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Role of Reactive Oxygen Species On Developments Of Osteoclastogenesis In Aging

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Abstract - Osteogenesis is a process of development which is perfectly controlled by a number of extrinsic and instrinsic factors that consists of hormones, growth factors, cytokines produced in the bone marrow micro environment, due to process of molecules adhesion that mediate by the interaction of cells and cell-matrix, osteoblasts-specific signaling proteins and transcription factors (TFs). Recently, research explained that adherence osteoclasts on the bone surface and secretion of protons into an extracellular compartment between osteoclasts and bone surface together with the production of Reactive Oxygen Species (ROS) involved in the complex process of bone resorption. Superoxide role in the activation and transcription NF-κB factor is to increase osteoclastogenesis. Furthermore, it is alleged that ROS is involved in both differentiation of osteoclasts and osteoblast cell. Aging and diseases which associated with aging is a result of ROS that cause damage was reported increase with age. This review, which is dedicated to geriatric physicians, geriatric dentistry or gerodontology reviews ROS-related osteoclastogenesis in aging and as the basic of the research to determine benefit of ROS through NADPH oxidase activation in the osteopetrotic case.

Keywords— Reactive Oxygen Species, Osteogenesis, aging

INTRODUCTION

When the human is born, the bone marrow is hematopoietic (red marrow), but next it is replace by fat or yellow marrow. The bone marrow decreases approximately 10% in each decade of life. These changes greatly affect the skeletal heterogeneity. At the age of 70 years and next ages cells of the bone marrow in vertebral are only about 30% of the total. The relationship between age change with change in the composition of the marrow and bone mass has not been fully clarified. Increasing the number of adipose cells has been observed in bone marrow patients with osteoporosis [1]. The bone is a dynamic organ. Where there is cooperation between osteoblasts and osteoclasts not only on bone remodeling, but also during osteoclastogenesis. Osteoblast cells are cells derived from pluripotent stromal stem cells. Contact between stromal cells and osteoclast precursors in the bone marrow is needed to osteoclastogenesis. The understanding about the differentiation and activity of osteoclasts are obtained by analyzing a protein that is biologically a family of TNF receptor (TNFR)/TNF-like protein that is; Osteoprotegerin (OPG), Receptor Activactor of Nuclear-κB /(NF)-κB (RANK) dan Receptor Activator of Nuclear-κB Ligand (RANKL), which together perform the functions of regulation on osteoclasts. RANKL/Osteoclasts Differential Factor (ODF) and OPG is a protein produced by osteoblasts/stromal cells are key extracellular regulation of osteoclastogenesis [2, 3]. OPG is a receptor for RANKL / ODF which is expressed on the cell membrane surface of osteoblasts. Bond OPG and RANKL can obstruct osteoclastogenesis. RANKL is also a ligand for RANK, a receptor expressed on the surface membrane of osteoclast precursor cells . The bond of RANKL and RANK can increase osteoclastogenesis [4, 3, 5, 6]. Osteoclasts is one type of bone cell that is capable of destroying bone tissue by removing the bone matrix and destroying organic part of the bone. This process is called bone resorption. Osteoclasts are derived from hematopoietic stem cells (CFU-GM) that have characteristics of similar phenotype with monocytes in circulation and makrophag [7, 8]. In carrying out its functions, osteoblasts and osteoclasts have to work together. Bone building activity and bone resorbing can be said sequentially. However, there are some circumstances that lead to imbalances osteoblasts and osteoclasts [9, 10]. Increasing the number of adipose cells has been observed in bone marrow patients with osteoporosis[1]. According Soejono, C.H, their aging on the skeletal as consequences of aging in the bone marrow. The increasing age resultes in a reduction of stromal cells that can differentiate into osteoblasts and increasing the differentiation of osteoclasts. The result is a disruption in the balance of osteoclasts osteoblasts (10). Decrease of bone which turn over can also lead to sceletal abnormalities. There are severa syndrome of osteopetrosis or osteosclerosis in which bone resoption is defective because of impaired formation of osteoclast or lost osteoclast function [11].

Many kinds of cytokines can enhance the differentiation of osteoclasts in animal culture. However, there is still little which is known about the mechanisms of human osteoclast activity increasing in the aging process. An increase in the secretion of IL-6 and IL-11 at the age of 49-88 may stimulate osteoclasts in vitro. Losing another possibility mediators which associated with increased osteoclastogenesis on aging cause the increasing of osteoporosis [12, 13, 11]. In a younger person associated is found an increasing of transcription of mRNA for OPG than older people. Osteoprotegerin (OPG) is secreted by osteoblasts for inhibiting osteoclast differentiation. The decreased expression of OPG because of advancing of age can increase the possibility of osteoclastogenesis. This statement is supported by several studies that explain that the decline OPG in mice showed an induced osteoporosis which is caused by increasing of osteoclast formation and age [13].

