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*Review*

# Applications of recombinant protein therapeutic agents in periodontics contributors

**Neha Sethi**

Department of Periodontics College, IDEAS Dental College, Address, Gwalior, India.

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**Based on the improved understanding of cell and molecular biology of periodontal wound healing, the recombinant technology comprising rh growth factors and carrier construct is applied in periodontal regeneration. In 1997, the first recombinant (that is, synthetic) protein therapeutic agent was approved by the US Food and Drug Administration (FDA). In this paper we review the two commercially available recombinant agents, that is, rhPDGF-BB and rh-BMP-2 used for periodontal regeneration.**

**Key words:** Periodontal regeneration, recombinant proteins, gene, tissue regeneration.

## INTRODUCTION

According to Murakami and Noda (2000), in normal wound healing multiple cytokines act in concert to regulate the cellular functions of various cell types within and adjacent to a wound in nearly all tissues, including the periodontium. Also, signalling molecules such as growth factors and morphogens are capable of stimulating cellular events. Tissue engineering or recombinant technology could be a more predictable modality, which can modulate the wound healing with supply of abundant growth factors.

Tissue engineering is a relatively new field of reconstructive biology which utilizes mechanical, cellular, or biologic mediators to facilitate reconstruction/regeneration of a particular tissue.

The goal of tissue engineering and regenerative medicine is to promote healing and ideally, true regeneration of a tissue's structure and function more predictably, more quickly, and less invasively than allowed by previous techniques.

## TISSUE ENGINEERING TRIAD

An ideal approach to tissue engineering is based on sound principles of developmental and molecular biology of signal transduction, and of the cell biology of tissue morphogenesis, including the supramolecular assembly of the extracellular matrix. Using tissue engineering, the wound healing progress is manipulated so that tissue regeneration occurs.

This tissue engineering approach to bone and periodontal regeneration combines three key elements to enhance regeneration (Lynch, 2008) (Figure 1).

1. Conductive scaffolds.
2. Signalling molecules.
3. Cells.

Cells are considered as a major component of the tissue regeneration process. Stem/progenitor cells contribute to the regeneration process. It also requires a scaffold or a supportive template which is necessary for the organization of these replicating cells. And in addition, it requires the presence of certain signaling molecules which act as growth and differentiating factors.

### Recombinant protein therapeutics

Characteristics include:

E-mail: neha.sethi85@gmail.com. Tel: +91-9780866984.

