Interaction between Bone Cells in Bone Remodelling

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1. Introduction

Bone is a rigid mineralized structure providing protection to vital organs, acts as a habitat for bone marrow and hematopoiesis, maintains acid – base balance and facilitates body movements. Bone is the mineral reservoir of calcium, phosphate, etc.

Based on the pattern of collagen formation the bone can be classified as woven bone (irregularly organized collagen fibres) and the lamellar bone (alternating orientation of collagen fibres with significant mechanical strength). Osteoblast, osteoclast, lining cells and osteocytes are the bone cells involved in bone remodelling.

2. Osteoblast

Osteoblast, commonly known as bone forming cells that are formed from the osteoprogenitor cells (mesenchymal stem cells). The osteoprogenitor cells undergo WNT (calcium) pathway or Bone Morphogenic Protein (BMP) pathway for the formation and differentiation of osteoblast. The mature osteoblasts are plump, cuboidal cells or flattened cells bearing the keys for the lock present in osteoclast precursors.

The primary function of osteoblast is bone matrix formation. Osteoblast secretes products such as type I and type V collagen, proteoglycans and non-collagenous proteins such as sialoprotein and osteopontin.

Osteoblasts bear receptors for the hormones like Parathormone (PTH)
Parathormone releasing protein (PTHrP) 1,2 dihydroxy cholecalciferol (VIT – D) Calcitonin
Estrogen
Glucocorticoids.

Osteoblast secretes enzymes like alkaline phosphatase, pyrophosphatase etc. The interaction between osteoblast and hormones plays a major role in the formation and inhibition of osteoclast. Alkaline phosphatase is called

Abstract

Bone forms the basic framework of the body and it consists of bone cells, ground substance and collagen fibres. The bone undergoes alternate deposition and resorption to withstand the biomechanical forces exerted. Osteoclasts, osteoblasts, osteocytes secrete numerous enzymes, cytokines, growth factors which interact among one another to perform the physiological activities taking place in bone. This article emphasizes on the secretions of osteoblast that interacts with the osteoclast precursor involved in osteoclastic stimulation and inhibition and vice versa osteoclastic activity on bone can trigger osteoblastic formation or inhibition. This interaction is explained based on 3 different modes: Direct cell-cell contact, gap junctions, diffusion of paracrine factors and the 3 phases of bone remodelling.

Keywords:
the osteoblast biomarker\(^5\). The role of osteoblasts such as bone formation, secretion of various enzymes, growth factors and regulations of osteoclasts comes to an end within the life span of 3 months, but still osteocytes (one of the end product of osteoblast and major component of bone architecture) have minor potential in secretion of certain proteins\(^4,5\).

Normally in lamellar bone the collagen fibres are arranged in well organized manner but in some conditions like fibrous dysplasia, callus formation in bone fracture, hyperparathyroidism etc., the collagen fibres are arranged irregularly in a basket like weave array manner. The mutations of growth factors are potential enough to cause multiple disorders. For eg: fibroblast growth factors are potential in altering the craniofacial skeleton such as Aperts Syndrome, Achondroplasia, Crouzons Syndrome\(^6\).

After the deposition of bone matrix the osteoblast has 4 fates: a) Become an osteocyte, b) Transformation into lining cells, c) Transdifferentiate into cells that deposit chondroid bone d) Undergo apoptosis\(^5\).

### 3. Osteocytes

Osteocytes are derived from osteoprogenitor osteocytes and it comprises the major cell count of bone. Osteocytes are relatively inert in nature. Osteocytes are stellate shaped with multiple cell process (40-60)\(^7\). These cell processes connect with adjacent osteocytes and osteoblasts and provide nutrition to bone and exchange of waste through gap junction thus maintaining the vitality of bone. Osteocytes help in transmission of signal over long distance. In the conversion of osteoblast to osteocyte three intermediate stages are seen. They are,

- Type I preosteocyte.
- Type II preosteocyte\(^4\).
- Type III preosteocyte\(^4\).

Osteocytes are said to be relatively inert because during the conversion of osteoblast to osteocyte, volume of osteoblast reduces by 70% .When osteocytes are destroyed bone resorption is induced and bone formation is decreased\(^8,9\). Inhibition of bone formation by osteocytes is done with the help of a protein called sclerostin. Sclerostin is inhibited by parathyroid hormone\(^5\).

Fate of osteocytes can occur as a result of apoptosis, necrosis, senescence or osteoclastic engulfment. Excess apoptosis can cause osteoporosis, osteoarthritis\(^8,10\).

### 4. Lining Cells

Lining cells are inactive osteoblasts that are flattened. These cells have 3 major functions:

- Protects the bone from chemical substances that can eat away bone.
- Acts as an immediate access to calcium when blood calcium level is low.
- Helps in maintenance of bone fluids\(^23\).

### 5. Osteoclast

Osteoclasts are multinucleated giant cells which take part both in physiological as well as pathological resorption of bone. Osteoclast originates from the hematopoietic stem cells. Macrophage is believed to give rise for osteoclasts with the help of monocyte macrophage colony stimulating factor (M-CSF) and RANKL (Receptor Activator Of Nuclear Factor Kappa-B Ligand)\(^1\).

