The Mechanisms of Osteoclastic Activity: Literature Review

Os Mecanismos da Atividade Osteoclástica: Revisão da Literatura

Ângelo Barbosa de Resende[1], Luciano Barreto Silva[1], Carolina dos Santos Guimarães[2], Rosana Maria Coelho Travassos[3], Ana Paula Veras Sobral[4]

Abstract

Bone tissues are continually being remodeled depending on the organic situation, with hard tissue resorption and neoformation. Homeostasis and inflammatory conditions concerning bone may bring about the activation of hematopoietic cells with the ability to generate or dissolve bone matrix for a number of organic needs. The mechanisms used by osteoclasts to resorb hard tissues and the basic cell to cell interactions, with the participation of immune cells such as lymphocytes, along with other substances, as well as bone composition are described in this review.

Keywords: Bone tissue; T-lymphocytes; Bone matrix.

Resumo

O tecido ósseo é continuamente remodelado dependendo da situação orgânica, com reabsorção dos tecidos duros e neoformação óssea. A homeostasia e condições inflamatórias envolvendo o osso podem ativar células hematopoiéticas com a capacidade de gerar ou dissolver a matriz óssea para uma grande variedade de necessidades orgânicas. Os mecanismos usados pelos osteoclastos para reabsorver tecidos duros e as interações básicas intercelulares, com a participação de células imunológicas tais como linfócitos, junto com outras substâncias, bem como a composição óssea são descritas nesta revisão.

Palavras-chave: Tecido ósseo; Linfócitos T; Matriz óssea.

[1] Aluno do mestrado, Faculdade de Odontologia de Pernambuco, Universidade de Pernambuco (UPE)

[2] Aluno do doutorado, Faculdade de Odontologia de Pernambuco (UPE)

[3] Professor Adjunto, Disciplina de Endodontia, Faculdade de Odontologia de Pernambuco , Doutor (UPE)

[4] Professor Adjunto, Disciplina de Patologia Bucal, Faculdade de Odontologia de Pernambuco, Doutor (UPE)

Correspondência: Ângelo Barbosa de Resende
Endereço: Rua Afonso Batista, 175, Apto 403, Espinheiro – CEP 52021-020, Recife – PE, Brasil.
E-mail: angeloresende@ig.com.br

Data de Submissão: 24/05/2011
Data de Aceite: 06/03/2012

Introduction

The bone composition, the mechanisms used by osteoclasts to resorb hard tissue and cell interactions during the process of resorption are described in this literature review. The sources for information on the mechanisms of osteoclastic activity were books, LILACS, MEDLINE, Cochrane Library, SciELO, BBO, PubMed databases and the bibliographical references of the works selected, searching for studies which, potentially, could be included in this review. The words used in the search for relevant studies were: ("osteoclast activity") and ("odontology"). The search was accomplished in Portuguese and in English. With the aim of producing an updated literature review, only the studies published in English, Portuguese and Spanish from 1999 to 2011 were selected.

Literature Review

For a long time researchers have been trying to comprehend the formation and degradation of hard tissues, not only with the aim of understanding the course of certain illnesses, but also to understand the growing process of the vertebrates; particularly the degradation process, for the fact that many pathologic diseases affect both osseous and dental structures. It is known that the cells responsible for the reposition of newly formed hard tissues are called osteoblasts or odontoblasts, depending on which structures they form; the former generates osseous structures and the latter, dental structures.

Osteoblasts are characterized by their morphology, ability to form bone matrix, capacity for calcification of bone matrix and high alkaline phosphatases activity on the outer surface of the cell. Another possible property is the increasing of intracellular cAMP production in the presence of parathormone (PTH). (YOUNG et al., 2005; LYRITIS; BOSCAINOS, 2001; KWON, 2010). Internal signaling systems translate many external stimuli to a narrow range of internal signals or second messengers (MARANGALOU; GHALICHI; OUCHAKI, 2009). cAMP and cGMP are two second messengers associated with bone remodeling (REITAN; RYGH, 2006). Bone cells, in response to hormonal and mechanical stimuli, produce cAMP in vivo and in vitro.