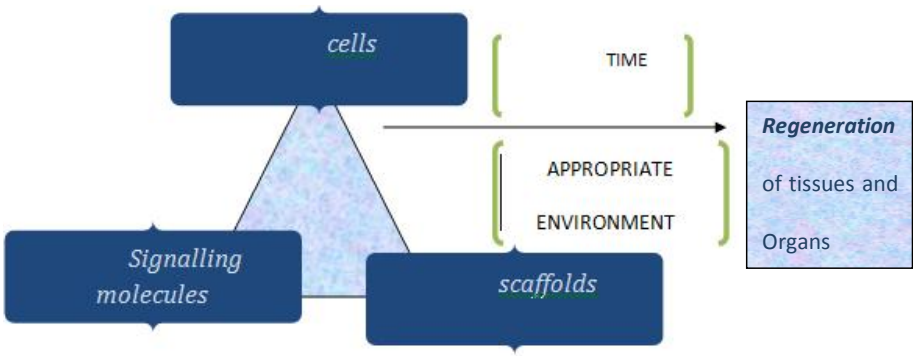


**Figure 1.** Tissue engineering triad.

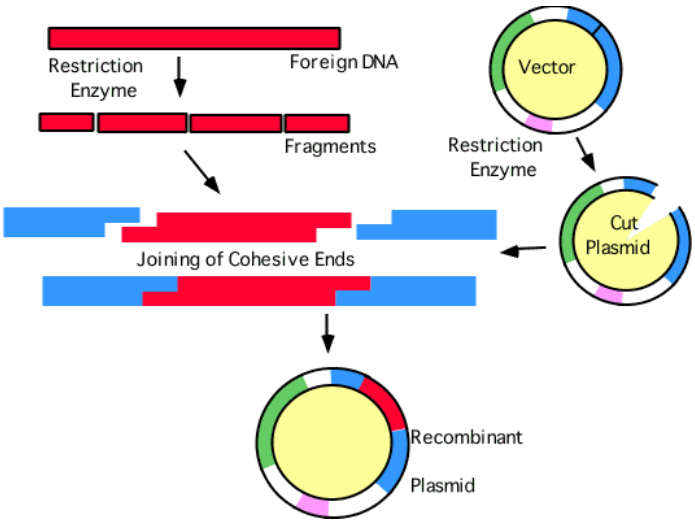


**Figure 2.** Formation of recombinant protein.

1. Highly concentrated growth modulating molecules. Sutherland and Bostrom, 2007
2. Increased predictability of regenerative results for clinician and patients. Sutherland and Bostrom, 2007
3. Combination products such as regenerative proteins with tissue specific matrices (scaffolds) is the emerging trend.
4. Promising approach to periodontal regeneration.

## RECOMBINANT PROTEINS

**Derived from:** Recombinant DNA.
**Recombinant DNA:** Is a form of artificial DNA created by either combining 2 or more DNA sequences or inserting it into another DNA strand.

### Mechanism/ Procedure

-The gene/ specific DNA sequence from the human cell is selected and isolated.

- Gene is isolated and carefully grafted onto a vector in order to be cloned (the vectors like bacteria are able to grow independently hence are the ideal choice for this purpose).
-Little section of the vector's gene is removed and the gene of interest is implanted into it.
-Bacterial plasmid is the transfected into host cells like yeast/ Chinese hamster ovarian cells/ *Escherichia coli*, capable of large scale growth.
-The grafted gene grows with the vector gene it thereby creating the gene scientists wanted.
- Once it has been created, scientists carefully extract it from them.
-Thus, proteins are synthesized, concentrated purified and packaged in large sterile quantities (Figure 2).

### Applications

i) To diagnose and treat a number of genetic disorders.
ii) To isolate proteins and for therapeutic purposes.
iii) To determine gene sequences and mutations.

**Table 1.** US FDA approved recombinant protein therapeutics.

| Recombinant protein therapeutics | Approved indication |
|---|---|
| rhPDGF-BB (gel) | - Treatment of neuropathic ulcers (Wieman et al., 1998).<br>- No applications in periodontics (Nevins and Giannobile, 2003). |
| rhPDGF-BB (with β tricalcium phosphate) | Treatment of intrabony and furcation periodontal defects and gingival recession associated with periodontal defects (Nevins and Giannobile, 2003) |
| rhBMP-2 (with type 1 collagen sponge) | As an alternative to autogeneous bone graft for sinus augmentations and for localized alveolar ridge augmentations for defects associated with extraction sockets (Boyne and Marx, 1997) |

**Table 2.** *In vivo* studies.

| Factor | Animal studies | Result | Studies |
|---|---|---|---|
| PDGF-BB | Beagle dog | Promoted PDL fibroblast proliferation | Wang et al. (1994) |
| | Monkey | New attachment | Giannobile et al. (1996) |
| PDGF/dexamethasone | Monkey | Regeneration | Rutherford et al. (1993) |
| PDGF –BB/ePTFE/citric acid | Beagle dog | Regeneration | Cho et al. (2002) |
| BMP-2 | Baboon | Regeneration | Ripamonti et al. (1994) |
| | Beagle dog | Increase in bone and cementum (rare ankylosis or resorption) | Sigurdsson et al. (1993) |

iv) First applied clinically tube used as recombinant human insulin.

Other medically used recombinant forms include:

i) rh growth hormone
ii) rh blood clotting factors
iii) rh hepatitis vaccine
iv) rh PDGF
v) rh BMP 2,7.

## RECOMBINANT PROTEINS IN PERIODONTAL REGENERATION

List of recombinant proteins used in various trials of periodontal regeneration include:

1) rh PDGF-BB plus rh IGF-1
2) rh PDGF-BB plus β-TCP
3) rh BMP-2 with type I collagen
4) rh bFGF (rh FGF-2)
5) rh TGF-β
6) rh osteogenic potential 1/ BMP-7
7) BDNF
8) GDF-5.

To date, only three recombinant growth factor products have been widely *commercialised*, for use in tissue regeneration (Table 1) ( Lynch, 2008)

Among these only rh PDGF-BB with tricalcium phosphate and rh BMP-2 with type 1 collagen sponge are used in periodontics.

Review of articles on *in vivo*, *in vitro* and human studies are as shown in Tables 2, 3 and 4, respectively.