Osteoclasts are larger in dimension with multiple nuclei, abundant mitochondria and extensive golgi
bodies. The osteoclasts can be functional or nonfunctional. The functional osteoclasts are polarized with three distinct domains such as ruffled border, functional secretory domain and a basolateral membrane whereas the non-functional osteoclast lacks polarization and different domains.

Osteoclasts also contain Tartrate Resistant Acid Phosphatase (TRAP), calcitonin, vitronectin receptor and vacuolar proton Adenosine Triphosphatase. Osteoclast exhibits terminal differentiation (i.e.,) unable to differentiate back to macrophage or any other cell.

Osteoclasts have unique machinery for dissolving mineral within bone matrix. Osteoclast reaches the bone resorption site and secretes numerous integrin receptors like \( \alpha v\beta 1, \alpha 2\beta 1, \alpha v\beta 3, \alpha v\beta 5 \) which helps in cell to cell interaction and cell matrix migration. These integrin receptors are also involved in leukocyte activation and aggregation of tumour cells. Once osteoclast determines the site of activation, it binds to bone, gets polarized and forms ruffled borders which increases the surface area for resorption. The vitronectin part of the osteoclast gets attached to the bone and is called the sealing zone. The resorbed area is irregular which forms the resorption lacunae.

The study conducted by Lakkakorpi et al 1991 failed to demonstrate the presence of sealing zone whereas Vannen and Horton in 1995 hypothesized the plasma membrane attached tightly to the bone by sealing zone.

The main function of osteoclast is resorption of mineralized bone matrix by dissolution of hydroxyapatite crystal and cleavage of organic matrix. Before the collagen rich organic matrix gets degraded, the hydroxyapatite crystal has to be degraded. Most accepted way of dissolution of mineral occurs by secretion of HCl through ruffled borders. Area of low pH seen in the resorption lacunae is attained by ATP proton pumps. The number of intracellular acidic compartments decreases as the vesicles containing proton pumps are transported to the ruffled borders.

Osteoclast secretes several proteolytic enzymes like lysosomal cysteine proteinases, matrixmetalloproteinase, cathepsin K etc., that degrades the organic matrix in the resorption lacunae. Matrix metalloproteinase is a key player in the resorption of organic matrix. The role of Cathepsin K in the resorption process is explained with the study on osteopetrosic mice. Cathepsin K is a serine theorine protease that can sever type I collagen the major component of bone matrix.

6. Mechanism of Interaction between Osteoblast and Osteoclast can be, Direct cell - Cell Contact

The intercellular signaling occurs through membrane bound ligands. Direct cell to cell contact occurs through RANK Ligand/RANK interaction and Ephrin signaling.

- RANK Ligand/RANK Interaction:
- RANK Ligand interacts with RANK on the surface of osteoclast precursors for differentiation of osteoclast.
- Ephrin/ Eph signaling: It is a bidirectional signaling between osteoblast and osteoclasts.

Ephrin B2, a ligand expressed by osteoclast precursor suppresses osteoclast differentiation.

Ephrin B4 in osteoblastic cells promote osteoblast differentiation.

7. Gap junctions

Gap junctions permit small water soluble molecules between osteoblast and osteoclast.

8. Paracrine signaling

This type of communication occurs through growth factors, cytokines, chemokines and other small molecules.

Paracrine cell signaling occurs at all stages throughout the process of remodeling via various signaling molecules. Signals from osteoblasts activate osteoclast. For example PTH receptors are present in osteoblast but not in osteoclast. PTH induced osteoclastic bone resorption does not occur in the absence of osteoblast. In low physiological concentration of parathormone and vitamin D3 bone formation occurs whereas in high concentrations it enhance bone resorption. The action of osteoblast and osteoclast is controlled by numerous numbers of cytokines, enzymes, hormone in regulating the bone remodeling.

9. Bone Remodeling

Mechanisms involved in bone remodeling explain the interaction between osteoblast and osteoclast. Bone remodeling occurs in three phases; initiation, transition, and termination.
| Proteins | Type I collagen | Stimulates osteoblast differentiation |
|----------|----------------|-------------------------------------|
|          | Type V collagen | Stimulates osteoblast differentiation |
|          | Proteoglycans   | Stimulates osteoblast differentiation |
|          | Non-collagenous protein-sialoprotein, osteopontin | Inhibits osteoclast activity |
| Cytokines | Interleukin 1   | Stimulate bone resorption |
|          | Interleukin 6   | Stimulate bone resorption |
|          | Interleukin 8   | Stimulate bone resorption |
|          | Interleukin 11  | Stimulate bone resorption |
|          | Interleukin 4,13| Stimulates bone formation |
|          | Tumour necrosis factor (alfa) | Stimulate bone resorption |
|          | RANKLigand      | Stimulates osteoclast differentiation |
|          | Osteoprotegrin  | Inhibits bone resorption |
|          | Bone morphogenic protein | Stimulates bone formation |
|          | Leukotrienes    | Regulates bone resorption |
|          | Prostaglandins  | Stimulate bone resorption |
|          | Monocyte        | Stimulates bone formation |
|          | Monocyte chemoattractant protein-1 | Stimulates bone formation |
|          | Insulin growth factors | Stimulates bone formation |
|          | Transforming growth factor | Stimulates bone formation |
|          | Platelet derived growth factor | Stimulates protein synthesis |
|          | Fibroblast growth factor | Maturation of bone matrix |
|          | Cathepsin K     | Stimulates bone resorption |
|          | Ephrin B2       | Suppresses osteoclast differentiation |
|          | OCIL            | Inhibits osteoblast differentiation |
|          | M-CSF           | Stimulates osteoclast differentiation |
10. **Initiation Phase**

