Role of Bone Morphogenetic Proteins in Periodontics

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Abstract- Bone Morphogenetic Proteins which are part of extracellular matrix are capable of inducing de novo bone formation. De novo bone formation is essential in treating non-union of fractures, periodontal defects or in patients with tissue irradiation, those in whom there is a necessity to augment alveolar ridge or maxillary sinus for implant placement. BMPs are essential components of tissue engineering. This article focuses on history of isolation and purification of BMPs, mechanism of action, carriers and various clinical applications of BMPs.

Index Terms- Bone Morphogenetic Proteins, Periodontal Regeneration, Tissue engineering.

I. INTRODUCTION

Extracellular matrix (ECM) contains 'morphogenetic factors' apart from growth factors which are capable of inducing de-novo bone formation. (Urist, 1965,Reddi)[1] 'Inductive interactions' between these morphogenetic proteins and target cell leads to sequential biochemical and morphogenetic events resulting in differentiation of endochondral bone.

BMPs were acknowledged to the world when Marshall Urist implanted demineralized bone matrix intra-muscularly into the rodents and observed new bone formation. Urist[2] named them "bone morphogenetic protein" believing that they are capable of determining morphogenesis of structures such as bone. Subsequently he was able to isolate the protein responsible for inducing new bone formation. However isolation and purification of BMPs were laborious tasks [Wang et al., 1988] [3,4] until bovine bone sequencing was done by Wozney et al and highly purified form was extracted by Sampath et al in 1980s.[7]

II. BONE MORPHOGENETIC PROTEINS IN PERIODONTICS

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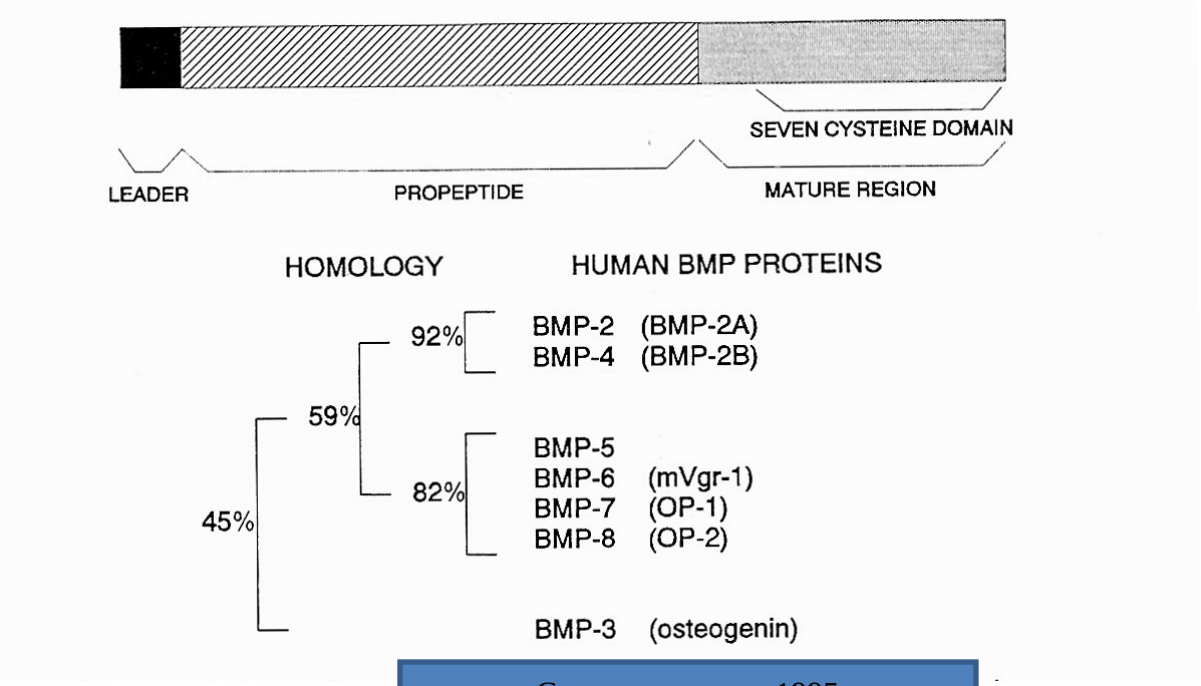
III. STRUCTURE OF BMPS

There are about 20 BMPs identified till now. They belong to Transforming Growth Factor β superfamily except BMP1 which is a metalloproteinase.[4]

The basic structure of a BMP protein has :

- hydrophobic secretory leader sequence,
- large propeptide region, and
- mature domain.