## RECOMBINANT HUMAN PLATELET DERIVED GROWTH FACTOR (PDGF)–BB

Platelet derived growth factor (PDGF) is a naturally occurring protein found abundantly in bone matrix forms which includes PDGF-AA, PDGFB and PDGF-AB. Major sources include platelets, macrophages, epithelial cells, endothelial cells, smooth muscles and bone matrix.

### Significance

1. Released locally during clotting by blood platelets at the site of injury.
2. Stimulates wound healing response.
3. Promotes raid cellular migration (chemotaxis).

**Table 3.** *In vitro* studies.

| Factor | Cell type | Result | Studies |
|---|---|---|---|
| PDGF- BB | Rat PDL cells | Mitogenic effect PDGF>FGF>EGF | Blom et al. (1994) |
| | Human PDL cells | Increased mitogenic activity | Oates et al. (1993) |
| | | An increased proliferation plus chemotaxis: PDGF BB>AB>AA | Boyan et al. (1994) |
| | | Capable of upregulating collagen synthesis in the extracellular matrix | Tomoyuki kawase (2003) |
| | Human gingival fibrobalsts | Increased hyaluronate synthesis, blocked inhibitory effects of LPS on cell growth | Bartold and Raben (1992) |
| | | Mitogenic | Piche et al. (1992) |
| | Human PDL and gingival fibroblasts | PDGF alone had a greater proliferation effect | Boyan et al. (1994) |
| | Osteoblasts | Promote chemotaxis, matrix synthesis, and mitogeneis | Canalis et al. (1989)<br>Piches Graves (1992)<br>Hughes and Aubin (1992) |
| | | Involved in maturation and remodelling of newly formed blood vessel, angiogenic and vasculogenic cells might act as important target initially responding to this mitogenic factor | Risau (1997). |
| | | Have direct and indirect effects on bone resorption by the upregulation of collagenase transcription | Rydziel et al. (2000) |
| | | increase in IL 6 expression in osteoblasts | Franchimont et al. (1997) |
| | Mesenchymal cells | Accelerated provisional extracellular matrix deposition and subsequent collagen formation | Glenn et al. (2004) |
| BMPs | Osteoblasts | Promote osteoblast phenotype | Ripamonti et al. 1994 |
| | | osteopontin and osteocalcin expressed in late stages of osteoblast differentiation | Hirota et al. (1994) |
| | | Sequential expression of osteopontin and osteocalcin mRNA in the process of ectopic bone formation | |
| | | Osteocalcin production depends on BMP-2 concentration | Zhao et al. (2003) |
| | | Upregulate Cbfa1/Runx2 under certain conditions during osteoblast differention therfore, this is a candidate down stream target of BMPs , although Smad complexes can also directially interact and activate target genes independently of Cbfa1/Runx2 | Jonk et al. (1998) |
| | Mesenchymal cells | Stimulates osteopontin and osteocalcin. | Lecanda et al. (1997) |
| | | BMP-2,induces the differentiation of undifferentiated cells, 2T9 (osteoblast progenitor cells), in the lineage | Schwartz and Ren (2000) |
| | | Recombinant BMP-2 increased alkaline phosphatase activity and osteocalcin production in the bone marrow stromal cell line | Rosen and Thies (1992) |
| | | Raising the possibility that BMP-2 may be involved in the differentiation of osteoblasts from progenitor cells resident in the bone marrow. | |
| | Periodontal ligament cells | No increase in osteopontin or bone sialoprotein within periodontal ligament | Rajshankar et al. (1998) |

**Table 4.** Human studies.

| Factor | Defect | Result | Studies |
|---|---|---|---|
| PDGF-BB | Intrabony defect and furcations | Bone fill was seen and there was gain in attachment. | Nevins and Giannobile (2003), Nevins et al. (2005), Nevins et al. (2007) and McGuire et al. (2006) |
| BMP-2 | Intrabony defects<br>Furcation defects | Only clinical attachment was gained | Nevins and Giannobile (2003). |
| | Sinus elevation | Bone fill of about 13.4 mm was obtained | van den Bergh et al. (2000) |

4. Promotes cellular proliferation (mitogenesis).
5. Promotes regeneration of periodontal tissues including bone, cementum, PDL – (Lynch, 1980).

In 1997, the first recombinant (that is, synthetic) protein therapeutic agent was approved by the US Food and Drug Administration (FDA). The product provides recombinant human PDGF-BB in gel formulation for the treatment of recalcitrant neuropathic dermal ulcers in diabetic patients.