Initiation phase includes beginning of osteoclastogenesis. Interaction between precursors of osteoclast and the osteoblast cell lineages initiate osteoclastogenesis. Bone lining cells express RANKL & stimulate RANK on osteoclast precursors. Osteocyte determines the surface of bone to be resorbed by osteoclasts. Osteoblasts express RANKL and stimulate osteoclastogenic cascade. It also produces M-CSF required for survival of osteoclast lineage cells. Cell migration and cytoskeletal reorganization in macrophages and osteoclasts are controlled by M-CSF. The osteoclastic precursor cell expresses c-Fms, tyrosine kinase receptor for M-CSF. Other inflammatory cytokines like Interleukin 1 Beta, Tumor Necrosis Factor Alfa, Monocyte Chemo attractant Protein Stromal Cell Derived Protein12.

11. **Transition Phase**

Osteoblastic bone resorption liberates growth factors from bone matrix and activates osteoblastic bone formation. Calcium released from bone during resorption results in apoptosis of osteoclasts. Coupling is an interesting process that takes place in Bone Multicellular Unit (BMU) that determines the transition from bone resorption to bone formation at the cellular level. There are two types of coupling factors; Liberated coupling factors and Secreted coupling factors ( usha kini et al). Based on natalie et al., coupling factors are of four types: a) Matrix developed factors b) Secreted by osteoclasts c) Expressed by osteoclast d) Topographical changes effected by the osteoclast on the bone surface. The membrane bound molecules produced by osteoclast acts on osteoblast precursors to stimulate bone formation. Connexin mediates the gap junction communication between osteoblasts that stimulates osteoblast differentiation and bone formation.

12. **Matrix Derived Factor**

Bone matrix contains Transforming Growth Factor Beta, Insulin Growth Factor I, II, Platelet Derived Growth Factor, Fibroblast Growth Factor, Tumour Necrosis Factor Alfa, Epidermal Growth Factor etc., which are released from the bone matrix due to resorption (osteoclastic activity). Thus once released remains in the bone micro environment for 5-8 weeks and these factors are readily available for bone formation.

13. **Osteoclast Secreted Factors**

Osteoclast secretes products to motivate osteoblast precursor cells to differentiate into osteoblast. Based on the study conducted on mice the secretions are cardiotoxin-1, sphingosine-1- phosphate, Wnt 10b, Bone Morphogenic Protein -6 , and complement factor 3a (C3a), cathepsin K inhibitors. These coupling factors are produced by both functional and nonfunctional osteoclasts are also called as anti-resorptive inhibitors. Osteoclast secreted coupling factors like cathepsin K inhibitors are important clinically.
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14. Osteoclast Membrane Bound Factors

Osteoclast Inhibitory Lectin (OCIL) is a type II transmembrane c-type lectin that suppresses osteoclast differentiation. Few authors have suggested that osteoclast interacts directly through cell surface regulatory proteins to promote mature osteoblast activity. EphrinB2 and Semaphorin D are factors proposed for cell to cell contact. Based on the in vitro studies the cell contact dependent mechanisms are considered difficult in bone multicellular unit. If the cell to cell contact takes place it is found in the bone marrow or in the remodeling canopy (an anatomical structure observed above the BMU).

15. Termination Phase

During termination phase osteoblastic bone formation continues much longer than resorption. Osteoclastic differentiation is suppressed and bone formation is enhanced. Osteoprotegrin is produced by osteoblasts prevents the interaction of RANK with RANK Ligand. Osteoprotegrin activity on osteoclast precursors (RANK) inhibits osteoclastic formation and when osteoprotegrin attacks the mature osteoclast they undergo apoptosis.

Sclerostin is another protein synthesized by osteocytes that suppresses osteoblastic bone formation.

16. Conclusion

Understanding the concept behind osteoblast and osteoclast interaction provides deep understanding of bone pathology, also in treatment and prognosis. The complex interaction of the growth factors, cytokines, enzymes, hormones has significant role in dental therapy. On a gross evaluation of all pathologies of bone reflects an imbalance between osteoblastic and osteoclastic activity. The quantitative and qualitative assessment of bone can be made through the interaction between osteoblast and osteoclast. This is achieved through pathological or laboratory analysis.

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