Courtesy: wozney 1995

BMPs are divided into 3 groups:

- BMP- 2 and BMP-4 –has similar seven-cysteine domains but varies in amino-terminal regions- share 92% of homology.
- BMP-5, BMP-6, BMP-7,BMP-8 share sequence homology

BMP-7 is OP-1osteoprotegrin 1 and BMP-8 is OP-1osteoprotegrin 2

- BMP-3 (osteogenin), differs from these two subgroups, form a different entity.

Recombinant human BMP-2 (rhBMP-2) has been produced using a Chinese hamster ovary (CHO) cell expression system.

- rhBMP-2 causes bone formation by intramembranous as well as the endochondral method. Other BMPs, including BMP-4, BMP-5, BMP-6, and BMP-7, also induce bone in a similar manner although there could be variation in the amount and rate of bone and cartilage formation.

IV. ROLE OF BMPS IN EMBRYO DEVELOPMENT

The bone inducing property of BMPs in extra-skeletal tissues gives clue regarding their involvement in embryonic development and also in post-natal bone differentiation (RIPOMANTI 1992) [8]. BMP2 was localised in mouse embryo at condensing precartilagenous mesenchyme, and in developing bones thus indicating it could also regulate cartilage and bone formation. Similarly, Osteogenin was also observed in rat embryo. BMP-2A was found localized in developing mouse hair follicles, limb buds, tooth buds – including the dental papilla and the odontoblastic layer, and in the mesenchyme of craniofacial region including Meckel's and nasal cartilage and that of the palatal shelves. Thus BMPs are believed to regulate embryogenesis.

V. BONE INDUCING PROPERTY OF BMPS

Subcutaneous implantation of demineralized bone matrix leads to endochondral bone formation similar to embryonic bone development. The sequential developmental cascade includes

1. Activation and migration of undifferentiated mesenchymal cells by chemotaxis;
2. anchorage-dependent cell attachment to the matrix via fibronectin;
3. mitosis and proliferation of mesenchymal cells;
4. differentiation of cartilage;
5. mineralization of the cartilage;
6. vascular invasion and chondrolysis;
7. differentiation of osteoblasts and deposition of bone matrix;
8. Mineralization of bone
9. Differentiation of hemopoietic marrow in the newly developed ossicle.

VI. MECHANISM OF ACTION OF BMPS

The ability of rhBMPs to induce intramembranous bone formation without endochondral formation has created interest in role of BMPs in periodontal regeneration. Since periodontal

regeneration also involves regeneration of periodontal ligament and bone, the ability of BMPs to stimulate cementum formation is believed to be similar to that of its bone-forming ability [8] Cementoblast and osteoblast share similar progenitor cells and rhBMP2 is expressed just before the initiation of tooth development during cementogenesis has opened way for research for the possibility of regeneration of periodontium with BMPs.

BMPs can stimulate the following cells to regenerate periodontium

- Residual cells in periodontal ligament
- Blood clot in the wound of periodontal wound
- Adjacent endosteal spaces and beyond the defect.

Two pathways may be hypothesized to reestablish tissue relationship in the periodontium following periodontal reconstructive surgery: [9]

1) growth and migration of already differentiated cells into the wound site from existing tissue resources (i.e., alveolar bone and periodontal ligament),
2) growth and subsequent differentiation of pluripotent progenitor cells (mesenchymal stem cells).

rhBMP-2 has been demonstrated to induce endochondral ossification through differentiation of mesenchymal cells into cartilage and bone cells. This, however, may only in part explain periodontal regeneration following surgical implantation of rhBMP-2. Possibly, when the healing sequence is initiated by rhBMP-2, other tissue specific cytokines and growth factors may in turn support differentiation of mesenchymal stem cells into additional periodontal phenotypes.
Due to high osteogenic potential rhBMP-2/ACS (Absorbable Collagen Sponge) was tried by many authors for better bone regeneration.

VII. CARRIERS

Carriers play an important part in bone induction by contributing to the following functions :[10]

- Localisation and retention of BMPs at the site of application (reduces the dosage)
- Providing a matrix for mesenchymal cell infilteration
- Providing substrate for cell growth and differentiation
- Shapes the new bone formation
- Degradation rate that does not inhibit bone growth and remodelling [4]

The ideal scaffold should not only deliver BMP-2, but also have the following characteristics: [11] It should be

- non-immunogenic,
- biocompatible,
- be biodegradable,
- present adhesion for cell ligands;
- contain affinity sites for growth factor (GF) binding;
- permit the integration of the newly formed bone with native surrounding tissue; and
- to fill the defect;

Components of ECM may act as carrier for collagenous matrix but due to potential problems of antigenicity and viral contamination, there is a constant search for carrier materials.