In 2005, rhPDGF-BB + β tricalcium phosphate product was approved for bone and periodontal regeneration and treatment of gingival recession. This product contains approximately 1,000 times higher concentration of PDGF than the level commonly obtained through platelet concentration. (Bowen-Pope etal., 1988, Huang et al., 1983)
rh PDGF-BB is more than 98% pure recombinant protein developed using conventional recombinant expression techniques under highly controlled conditions.

In a landmark study:

1. Nevins and Giannobile (2003) showed histological evidence of periodontal regeneration in treated intrabony furcation defects with rh PDGF-BB.
2. Nevins (2005) conducted a clinical study on humans with rh PDGF-BB delivered with β-TCP for advanced periodontal osseous defects. Results showed larger gain of CAL, greater bone gain and percentage of defect fill with the combination.
3. No adverse affects such as root resortion, ankylosis, inflammation were reported.
4. Also, rh PDGF improves bone healing at tooth extraction sites (Cardaropoli, 2003), in patients with diabetes and osteoporosis and in peri-implant bone (Berglundh, 2008).
5. Also, McGurie (2006) use PDGF-BB with bone graft material and covered it with collagen membranes in recession sites, thus providing results comparable to CT grafts with no need for second surgical site.

## Role of rh PDGF-BB in periodontal regeneration

1. It promotes DNA synthesis and chemotaxis in periodontal ligament cells especially in osteoblastic phenotype (Wang et al., 2004).
2. It stimulates collagen and non collagen proteins synthesis (Lynch et al., 1991).
3. In cultures of osteoblast-like cells, it downregulates alkaline phosphatase activity and osteocalcin (Zaman et al., 1999).
4. It enhances demineralised bone matrix induced cartilage and bone formation (Howeels, 1997).
5. PDGF increases the pool of osteogenic cells, and cells that will differentiate into cementoblasts and periodontal ligament cells (that is, acts as a chemotactic agent and mitogen); whereas their subsequent differentiation into osteoblasts or chondrocytes is directed by BMP family (Cho et al., 2002; kugimiya et al., 2005), heghehog proteins, (Murakami and Noda, 2000) and activation of the Wnt- signalling pathway. (Hadjiargyrou et al., 2002)
6. It exerts indirect effects on bone regeneration by increasing the expression of angiogenic molecules such as vascular endothelial growth factor (VEGF) (Bouletreau et al., 2002) and hepatocyte growth factor/scatter factor, as well as the proinflammatory cytokine interleukin-6. VEGF is a key molecule in bone regeneration.

## Mechanism of action

Genetic models demonstrate that endothelial cell derived PDGF-BB is required to recruit PDGFR-β positive cells and stimulate blood vessel maturation.

PDGF-BB from endothelial cells
↓
chemoattractant & mitogen for mural cells
(pericytes & smooth muscle cells)
↓
Destabilizes blood vessels
↓
Sprouting and filamentous web formation
↓
Recruits PDGFRβ+ cells essential for vessel formation

PDGFs can modulate the responsiveness of osteogenic cells to BMPs by increasing the expression of gremlin and IGF signalling. The responsiveness of osteogenic cells to PDGFs can be regulated by the inflammatory cytokine interleukin-1, which inhibits PDGFRα expression in MG-63 cells and human osteoblastic cells

**Clinical applications**

1. rh PDGF-BB has been used to promote bone regeneration around endosseous implants. Allori et al., 2008
2. FDA has cleared the clinical use of rhPDGF-BB for chronic skin wounds in diabetic patients (Regranex, Ethicon) and for periodontally related osseous defects (GEM 21S, BioMimetic Therapeutics) and rh BMP-2 (InFuse Medtronic Sofamor Danek) for anterior interbody spine fusion, open tibial fractures, sinus elevations, and defects associated with tooth extraction (Lynch 2008).
3. It is noteworthy to consider the therapeutic role for rh PDGF for the compromised bone wound healing in patients with diabetes. It has been shown there is a decrease in cellular proliferation in the fracture callus and a decrease in levels of PDGF transcripts in diabetic rats, suggesting a correlation between PDGF levels and fracture healing response. (Pietrzak and Eppley 2005)
4. In fenestration defects in alveolar bone, recombinant PDGF–BB applied to root surfaces increased proliferation of periodontal ligament, cementoblasts, osteoblasts, perivascular cells and endothelial cells Hollinger et al., 2008.