It is generally established that aging and diseases which associates with aging can be caused by Reactive Oxygen Species (ROS) such as superoxide anion and H2O2. ROS can cause damage DNA, proteins and lipids, nuclear and mitochondrial DNA. The concentration of the breakdown of proteins, fats and DNA is reported that increased with increasing age [14, 15]. From the results of our study (Thesis) it is also found a positive correlation between osteoclasts and superoxide in mice older ages.

Garrett, et al (1990) and Steinbeck, (1994), reported that the decreasing in NBT by osteoclasts in the cultured of rat calvarial is a response from hormones parathyroid, IL I, TNF, and 1α ,25-dihydroxy vitamin D3 associated with the formation and activity of osteoclasts. It is also reported that the addition of superoxide dismutase (SOD), catalase, and a block on NBT decrease causes a decreasing in osteoclast formation. The addition of the catalyst can inhibit the formation of osteoblasts from precursor cells and inhibits bone resorption by osteoclasts in culture of calvarial mice were stimulated with 1α-25 dihydroxy vitamin D3. The research results proved that the superoxide and H2O2 role in osteoclastogenesis and osteoclast activity [13] .

Superoxide participate in the activation and the transcription of factor NF-κB on osteoclasts. NF-κB increases the transcription of genes for doing activation of osteoclasts. Besides that, the superoxide is allegea that also mediates osteoclast differentiation [17]. RANK activation results in activation of a signaling cascade, (NF) -B and nitrogen such as nuclear factor- ctivated protein kinases (MAPKs), including p38 MAPK, C-jun

N-terminal kinase signals extracellular regulated kinase through several adapter proteins and cofactors [18].

The aim of the study is to understand the role of Reactive Oxygen Species (ROS) to osteoclastogenesis in aging.

DISCUSSION

Bone is a hard connective tissue that compose the majority of the vertebrates order. It consists of organic components (cells and matrix) and inorganic components such as minerals, especially calcium phosphate (85 %) and calcium carbonate (10 %). In the bone there is soft tissue that is found in holes or depressions in the bone, which is called bone marrow . Bone and bone marrow are anatomically located side by side and has the role of the interplay between one another. Haemopoietic bone marrow containing stem cells and non haematopoietik which is the origin of bone cells (osteoclasts and osteoblasts). The bone marrow also plays a role in the regulation of osteoclastogenesis. Cellular processes of bone activity associates with bone remodeling, in this case there is a balance between bone formation and resopsi. There are interdependencies between these processes, where osteoclast do resorption activity while the osteoblast activity in the form of bone formation [19, 1]. Bone marrow contains three types of stem cells ; ie haematopoietic stem cells (HSCs) are multipotent stem cells that can reduce all types of blood cells such as myeloid (monocytes and makrophag, neutrophils, basophils, eosinophils, erythrocytes, megakaryocytes / platelets , and dendritic cells) and lymphoid (T cells , B cells , NK cells). Both types of stem cells are mesenchymal stem cells , which have the ability to differentiate into osteoblasts , chondrosit , myocytes , and several other types of cells . While the third types of stem cells is endothelial stem cells.[20, 1, 21]. Osteogenesis is a developmental process that is controlled completely by a number of extrinsic factors that consists of hormones , growth factors , cytokines that are produced in the bone marrow microenvironment, adhesion molecules that mediate cell-cell and cell-matrix interactions, specific signaling protein osteoblast- and transcription factors (TFs). These factors are required for differentiation of osteoblasts and osteoclasts[22, 23, 8].

Beside some of the factors that may affect osteogenesis as described above , other studies explain that osteogenesis is also influenced by age . Aging causes changes in status and decreasing MMSC commitment to differentiate into osteoblasts and increased its commitment to differentiate into adipocytes cells [10]. Increasing age is also associated with increased production of free radicals. Nowadays various studies using O^2 isissued by osteoclasts as an index of osteoclast function. It was alleged that the production of ROS superoxide or activity of osteoclasts play an important role in osteoclastogenesis and osteoclast activity [24, 25, 16].

At the cellular level of ROS act as second messengers in a wide variety of signal transduction . The effect is large from proliferation to growth arrest or differentiation until senescence and cell death. ROS can activate a large number of signaling pathways include; phosphoinositide 3 - kinase (PI-3K) , phospholipase C- γ1 (PLC- γ1), p53, CREB, HSF and mitogen-actived protein kinases (MAPKs), which can be classified into extracelluler signal regulated kinases (ERKs), c-JunN-terminal kinase p38 MAPK [25, 14, 26].