Carriers can be

- ✓ Solid xenogenic (HA)
- ✓ Solid alloplastic (polyethylene polymers)
- ✓ Gels of
  - o autogenous/
  - o allogenic
  - o alloplastic origin
- ✓ Combinations of the above.

VIII. RELEASE KINETICS OF BMPS

Release kinetics is important for example, a study by Talwar et al have shown that rapid release of BMPs resulted in bone formation and slow release promotes cementum formation [12].
By affecting the degradation rate of carrier, its release kinetics could be altered. Resorbable carrier matrices have an unpredictable degradation rate. Regeneration may be limited since earlier resorption leads to premature obliteration of space. In case of non-resorbable carriers such as methacrylate/tetrahydrofurfuryl methacrylate (PEM/THFM), amount and duration of release can be altered by adjusting the preparation method. They have been observed to have an initial rapid relief followed by a slow release and resiliency in maintaining the space necessary for proliferation and differentiation of osteogenic cells. However they necessitate a second surgery for removal. [13]
Release kinetics could be altered through

1. Chemical method- for example gelatin carrier is altered by cross-linking with glutraldehyde.
2. Magnetic field,
3. Ultrasound
4. Emission of photons.

IX. CLINICAL APPLICATIONS

BMPs are of tremendous interest as therapeutic agents for healing bone fractures, including non-union and in open tibial fracture.[4] Also used in spinal fusion and reported to prevent osteoporosis.

In dentistry, it is used for augmentation of maxillary sinus floor and alveolar ridge. BMPs may provide a promising alternative to traditional grafting procedures. Its scope further extends in treating periodontal bone defects and in implant placement along with alloplastic materials, root coverage procedures and in periodontal regeneration.

The combination of BMP2/ACS is commercially available as INFUSEVR bone graft (Medtronic, Minneapolis, MN), is useful for sinus lifting and implant dentistry.

Potential of BMP-2 helps in regenerating bone in irradiated tissues seems promising in rehabilitating patients who have undergone radiation therapy and need bone reconstruction.

X. BMPS IN TISSUE ENGINEERING

Tissue engineering aims to reconstruct lost tissues or organs and is considered as the ultimate regenerative technique. Using tissue engineering, unwanted reaction which arise due to grafts such as tissue biocompatibility or rejection could be avoided.

With the help of tissue engineering, therapies such as the production of skin to treat burns, bone grafts, arteries to treat atherosclerotic vascular disease and cartilage for plastic and reconstructive surgeries have been achieved. Tissue engineering is being applied in dentistry for the regeneration of temporo-mandibular joint, periodontal ligament, dentin, enamel, pulp and integrated tooth tissues.

Tissue engineering has three key features namely

- Cells
- Scaffolds
- Signaling molecules such as growth factors

Cells synthesize the matrix essential for the new tissue. Scaffolds provide the environment for the cells to synthesize matrix. Growth factors facilitate and promote this action. [13]