**GEM - 21S GROWTH FACTOR ENHANCED MATRIX**

FDA approved components include:

- Synthetic β tricalcium phosphate.
- Highly porous and resorbable.
- Osteoconductive scaffold/ matrix.
- Provides framework for bone growth.
- Aids in preventing collapse of soft tissue.
- Promotes stabilization of blood clot.
- Pore diameter of scaffold → 1 to 500 µm.
- Particle size → 0.25 to 1 mm.
- Recombinant PDGF-BB.
- Native protein constituent of blood platelets.
- Causes mitogenis, angiogenic and chemptactic effects on bone and PDL cells.

**Indications**

- Intrabony periodontal defects.
- Furcation periodontal defects.
- Gingival recession associated with periodontal defects.

**Contra-indications**

- Untreated acute infections at surgical site.
- Untreated malignant neoplasm at the surgical site.
- Known hypersensitivity to the product components.
- General contra indications to grafting / surgery.

**Warnings**

- Various features of recombinant human PDGF (GEM 21s) are yet unknown.
- Interactions with other medications are unknown.
- Carcinogenesis, reproductive toxicity are unknown.
- Effects in pregnant and nursing women are unknown.
- Effects in smokers/ tobacco users are unknown.
- Effects in pediatric patients are unknown.
- Also GEM -21s is intended to be placed in periodontally related defects. Must NOT be injected systemically.
- Radio-opaque in nature and should be considered during evaluation. It is comparable to the radio-opacity of bone initially, diminishes as it is resorbed.

**Supply**

Each kit consists of:

- One cup containing 0.5cc of β-TCP particles (0.25 to 1 mm).
- One syringe containing solution of 0.5 ml rh PDGF (0.3 mg/ml).

**Cost**

- 0.5 cc of β TCP / 0.5 ml of PDGF (0.3 mg/ml) →$300.

**Directions to use**

1. Appropriate sterile conditions should be maintained.
2. B βTCP and PDGF are to be mixed. Following a waiting period of 10 min, saturated GEM 21S is placed into the defect.
3. Placement should be with moderate pressure at the level of surrounding bone walls.
4. The kit should not be resterlized/ reused.

**Storage**

- To be refrigerated at 2 to 8°C.
- β TCP can be stored at room temperature.

**Clinical trial**

The use of GEM 21 s based on the study by Nevins et

al., (2005) concluded:

1. Dosage of rh PDGF → 0.3 mg/ml.
2. Showed improved periodontal parameters over two years.
3. Resulted in better regeneration in comparison to emdogain.

**Advantages**

1. Clinical and radiographic benefits of regeneration.
2. Better outcomes than enamel matrix derivates.
3. Speedy clinical attachment level gains, reduction in gingival recession and improved bone growth.
4. No need for second surgery (as in autogenous bone graft sites).
5. Also, when combining periodontal therapy with rh PDGF and Er:YAG laser, promising results have been shown.

**Disadvantages**

1. High cost.
2. Various interactions with drugs and systemic health – unknown.
3. Handling difficulties.
4. Mild surgical adverse events – swelling, bleeding, dizziness, difficult breathing, headaches, anaphylaxis.
5. Long term benefits / adverse effects are still unknown.

**BONE MORPHOGENETIC PROTEINS (BMPs)**

Bone morphogenetic proteins (BMPs) are morphogens and differentiation factors originally isolated from bone matrix based on their ability to induce ectopic bone formation, that is, bone formation de novo where bone does not normally exist, such as in subcutaneous or intramuscular site. Wozney et al., 1988). It should be noted that BMP is a member of TGF–β family. In 1965, Urist showed that crude bone extracts induced new bone in ectopic site in muscle pouch in rat model. He coined the term 'bone morphogenetic protein'. The main sources of BMPs are the bone and kidney cells.

**Role**

- Act as growth and differentiation factors.
- Act as chemotactic factors/ agents.
- Differentiate stem cells from surrounding mesenchymal cells/ tissue and bone forming cells.
- Also stimulate angiogenesis and migration and proliferation of stem cells.