Seemingly, bone or dental degradation have been a challenge for researchers, especially as for what concerns a condition known as root resorption, a pathologic process which recognizes and destroys dental structures while trigger immune cells. The attention is turned to the osteoclasts (degradation of osseous tissues) and odontoclasts (degradation of dental tissues).

It has been reported that the composition of calcified tissue is similar to a precipitate formed from salt solutions supersaturated of calcium phosphate. Both bone and precipitated of calcium phosphate present similar x-ray diffraction patterns characteristic of the apatite structure (HORVATH, 2006). Carbonate is present in bone and in the precipitate formed from salt solutions even when the calcium carbonate solubility product is not exceeded (DALAPICULA et al., 2006). The substance which is the first to be formed both in calcification and in precipitation of the salt solution is dicalcium phosphate which can be rapidly converted to a basic calcium phosphate. Bone contains abundant amounts of transforming growth factor β (TGFβ), which includes TGFβ1, activins, inhibins, and bone morphogenetic protein (KATAGIRI; TAKAHASHI, 2002). This small polypeptide is produced by several cell types, such as fibroblasts and osteoblasts, and is deposited in the extracellular bone matrix in a latent form. The richest sources of TGFβ are platelets and bone, and it attracts monocytes and fibroblasts, and stimulates angiogenesis in vitro (VIÑALS; POUYSSÉGUR, 2001).

Collagens and proteoglycans have been considered the two major classes of extracellular matrix macromolecules in skeletal and dental tissues. An analysis of changes in these matrix constituents during cell differentiation and morphogenesis may lead to a better understanding of the biological role of these macromolecules in development. The transplantation of demineralized bone matrix from the rat diaphysis to subcutaneous sites, resulted in new endochondral bone formation accompanied by hematopoietic bone marrow differentiation in the newly formed tissue (SHOULDERS; RAINES, 2009).

The degradation and consequent removal of hard tissue are a cellular event brought about by giant multinucleated cells which seem to be formed through asynchronous fusion of mononuclear cells belonging to the macrophage lineage and originating from the hematopoietic system. These cells are named clasts and can be identified under the light microscope because of their size (50 to 100μm), their multinucleation (2 to 10 nuclei per cell), and because they occupy shallow depressions known as Howship´s lacunae (THEILL; BOYLE; PENNINGER, 2002).

Changes in the width of periodontal ligament (PDL) cause changes in cell population and increases in cellular activity during stress caused mainly by orthodontic forces. There is an apparent disruption of collagen fibers in the PDL, with evidence of cell and tissue damage. Hyalinization may appear as a consequence of such damage. The first sign of hyalinization is the presence of pyknotic nuclei in cells, followed by areas of acellularity, or cell-free zones. The resolution of the problem starts when cellular elements such as macrophages, foreign body giant cells, and osteoclasts of adjacent undamaged areas invade the necrotic tissue. These cells also resorb the underside of bone immediately adjacent to the necrotic PDL area and remove it together with the necrotic tissue. This process is known as undermining resorption (LEE; MOTURI; LEE, 2009; MABUCHI; MATSUZAKA; SHIMONO, 2002).

Before the last decade, little was known regarding the ontogeny of osteoclasts or how these cells could resorb bone. There has been doubt whether clasts could degrade both organic and inorganic phases of bone (BLAIR; ATHANASOU, 2004). Ultimate proof that the osteoclast is of myeloid ontogeny came with the capacity to generate these resorptive cells in culture of pure populations of mononuclear phagocytes (YANG et al., 2001). Therefore, when it comes to mention the clastic cells, the macrophage lineage is indirectly mentioned as well.

