated with increased gingival inflammation and alveolar bone loss (Noguchi and Ishikava, 2007; Reynolds *et al.*, 2007; Tripton *et al.*, 2003).[26] Arachidonic acid metabolites are pro-inflammatory mediators that have been involved in a wide variety of resorptive processes involving bone, including diseases such as chronic periodontitis. These mediators can be potentially inhibited by NSAIDs, such as aspirin, ibuprofen, flurbiprofen, and naproxen. NSAIDs inhibit the enzyme COX, thereby preventing the production of arachidonic acid metabolites. Use of NSAIDs results in decreased levels of pro-inflammatory mediators that may limit the host-mediated alveolar bone destruction observed in periodontitis and peri-implant diseas.[27]

*Bisphosphonates (BP's)*

BPs were introduced in 1990 for the treatment of osteoporosis and osteolytic tumors. They are second group of drugs under investigation for their ability to modulate the bone loss and prevent bone resorption. They are non-biodegradable

analogs of pyrophosphate that have a high affinity for calcium phosphate crystals and that inhibit osteoclast activity.[28] BPs are drugs that suppress bone turnover, primarily through effects on osteoclasts, and are commonly prescribed to prevent skeletal-related events in malignancy and for benign bone diseases such as osteoporosis.[29]

***Hormonal therapy***

*Parathyroid hormone (PTH)*

The mechanism of the PTHs action is complex and involves pathways linked to common signaling peptides that affect osteoblast gene transcription. *In vitro* studies, *in vivo* experiments, and clinical trials demonstrated that intermittent PTH 1-34 administration induced anabolic effects on cancellous and cortical bone, enhanced bone mass, and increased mechanical bone strength.[30]

*Estrogen*

The relationship between estrogens and periodontal tissues was studied mainly for its possible implication in the inflammatory process, with a clear demonstration that estrogens do not increase inflammation. Most of the cells in the periodontium (fibroblasts, endothelial cells, epithelial gingival cells, osteoclasts, and osteoblasts) express estrogen receptors $\alpha$ and $\beta$. It is well known that estrogens decrease bone resorption and have a positive effect on bone formation.[31] Treatment with estrogens clearly inhibits bone loss as well as bone turnover and increases bone mineral density. There is substantial evidence that estrogen inhibits both osteoclast activity and differentiation by regulating the production of stimulatory and inhibitory cytokines by osteoblasts and monocytes. Various reports also have linked estrogen deficiency and osteoporosis to increased oral bone resorption, attachment loss, and tooth loss.[32-34]

**Conclusion**

To achieve our ultimate goal, we must take advantage of new scientific knowledge and tools, strengthen and expand partnerships, ensure that research advances are translated into useful technologies, and above all make sure that our scientific efforts benefit people. Despite the grandiosity of recent discoveries in the field of tissue engineering, more questions than answers exist due certain limitations, our ability to provide regenerative therapeutics continues to evolve. The challenge will be to select the optimal concentration of matrix, cells, and soluble regulators and aim to engineer tissues *in vivo*. Hence, we need to continue to improve our understanding of the physical and biological requirements necessary for specific tissue regeneration.

**References**

1. Lynch SE. Introduction to tissue engineering. In: Lynch SE, Genco RJ, Marx RE, editors. Tissue Engineering: Applications in Maxillofacial Surgery and Periodontics, Hanover, IL: Quintessence Publishing Co.; 1999. p. xi-xviii.
2. Kao RT, Murakami S, Beirne OR. The use of biologic mediators and tissue engineering in dentistry. Periodontol 2000 2009;50:127-53.
3. Marx RE, Carlson ER, Eichstaedt RM, Schimmele SR, Strauss JE, Georgeff KR. Platelet-rich plasma: Growth factor enhancement for bone grafts. Oral Surg Oral Med Oral Pathol Oral Radiol Endod 1998;85(6):638-46.
4. Landesberg R, Roy M, Glickman RS. Quantification of growth factor levels using a simplified method of platelet-rich plasma gel preparation. J Oral Maxillofac Surg 2000;58(3):297-300.
5. Döri F, Huszár T, Nikolidakis D, Arweiler NB, Gera I, Sculean A. Effect of platelet-rich plasma on the healing of intrabony defects treated with an anorganic bovine bone mineral and expanded polytetrafluoroethylene membranes. J Periodontol 2007;78(6):983-90.
6. Yassibag-Berkman Z, Tuncer O, Subasioglu T, Kantarci A. Combined use of platelet-rich plasma and bone grafting with or without guided tissue regeneration in the treatment of anterior interproximal defects. J Periodontol 2007;78(5):801-9.
7. Heijl L. Periodontal regeneration with enamel matrix derivative in one human experimental defect. A case report. J Clin Periodontol 1997;24:693-6.
8. The potential role of growth and differentiation factors in periodontal regeneration. J Periodontol 1996;67(5):545-53.
9. Massagué J. TGF-beta signal transduction. Annu Rev Biochem 1998;67:753-91.
10. Fang J, Zhu YY, Smiley E, Bonadio J, Rouleau JP, Goldstein SA, *et al.* Stimulation of new bone formation by direct transfer of osteogenic plasmid genes. Proc Natl Acad Sci U S A 1996;93(12):5753-8.
11. Giannobile WV, Lee CS, Tomala MP, Tejeda KM, Zhu Z. Platelet-derived growth factor (PDGF) gene delivery for application in periodontal tissue engineering. J Periodontol 2001;72(6):815-23.
12. Jin Q, Anusaksathien O, Webb SA, Printz MA, Giannobile WV. Engineering of tooth-supporting structures by delivery of PDGF gene therapy vectors. Mol Ther 2004;9(4):519-26.
13. Healy KE, Rezania A, Stile RA. Designing biomaterials to direct biological responses. Ann N Y Acad Sci 1999;875:24-35.
14. Reddy MS. Achieving gingival esthetics. J Am Dent Assoc 2003;134(3):295-304.
15. 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. J Periodontol 2005;76(12):2205-15.
16. Takayama S, Murakami S, Shimabukuro Y, Kitamura M, Okada H. Periodontal regeneration by FGF-2 (bFGF) in primate models. J Dent Res 2001;80(12):2075-9.
17. Boyne PJ. Animal studies of application of rhBMP-2 in maxillofacial reconstruction. Bone 1996;19:83S-92.