The Influence of Reaktive Oxygen Species on Osteoclasts

Osteoclasts is derived from the word “osteo” (from the Greek osteon, meaning bone) and “clast” (from Greek clastos, which means destroyer). Osteoclasts is one type of bone cell that is capable of destroying bone tissue by removing the bone matrix and destroy organic part of the bone. This process is called bone resorption. In the 60's, itis probably derived from the fusion of osteoclasts osteoblasts [7]. For several years the origins ors) of osteoclasts are debated. Then demonstrated that osteoclasts come from hemapoietik and has nothing to do with the stromal cells. Osteoclast cells derived from mononuclear precursors called preosteoclasts Of Quail-chick chimera research demonstrated that osteoclasts come from hemapoietik lineage is the lineage of monocytes [7, 8].

Molecular events of osteoclastogenesis begins with the bond between RANKLsekresikan by osteoblasts on RANK on the surface of osteoclast precursors. Furthermore, activation occurres TRAF6 (figure 1). This stage is the initial stage that describes the signaling cascade. Phosphorilation activates c-jun, and NFAT1 activated by dephosporylation through calcium-mediated induction of calcineurin. RANKL / RANK also affect the c-Fos through mechanisms not yet understood. NFAT1 collaborate with AP-1 proteins in Fos / June to form NFAT2 gene, result ternary complex trancriptions on osteoclast genes to reveal the mature osteoclast phenotype. [27] .

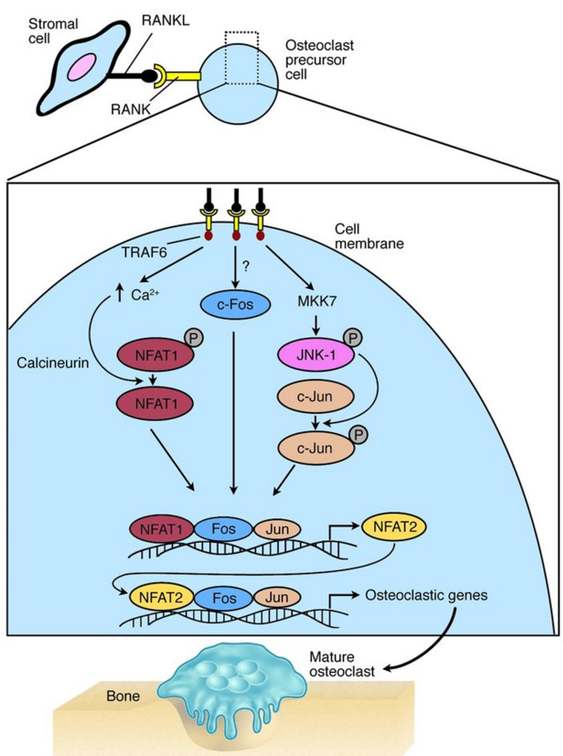

Fig. 1 Osteoclastogenesis. Osteoclastogenesis event begins with a pad bond between RANKL /RANK on the surface of osteoclast precursors, which happened TRAF6 activation is an early stage signaling cascade (27)

ROS contributes on bone resorption by osteoclasts. Strong evidence indicates that NADPH oxidase is an enzyme system responsible for the formation of superoxide on osteoclasts. Constraints on osteoclastic superoxide causes a decrease in bone function . Handling using interferon γ, a stimulator to for NADPH oxidase activity can improve the function of osteoclasts in osteopetrotic disorders. Osteoclasts express Nox2 an enzyme thought to play a role in the ROS-dependent differentiation and resorption activity. Although it is found that the mature osteoclasts whose role is NOX4 while on osteoclast precursors is Nox1 [18].

In active osteoclasts Ruffled border will form near the surface of the bone to prepare the bone resorption. Osteoclasts secrete hydrogen ions, proteinases and superoxide into Ruffled border for excavating the lacuna in the bone surface. Superoxide plays a role in the activation and the transcription factor NF-κB on

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osteoclasts. NF-κB increases the transcription of genes for activation of osteoclasts. Besides that, superoxide is also alleged mediates osteoclast differentiation [17, 28] .