The osteoclast shares many characteristics with other macrophage polykaryons; however it is a unique cell. Those features distinguishing the osteoclast are expression of calcitonin receptors, the capacity to degrade bone and, in so doing, produce lacunae resorption, abundant synthesis of tartrateresistant acid phosphatase and distinct polarization, the latter, forming a unique ruffled membrane at the osteoclast-bone interface. Under the electron microscope, multinucleated osteoclasts exhibit a unique set of morphologic characteristics. Adjacent to the mineralized surface their cell membrane is thrown into a myriad of deep folds that form a brush border sometimes visible by light microscopy. Surrounding the brush border, the plasma membrane is closely apposed to the mineralized surface (within 0.2 to 0.5 nm), and the adjacent cytoplasm, devoid of cell organelles, is filled with fibrilar contractile proteins. This clear or sealing zone not only attaches the cells to the mineralized surface but also (by sealing the periphery of the brush border) isolates a microenvironment between them and the mineralized surface. The cells organelles consist of many nuclei, each surrounded by multiple Golgi complexes, an array of mitochondria and free polysomes, a rough endoplasmic reticulum, many coated transport vesicles, and numerous vacuolar structures. It has been known for years that osteoclasts are rich in acid phosphatases as well as other lysosomal enzymes. Several light microscopic investigations suggest that the osteoclast, fibroblast, foreign body giant cell, macrophage, and endothelial cells are involved in phagocytosis of collagen during resorption (PARRENO; HART, 2009).

The concentration of enzymes, however, is not associated with lysosomal structures as in most other cells. Instead, it is known that the enzymes are synthesized in the rough endoplasmic reticulum, transported to the Golgi complexes, and from there, in coated transport vesicles, moved to the brush border (where by a process of exocytosis, their release occurs into the sealed compartment adjacent to the mineralized surface). Biochemical investigations have shown that during remodeling of tissues hydrolytic enzymes (such as acid hydrolases) increase many times (MORALES; ZHAO;LEFRANCOIS, 1999; PERRENO; HART, 2009). Another recognized feature of osteoclasts is the presence of a proton pump associated with the ruffled border, pumping hydrogen ions into the sealed compartment (LUNDQUIST, 2002; GOLDBERG et al., 2011).

The sequence of resorptive events might be resumed as the following: attachment of clasts to the mineralized surface; creation of a sealed acidic environment through action of the proton pump, demineralizing and exposing the organic matrix; degradation of this exposed organic matrix to its constituent amino acids by the action of released enzymes such as acid phosphatase and cathepsin B, and; uptake of mineral ions and amino acids by the cell. The resorptive cells of dental hard tissue (odontoclasts) are similar in most respects to osteoclasts although maybe somewhat smaller. Odontoclasts possess endocytotic vesicles containing liberated apatite crystals, which suggests that demineralization in the resorptive environment is not as complete as occurs in relation to osteoclasts (TEN CATE, 2008).

The activation of clasts is not a single event and takes place in association with other cell types such as lymphocytes. The interaction of the immune cells as well as cytokines and chemokines constitutes the main defense mechanism of a host. They are some of the substances used by the immunological cells to communicate. With special interest for the interaction with clasts is the fact that only interleukin (IL)-1α has a potent capacity, as far as it is known, to increase root resorption with odontoclastic activities (RIVOLLIER et al., 2004). IL-1 is the most potent among them, which directly stimulates osteoclast function through IL-1 type 1 receptor, expressed by osteoclasts. Secretion of IL-1 is triggered by various stimuli,

including neurotransmitters, bacterial products, other cytokines, and mechanical forces (NANCI; BOSSHARDT, 2006). Its actions include attracting leukocytes and stimulating fibroblasts, cells, osteoclasts, and osteoblasts to promote bone resorption and inhibit bone formation. Osteoblasts are target cells for IL-1, which in turn conveys messages to osteoclasts to resorb bone (NANCI; BOSSHARDT, 2006). Suda et al. (1999) reported increased levels of IL-8 at PDL tension sites and proposed it to be a triggering factor for bone remodeling. As the studies concerning root resorption continued, some researchers have tried to associate its origins with an specific antigen, present in the dentin, which would trigger the immunologic system.