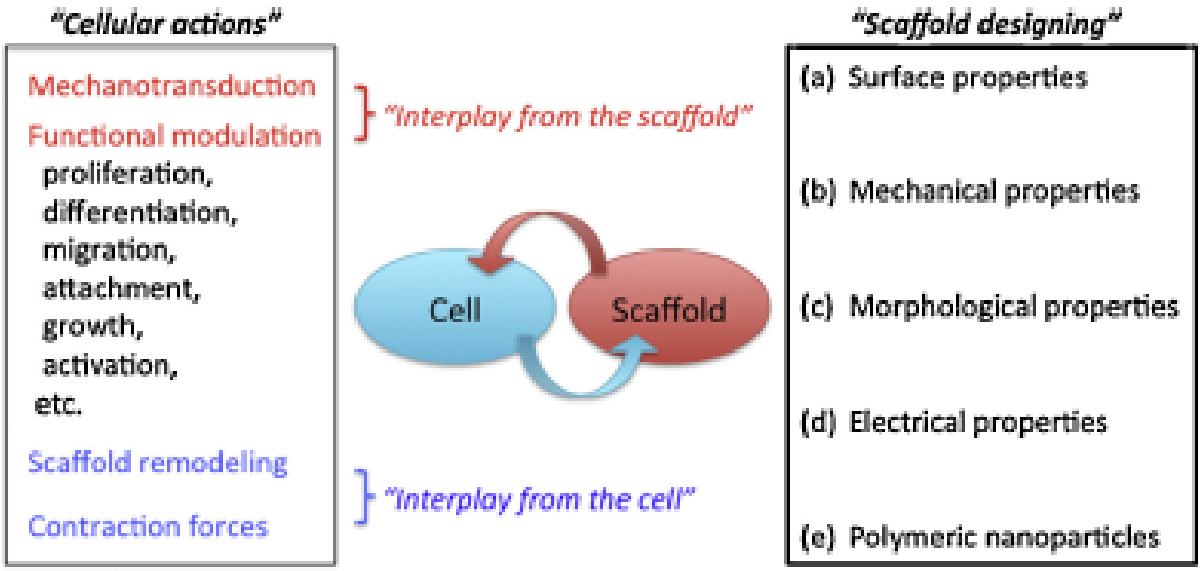


Figure 3 Cell–scaffold interplay and the components of scaffold designing.

Courtesy: Eiji Nemoto Japanese Dental Science Review

The growth factors that have frequently been applied to tissue engineering include bone morphogenetic proteins (BMPs), basic fibroblast growth factor (bFGF or FGF-2), vascular epithelial growth factor and transforming growth factor-b (TGF-b).

The BMP/TGF-b signaling pathway mediates osteoblastic differentiation and in vivo bone formation; BMP-2 and -7 were reported to the play a role in the differentiation of periodontal ligament stem cells (PDLSC) and dental follicle stem cells. Reparative dentin formation was promoted by BMP-2 and 7. Other members of the BMP family, such as BMP-7/OP-1 have observed periodontal regeneration in animal model. [8]

XI. BMPS IN SOCKET AUGMENTATION

Bmps when used in augmentation of socket and maxillary sinus wall [18] were found to promote soft-tissue healing, minimize surgery time, reduce potential postsurgical infection, accelerate cell migration and promotes early bone formation.

XII. BMPS IN IMPLANTOLOGY

Application of BMPs for the osseointegration of Endosseous implant has been evaluated by some authors [15]. Osseo-integration is critical for endosseous implant in which there is complete union of implant with bone. Sometimes there would be insufficiency in quality or amount of bone, which is addressed by using grafts or growth factors.

In human trial studies conducted by Howell in 1997 and Cochran et al in 2000 using Recombinant human BMP-2 in collagen sponge carrier, bone formation at the extracted site was observed, which helped in endosseous implant placement. [15,16]

Boyne et al in 1997, observed bone formation in sinus lift procedure using the same combination and this aided in implant placement (Boyne PJ). A feasibility study evaluating rhBMP-2/absorbable collagen sponge for maxillary sinus floor augmentation [17]

XIII. BMPS USED ALONG WITH DISTRACTION OSTEOGENESIS (DO)

Rachmiel et al I 2006, evaluated the effect of rhBMP in distraction osteogenesis in sheep model. 1.5 mm distraction devices were placed following alveolar segmental osteotomy in sheeps. [19] 5 days later rhBMP was injected. Radiographic analysis showed lifting of the transported segment and union of the distracted segment, newly formed bone and the native bone. Thus BMPs when used in the process of distraction osteogenesis seemed to minimise the consolidation period, allowing early placement of implants.

BMP implanted at the distraction site, may induce the noncommitted mesenchymal cells to form cells of osteoblastic or chondroblastic lineage. Thus to reduce the consolidation phase and improve quality of bone, BMPs can be used in DO procedure.

XIV. LIMITATIONS

Though BMPs are potential candidates for promotion of regeneration of periodontium, [20] limitations do exist. Ankylosis of varying degrees has been observed in some studies. Still there are difficulties in the method of delivering BMPs to the target site to achieve a constant sully of BMPs. Thus gene therapy and other modes of targeted delivery are being developed. BMPs are also associated with some systemic toxicity

XV. CONCLUSION

BMPs apart from inducing new bone also seem applicable in multiple modes of regeneration like being used along with implants and distraction osteogenesis procedures .

Though the discovery and usage of BMPs are like the ‘light at the end of tunnel’ towards periodontal regeneration, more experimental studies and research are needed to fully tap their potential for regeneration.

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