18. Lim SM, Choi YS, Shin HC, Lee CW, Kim DI. Isolation of human periosteum-derived progenitor cells using immunophenotypes for chondrogenesis. Biotechnol Lett 2005;27(9):607-11.
19. O'Driscoll SW, Saris DB, Ito Y, Fitzimmons JS. The chondrogenic potential of periosteum decreases with age. J Orthop Res 2001;19(1):95-103.
20. Justesen J, Stenderup K, Eriksen EF, Kassem M. Maintenance of osteoblastic and adipocytic differentiation potential with age and osteoporosis in human marrow stromal cell cultures. Calcif Tissue Int 2002;71(1):36-44.
21. Diaz-Flores L, Gutierrez R, Lopez-Alonso A, Gonzalez R, Varela H. Pericytes as a supplementary source of osteoblasts in periosteal osteogenesis. Clin Orthop Relat Res 1992(275):280-6.
22. Mizuno H, Hata K, Kojima K, Bonassar LJ, Vacanti CA, Ueda M. A novel approach to regenerating periodontal tissue by grafting autologous cultured persiosteum. Tissue Eng 2006;12(5):1227-335.
23. Mizuno H, Kagami H, Mase J, Mizuno D, Ueda M. Efficacy of membranous cultured periosteum for the treatment of patients with severe periodontitis: A proof-of-concept study. Nagoya J Med Sci 2010;72(1-2):59-70.
24. Yamamiya K, Okuda K, Kawase T, Hata K, Wolff LF, Yoshie H. Tissue-engineered cultured periosteum used with platelet-rich plasma and hydroxyapatite in treating human osseous defects. J Periodontol 2008;79(5):811-8.
25. Okuda K, Yamamiya K, Kawase T, Mizuno H, Ueda M, Yoshie H. Treatment of human infrabony periodontal defects by grafting human cultured periosteum sheets combined with platelet-rich plasma and porous hydroxyapatite granules: Case series. J Int Acad Periodontol 2009;11(3):206-13.
26. Vlad C, Vlad DC, Bucur A, Popescu R, Dumitraşcu V. Beneficial effects of selective cyclooxigenase-2 inhibitor etoricoxib in periodontitis. Ann RSCB 2012;22:211-6.
27. Reddy MS, Geurs NC, Gunsolley JC. Periodontal host modulation with antiproteinase, anti-inflammatory, and bone-sparing agents. A systematic review. Ann Periodontol 2003;8(1):12-37.
28. Deshmukh J, Jawali MH, Kulkarni VK. Host modulation therapy – A promising new concept in treating periodontal diseases. Int J Dent Clin 2011;3:48-53.
29. Borromeo GL, Tsao CE, Darby IB, Ebeling PR. A review of the clinical implications of bisphosphonates in dentistry. Aust Dent J 2011;56(1):2-9.
30. Valderrama P, Jung RE, Thoma DS, Jones AA, Cochran DL. Evaluation of parathyroid hormone bound to a synthetic matrix for guided bone regeneration around dental implants: A histomorphometric study in dogs. J Periodontol 2010;81(5):737-47.
31. Nuñez J, Sanz-Blasco S, Vignoletti F, Muñoz F, Caffesse RG, Sanz M, *et al* 17beta-estradiol promotes cementoblast proliferation and cementum formation in experimental periodontitis. J Periodontol 2010;81(7):1064-74.
32. McCauley LK, Nohutcu RM. Mediators of periodontal osseous destruction and remodeling: Principles and implications for diagnosis and therapy. J Periodontol 2002;73(11):1377-91.
33. Singh N, Uppoor A, Naik DG. Bone's smart envelope - The periosteum: Unleashing its regenerative potential for periodontal reconstruction. Int J Contemp Dent Clin Med Rev 2015;2015:Article ID: 090215, doi: 10.15713/ins.ijcdmr.62.
34. Nandini TK, Mahantesha S, Mani R, Kranti K. Pharmacological agents for periodontal regeneration: A review. Int J Contemp Dent Med Rev 2015;2015: Article ID: 120115, doi: 10.15713/ins.ijcdmr.35.