Other substances like prostaglandins also seem to play a role in the activation of osteoclasts. Klein-Nulend et al. (2003) suggested that prostaglandins are important mediators of mechanical stress. This finding was followed by the work of Sekhavat et al. (2002) who found an increase in osteoclast numbers after a local injection of prostaglandins into the paradental tissues of rodents. Other study showed the reduced rate of tooth movement after the administration of indomethocin, an anti-inflammatory agent and a specific inhibitor of prostaglandin synthesis (RAMOS; FURQUIM; CONSOLARO, 2005). Other studies carried out in animals have identified the role of prostaglandins (PGE1 and PGE2) in stimulating bone resorption (BRETCHER, 2006; WISE; KING, 2008). They have reported a direct action of prostaglandins on osteoclasts in increasing their numbers and their capacity to form a ruffled border and effect bone resorption. Like other bone resorbing agents, PGE2 also stimulates osteoblastic cell differentiation and new bone formation, coupling bone resorption in vitro.

A great variety of chemically different substances seem to play important roles in bone remodeling. Growth factors (platelet-derived growth factors), hormones (PTH), and interleukins as well as other cytokines with the capacity to induce PGE2 production, are able to alter bone remodeling (KALE et al., 2004). Another study evaluated the effects of prostacyclin and thromboxane A2 in orthodontic tooth movement and osteoclastic activity on rats. It was found that these analogues increase the number of multinuclear osteoclasts, osteoclastic bone resorption, and the rate of orthodontic tooth movement (GURTON et al., 2004).

A cascade of events begins when the lymphocytes recognize and activate other cellular types to differentiate in order to eliminate the “nonself” structures once they are exposed to the immunologic system. As for what concerns dentin, odontoclasts are the main cellular type recruited in root resorption. As they come from the lineage of the macrophage, they play an important role in the immune response, since of the ways for macrophages to be activated is by microbial products, such as endotoxins and cytokines from T cells, such as interferon (IFN)-γ, and by the recognition of foreign agents. When activated, they destroy microorganisms, secrate pro-inflammatory cytokines and present the antigens to T auxiliary cells. Macrophages can also acquire different morphology in the varied tissues of the body, such as Kupffer cells in the liver, alveolar macrophages in the lung and osteoclasts in the bone, or even odontoclasts to destroy dental structures (HIDALGO, 2001).

The hard tissues in the body (bone, cementum, enamel and dentin) have many similarities in their formation. They are all specialized connective tissues, and collagen (mainly type I) plays a large role in determining their structure. Although enamel is not a connective tissue and has no collagen involved in its makeup, its formation still follows many of the principles involved in the formation of hard connective tissue (YAMAAI et al., 2005). The bone mass of a mammalian organism is maintained due to the balanced activities of bone forming and bone resorbing cells. Bone marrow stromal cells and osteoblasts support osteoclastogenesis via the synthesis of receptor activator of NF-κB ligand (RANKL). RANKL binds to its cell membrane receptor (RANK) in osteoclast precursors. In the presence of macrophage colony stimulating factor (M-CSF), these events lead to differentiation of osteoclast precursors to mature and active osteoclasts. Osteoprotegerin (OPG), also produced by osteoblasts and stromal cells, is a soluble decoy receptor for RANKL. Soluble OPG blocks the effects of RANKL on osteoclast precursors by preventing its binding to RANK (KALFAS, 2001). With both substances available, the organism regulates the need of resorption or neoformation of bone. The TNF-related ligand RANKL (receptor activator of nuclear factor-Kappa ligand) and its two receptors, RANK and osteoprotegrin (OPG), have been implicated in the remodeling process. RANKL is a downstream regulator of osteoclast formation and activation, through which many hormones and cytokines produce their osteoresorptive effect. In the bone system, RANKL is expressed on osteoblast cell lineage and exerts its effect by binding the RANK receptor on osteoclast lineage cells. This binding leads to rapid differentiation of hematopoietic osteoclast precursors to mature osteoclasts (DRUGARIN et al., 2003).