**Recombinant Human BMP-2**

RhBMP-2 in combination with a type I bovine collagen sponge has been approved in the US by the FDA for use in spinal fusion, tibial fracture repair, and most recently, as an alternative to autogenous grafts in sinus augumentation and extraction socket grafting procedures in skeletally mature patients. Preclinical results do not support the appropriateness of rhBMP-2 for the treatment of human periodontal defects. Lim et al., 2003) It is an active ingredient in osteoinductive grafts.

The primary activity of rhBMP-2 appears to be differentiating mesenchymal precursor cells into mature osteoblasts and/or chondroblasts. In addition, rhBMP-2 is chemotactic for some osteoblastic-type cells. RhBMP-2 has been shown to induce the complete sequence of endochondral ossification.

**Effect on cells in periodontal soft tissue and bone healing**

1. Stimulate proliferation and migration of undifferentiated bone cell precursors and induce new bone formation (Sigurdsson, 1993).
2. Helps undifferentiated pleuripotent cells to differentiate into cartilage and bone forming cells (Boden, 2001).
3. Act as chemoattractant for mesenchymal cells
4. Stimulate alkaline phosphatase activity, thus stimulates bone formation. Rosen and Thies, 1992 .
5. Helps in formation of bone matrix (Yasko 1992).
6. Along with bFGF, BMP-2 stimulates angiogenesis Li et al., 2005.

**Effect on periodontal ligament cells**

1. Stimulate matrix synthsesis.
2. Stimulate cementoblast proliferation.
3. Stimulate cementum production.
4. Regulate the proliferation and mitogenesis of the cells of osteoblastic lineage.
5. Stimulate maturation of osteoblastic cells.
6. Stimulate alkaline phosphatase activity, thus in turn stimulating increased bone formation.
7. Induce osteoblastic transformation of stromal cells.
8. Along with basic fibroblast growth factor it stimulates angiogenesis.

**Clinical applications**

1. Maxillofacial reconstruction (Boyne et al., 2005).
2. Alveolar ridge augmentation (Barboza et al., 2004).
3. Sinus floor augmentation (Boyne and Marx, 1997; Boyne et al., 2005).
4. Implant fixation (Hanisch et al., 2003; Bessho et al., 1999).

**Periodontal regeneration**

Wikup (2003, 2004) showed significant augmentation of alveolar ridge, used a dome shaped space providing porous expanded PTFE device to create unobstructed space to obviate the compression of rhBMP-2/ ACS. Thus allowing vascularity from gingival connective tissue.

**Carrier systems**

Several carrier systems have been screened to evaluate their efficacy and biocompatibility with BMPs. Ideal requirements include:

1. Maintaining its structural integrity at the target site.
2. Releasing BMPs in desired concentration over time.
3. Non obstruction of bone formation, thus undergo timely resorption.
4. Should not compromise the physiological and biochemical properties of bone.

**Carriers under evaluation**

Craniofacial indications in animal models include:

- Hydroxapatite- particulate/ putty formulations
- βTricalcium phosphate
- Calcium sulphate
- Calcium phosphate
- Calcium carbonate
- Bioglass
- Organic polymers
- Allogenic/ xenogenic collgen preparations
- Absorbable collgen sponge
- Wikup, 2003, 2004 – used rhBMP-2 and ACS with / without ePTFE
- Surpra alveolar defects: Need scaffolds with rhBMP-2
- Intrabony defects: May be treated successfully with rhBMP-2 only.

**Alternative carrier systems**

Wikisjo et al. (2003) investigated rhBMP-2 in calcium phosphate cement matrix.

**Indications**

- Can be easily shaped to desired contour.
- Provides space for rh BMP to induce bone formation.
- Injectable (for inlay and minially invasive technology).
- Maxillary sinus augmentation with titanium implants placement.

**Clinical applications**

- Infuse.
- FDA approved.

**Components**

- Rh BMP
- ACS- absorbable collagen sponge.
- Is a bovine type I collagen matrix.

**Supportive clinical trials**

1. van den Bergh et al., (200) reported significant sinus floor augmentation with both rhBMP-2 and rhOP-1.
2. Hanisch et al., (2003) reported re-osseointegeration of endoosseous implants exposed to peri-implantitis.
3. Jovanovic et al. (2003) established normal physiologic bone formation, osseointegration and long term functional loading of implants.

**Clinical indications**

1) Sinus augmentation.
2) Alveolar ridge augmentation:

a) Dose: 0.2 to 1.75 mg.
b) Inlay (extraction site) - exhibited significant bone formation (Florellini, 2005).
c) Onlay (ridge augmentation) showed negligible regeneration (Barboza et al., 2004; Barboza et al., 2000).