Activated T lymphocytes are emerging as regulators of bone cell function, particularly in chronic inflammatory diseases associated with bone loss (KONG et al., 1999; TAKAYANAGI et al., 2000). They can synthesize RANKL and induce osteoclast formation and activity in vitro. Co-cultures of murine or human mitogen-activated T lymphocytes with osteoclast precursors result in the formation of multiple functional osteoclasts (GILLESPIE, 2007; KOTAKE et al., 2001). Nevertheless, other methods of T-cell activation can result in complete inhibition of osteoclastogenesis, because IFN-γ is secreted by activated T cells, and is a known inhibitor of osteoclastogenesis (TAKAYANAGI et al., 2000).

Chronic inflammatory conditions, such as rheumatoid arthritis, periodontal disease, and inflammatory bowel disease, are associated with decreased bone mass (COMPSTON, 2003; PEREIRA; PEREIRA, 2004). By using adoptive transfer experiments, it has been shown that T lymphocytes appear to be crucial in periapical bone loss associated with periodontal disease (BAKER 2001; TAUBMAN; KAWAI, 2001). The effects of activated T lymphocytes on osteoclastogenesis are complex, and depend on the method of T-cell activation. The inhibition of osteoclastogenesis by activated T cells involves IFN-γ (and possibly other factors), but not OPG. This information should be helpful in interpreting the results of work examining the effects of T lymphocytes on osteoclast formation.

Conclusions

The balance of bone mass in living mammalian organisms is maintained by the action of osteoblasts and osteoclasts. Although many of the biological activities of the latter have been partially understood, further studies are necessary to comprehend the fundamentals of these cells, as well as the consequences that they may bring, not only for the establishment of illnesses, but also to enhance healing.

References

BAKER, P.J. T-cell contributions to alveolar bone loss in response to oral infection with Porphyromonas gingivallis. **Acta Odontol. Scand.**, London, v. 59, no. 4, p. 222-225, Aug. 2001.

BLAIR, H.C.; ATHANASOU, N.A. Recent advances in osteoclast biology and pathological bone resorption. **Histol. Histopathol.**,Murcia, v.19, no.1, p.189-199, Jan. 2004.

BRETCHER, A.B. **Kinins:** important regulators in inflammation induced bone resorption**.** 2006. 106f. Dissertations- Department of Oral Cell Biology, Umeå University, Umeå.

COMPSTON, J. Osteoporosis in inflammatory bowel disease. **Gut**, London, v. 52, no.1, p. 63-64, Jan. 2003.

DALAPICULA, S.S. Características físico-químicas dos biomateriais utilizados em enxertias ósseas. Uma revisão crítica. **Implant News**, Sao Paulo, v. 3, n. 5, p. 487-491, out. 2006.

DRUGARIN, D. et al. RANK-RANKL/OPG molecular complex - control factors in bone remodeling. **Timisoara Med. J.**, Timisoara, v. 53, no. 3-4, p. 296-302, 2003.

GILLESPIE, M.T. Impact of cytokines and T lymphocytes upon osteoclast differentiation and function. **Arthritis Res. Ther.**, London, v. 9, no. 2, p.1-3, 2007.

GOLDBERG, M. et al. Dentin: structure, composition and mineralization**. Front. Biosci.**, Searington, N.Y., v. 3, p. 711-735, Jan. 2011.

GURTON, A.U. et al. Effects of PGI2 and TxA2 analogs and inhibitors in orthodontic tooth movement. **Angle Orthod.**, Appleton, v. 74, no. 4, p. 526-532, Aug. 2004.

HIDALGO, M.M.A. **Estudo sobre o potencial imunogênico da dentina:** uma contribuição para a etiopatogenia da reabsorção dentária. 2001. 134 f. Tese (Doutorado em Patologia Bucal) - Faculdade de Odontologia, Universidade de São Paulo. Faculdade de Odontologia de Bauru, Bauru.

HORVATH, A.L. Solubility of structurally complicated materials: II. Bone. **J. Phys. Chem. Ref. Data**, Washington, v. 35, no. 4, p.1653-1668, 2006.

KALE, S. et al. Comparison of the effects of 1,25 dihydroxycholecalciferol and prostaglandin E2 on orthodontic tooth movement. **Am. J. Orthod. Dentofacial Orthop.**, Saint Louis, v. 125, no. 5, p. 607-614, May 2004.

KALFAS, I.H. Principles of bone healing. **Neurosurg. Focus**, Charlottesville, v. 10, no. 4, p. E1, Apr. 2001.

KATAGIRI, T.; TAKAHASHI, N. Regulatory mechanisms of osteoblast and osteoclast differentiation. **Oral. Dis.**, Copenhagen, v. 8, no. 3, p. 147-159, May 2002.

KOTARE, S. et al. Activated human T cells directly osteoclastogenesis from human monocytes: possible role of T cells in bone destruction in rheumatoid arthritis patients. **Arthritis Rheum.**, Hoboken, N.J., v. 44, no. 5, p. 1003-1012, May 2001.

KLEIN-NULEND, J. et al. Microgravity and bone cell mechanosensitivity. **Adv. Space Res.**, New York, v. 32, no. 8, p. 1551-1559, 2003.

KONG, Y.Y. et al. Activated T cells regulate bone loss and joint destruction in adjuvant arthritis through osteoprotegerin ligand. **Nature**, London, v. 402, no. 6759, p. 304-309, Nov. 1999.

KWON, R.Y. et al. Primary cilium-dependent mechanosensing is mediated by adenylyl cyclase 6 and cyclic AMP in bone cells. **FASEB J.,** Bethesda, v. 24, no. 8, p. 2859-2868, Aug. 2010.

LEE, C.H.; MOTURI, V.; LEE, Y. Thixotropic property in pharmaceutical formulations. **J. Control. Release**, Amsterdam, v. 136, no. 2, p. 88-98, June 2009.

LUNDQUIST, P. Odontoblast phosphate and calcium transport in dentinogenesis. **Swed. Dent. J. Suppl.**, Malmö, no. 154, p. 1-52, 2002.

LYRITIS, G.P.; BOSCAINOS, P.J. Calcitonin effects on cartilage and fracture healing. **J. Musculoskelet. Neuronal Interact.**, Kifissia, v. 2, no. 2, p.137-142, Dec. 2001.

MABUCHI, R.; MATSUZAKA, K; SHIMONO, M. Cell proliferation and cell death in periodontal ligaments during orthodontic tooth movement. **J. Periodont. Res.**, Copenhagen, v. 37, no. 2, p.118-124, Apr. 2002.

MARANGALOU, J.H.; GHALICHI, F.; OUCHAKI, B.M. Numerical simulation of orthodontic bone remodeling. **Orthodontic Waves**, Tokyo, v. 68, no. 2, p. 64-71, June 2009.

MORALES, C.R.; ZHAO, Q.; LEFRANCOIS, S. Biogenesis of lysosomes by endocytic flow of plasma membrane. **Biocell**, Mendoza, v. 23, no. 3, p. 149-160, Dec. 1999.

NANCI, A.; BOSSHARDT, D.D. Structure of periodontal tissues in health and disease. **Periodontol. 2000**, Copenhagen , v. 40, p. 11-28, 2006.

PARRENO, J.; HART, D.A. Molecular and mechano-biology of collagen gel contraction mediated by human MG-63 cells: involvement of specific intracellular signaling pathways and the cytoskeleton. **Biochem. Cell. Biol.**, Ottawa, v. 87, no. 6, p. 895-904, Dec. 2009.