3) Craniofacial reconstruction:

a) Acute / chronis post traumatic discontinuity defects
b) Congenital malformations
c) Tumour resection defects

4) Supports dental implants:

a) Significant occeointegration.
b) Recently,' Bone Inductive Implants' → titanium implants with purpose- designed surface serving as a vehicle for rh BMP-1 is being developed.

**Exclusion**

- Pregnancy.
- Hypersensitivity to the components.
- Infection or tumor.
- Systemic illness.

**Possible complications**

- Allergic reactions
- Bleeding
- Infection

- Pain, discomfort, swelling, etc.

**Drawbacks**

- Carrier – ACS→ is vulnerable to tissue compression.
- Less effective for inlay indications (intrabony defects).
- High cost.
- Possible adverse reactions.
- Poor results in periodontal regeneration. Recent study by Song (2011) suggests reduced collagen synthesis and increased adipogenic differentiation by human PDL cells under rhBMP-2 effect.

**GROWTH DIFFERENTIATION FACTOR**

- Member of TGF β superfamily.
- Also called cartilage derived morphogenetic potein-1.

**Roles**

***GDF -5,6,7***

- In animal studies suggest important regulatory roles in periodontal attachment.

***GDF-5***

- Plays critical role in mesenchymal cell recruitment inducing cartilage and bone formation, and ligament cell differentiation in morphogenesis.
- Promotes PDL cells proliferation by influencing ECM metabolism (Nakamura et al., 2003).
- Supports and accelerates periodontal tissue formation.
- Shows no evidence of ankylosis or root resorption.
- However, it induces bone regeneration less aggressively as compared to rh BMP-2, BMP-7.

**Recombinant forms**

***Rh GDF-5 + PLGA (polylacticoglycolic acid)***

- Cortellini Tonetti (2001, 2007) supported minimally invasive regenerative procedures.
- Herbery (2008) reported easy to use in contained and non contained periodontal defects.
- Kimura et al., (**2003**) demonstrated stimulation of periodontal regeneration.

***Rh GDF -5 + β TCP***

- Lee (2010) reported potential to support periodontal attachment in one walled intrabony defects.
- Further long term studies are necessary to confirm uneventful regeneration in human periodontal tissues.

***Osteogenic protein-1 (rh BMP-7)***

- Approved for bone regeneration in long bone fractures and lumbar spine fusion.
-van den Bergh et al., (2009) reported significant sinus floor augmentation with rh OP-1.
- Has been evaluated for significant sinus floor augmentation procedure with BMP–2.

**DISCUSSION**

**Pros**

Recombinant proteins appears to be a promising solution to clinical problems leading to

- rapid periodontal regenerative capability with more predictability.
- optimal compatibility for clinical applications.
- without risk of potential immunological reactions.
- without risk of transmission of infections.

1. rh PDGF- in intrabony and furcation defects.
2. rh BMP/ ACS- in augmentation of maxillary sinus and alveolar ridge.

-in osseointegration of endosseous implants and re-osseointegration of implants.

**Cons**

Although promising, the currently available growth factors provide limited clinical benefits. Loop holes include:

- Inappropriate doses.
- Inappropriate delivery systems.
- Expensive.
- Carriers with lacking ability of cell adhesion.
- Carrier systems resorbing untimely to the wound repair process.
- Recombinant technology relies on the inherent ability of transfected cells like yeast/ Chinese hamster cells / *E. coli* which could produce rh GFs of demonstrated biological activity.
- Variable healing responses lead to variable regenerative results.
- Wound healing requires various growth factors (and not just one) to act together to regulate cellular events.
- Recombinant protein therapy offers single growth factor which may be inadequate to achieve the desirable effects eg: rh BMP, rh GDF.

Thus an improved regenerative synthetic product may be synthesized by combining highly concentrated GFs in required amounts, so that the cocktail can successfully regenerate the lost periodontium.

## CONCLUSION

Based on the improved understanding of cell and molecular biology of periodontal wound healing, the recombinant technology comprising rh growth factors and carrier construct is applied in periodontal regeneration. Recombinant proteins represent a major evolution in regenerative therapies and have a potential to become a new standard of care broadening the scope of clinical practice. Whereas, still the knowledge in the area needs to be broadened to accept this tissue engineering trend as a regular regenerative therapy.

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