PEREIRA, I.A.; PEREIRA, R.M.R. Osteoporose e erosões ósseas focais na artrite reumatóide: da patogêneses ao tratamento. **Rev. Bras. Reumatol.**, Sao Paulo, v. 44, n. 5, p. 347-354, set./out. 2004.

RAMOS, L.V.T.; FURQUIM, L.Z.; CONSOLARO, A. A influência de medicamentos na movimentação ortodôntica: uma análise crítica da literatura. **Rev. Dent. Press Ortodont. Ortopedi. Facial**, Maringá, v.10, n.1, p.122-130, 2005.

REITAN, K.; RYGH, P. Biomechanical principles and reactions. In: GRABER, R.M.; VANARSDALL, R.L. (Ed.). **Orthodontics:** current principles and techniques. 4. ed. Saint Louis: Mosby, 2006. p. 96-192.

RIVOLLIER, A. et al. Immature dendritic cell transdifferentiation into osteoclasts: a novel pathway sustained by the rheumatoid arthritis microenvironment. **Blood**, Washington, v.104, no.13, p. 4029-4037, Dec. 2004.

SEKHAVAT, A.R. Effect of misoprostol, a prostaglandin E1 analog, on orthodontic tooth movement in rats. **Am. J. Orthod. Dentofacial. Orthop.**, St. Louis, v. 122, no. 5, p. 542-547, Nov. 2002.

SHOULDERS, M.D.; RAINES, R.T. Collagen structure and stability. **Annu. Rev. Biochem**., Palo Alto, v. 78, p. 929-958, 2009.

SUDA, T. et al. Modulation of osteoclast differentiation and function by the new members of the tumor necrosis factor receptor and ligand families. **Endocr. Rev.**, Baltimore, v. 20, no. 3, p. 345-357, June 1999.

TAKAYANAGI, H. et al. T-cell-mediated regulation of osteoclastogenesis by signalling cross-talk between RANKL and INF-γ. **Nature**, London, v. 408, no. 6821, p. 600-605, Nov. 2000.

TAUBMAN, M.A.; KAWAI, T. Involvement of T-lymphocytes in periodontal disease and in direct and indirect induction of bone resorption. **Crit. Rev. Oral Biol. Med.**, Boca Raton, v. 12, no. 2, p. 125-135, 2001.

TEN CATE, R. **Oral histology:** development, structure and function. 7th ed. St. Louis: Elsevier Mosby, 2008. 432 p.

THEILL, L.E.; BOYLE, W.J.; PENNINGER, J.M. RANK-L and RANK: T cells, bone loss, and mammalian evolution. **Annu. Rev. Immunol**., Palo Alto, v. 20, p. 795-823, 2002.

VIÑALS, F.; POUYSSÉGUR, J**.** Transforming growth factor β1 (TGF-β1) promotes endothelial cell survival during in vitro angiogenesis via an autocrine mechanism implicating TGF-α signaling. **Mol. Cell. Biol.**, Washington, v. 21, no. 21, p. 7218-7230, Nov. 2001.

WISE, G.E.; KING, G.J. Mechanisms of tooth eruption and orthodontic tooth movement. **J. Dent. Res.**, Chicago, v. 87, no. 5, p. 414-434, May 2008.

YAMAAI, T. et al. Gene expression of connective tissue growth factor (CTGF/CCN2) in calcifying tissues of normal and cbfa1-null mutant mice in late stage of embryonic development. **J. Bone. Miner. Metab.**, Tokyo, v. 23, no. 4, p. 280-288, 2005.

YANG, S. et al. A new superoxide-generating oxidase in murine osteoclasts. **J. Biol. Chem.,** Baltimore, v. 276, no. 8, p. 5452-5458, Feb. 2001.

YOUNG, N. et al. Differential regulation of osteoblast activity by th cell subsets mediated by parathyroid hormone and IFN-g. **J. Immunol.,** Baltimore, v. 175, no. 12 p. 8287-8295, Dec. 2005.