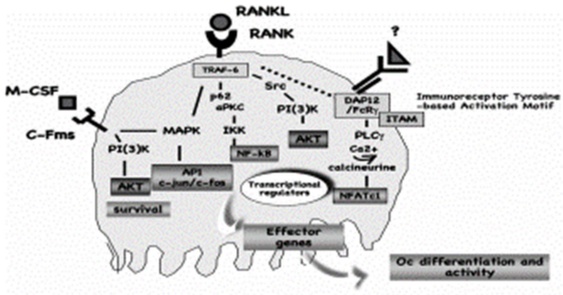

Figure 2. Signaling In Osteoklas.Diferensiasi and activation Bai Xiao-chun, 2005

Various studies have shown that ROS such as H2O2 and superoxide is involved in bone loss that is caused by the disease by stimulating osteoclast differentiation and bone resorption. NF-Bκ ligand (RANKL) osteoclastogenic important factor which is expresed by stromal cells/ osteoblasts. Although the influence of ROS on the expression of RANKL and how the effects of ROS signaling mechanism in RANKL gene have not been known yet. It was reported that the increase in intracellular levels of ROS, namely H2O2 or xanthin/ xanthine oxidase-generated superoxide anion can stimulate RANKL mRNA and protein expression of osteoblast-like MG63 human and rat [2]. Thus superoxide and H2O2 role in osteoclastogenesis and osteoclast activity. Superoxide plays a role in the activation and the transcription factor NF-κB on osteoclasts. NF-κB increases the transcription of genes for activation of osteoclasts. Besides that superoxide is also alleged mediate the differentiation of osteoclasts [17]. RANK activation results in activation of a signaling cascade (figure 2), such as nuclear factor (NF)-B and nitrogen- activated protein kinases (MAPKs), including p38 MAPK, C-jun N-terminal kinases extracellular signal-regulated kinases signal via multiple adapter proteins and cofactor [16, 18].

The Influence Of Reactive Oxygen Species On Osteoblasts

Factors that can afford to osteoblastogenesis precursor of osteoblasts is Bone Morphogenetic Protein (BMP). BMP has long been involved in skeletal development during embryonic life and fracture healing. From the research explained that BMP-2 and -4, start from the commitment of mesenchymal precursors from adult bone marrow into osteoblast lineage. BMP which is stimulates the transcription of genes encoding, that is an osteoblast-specific transcription factor, known as osteoblast-specific factor (Osf2) or core binding factor a1 (Cbfa1), which is referred to as Cbfa1/ Runx2/ PepbáA/ AML3. These transcription factors will join biological signals such as BMP/ TGFbeta and Wnt signaling pathway [22].

Wnt is an important part of bone formation. LDL-related protein 5 (LRP5) interacts with frizzled receptor's signaling transduction pathway by Wnt ligand. In the event of a deletion in LRP5 will cause osteoporosis syndrome. Osteoblast differentiation when remodeling depends on the activity of Wnt and BMP pathway. Wnt signaling requires the interaction of LPR5 and Frizzled receptor and can be inhibited by Dickkopf (DKK; an inhibitor of LPR5) and secretes Frizzled -relative Protein (sFRP). Antagonists such as sclerostin can perform well on the block BMP and Wnt signaling. Mediator in the canonical Wnt pathway that β-catenin can synergize with BMP2 to increase osteoblast differentiation and bone formation. Then Cbfa1 activate specific genes such as osteopontin, bone sialoprotein, collagen type 1 and osteocalcin. Besides Cbfa1, BMP-4 also induces homeobox-containing (Msx2) [14, 8]. Epedemiological evidence in humans and recent mechanistic studies in rodent indicated that aging and the associated increase in are the proximal culprits. ROS greatly influence the generation and survival of osteoclasts, osteoblasts, and osteocyte [29, 30]. The main function of osteoblasts on bone remodeling is forming new bone through the synthesis and secretion of non-mineral matrix proteins, proteins and polysaccharides and is responsible for the mineralization of the bone matrix. Another function but no less important is its involvement in the events of osteoclastogenesis. Precursor cells responsible for regulation of osteoclast differentiation [19, 31, 32]. Osteoblastogenesis is also affected by age. It is described on the results of these studies that osteoblasts and adipocytes cells derived from the same progenitor which mesenchymal stromal / stem cells (MMSC) derived from bone marrow. Aging causes changes in status and decreasing of MMSC commitment to differentiate into osteoblasts and increased its commitment to differentiate into adipocytes cells. Expression of osteoblast-specific transcription factor Runx2 and Dlx5 as well as markers for osteoblast decline due to aging[(33, 10, 11]. Aging is also associated with increased production of free radicals.

CONCLUSION

ROS such as H2O2 and O2 are involved in the osteoclast differentiation and bone resorption, by stimulating RANKL through ERKs and PKA-CREB pathway in rat osteoblasts and ERKs and HSF2 in humans.

ROS play a role in the activation and the transcription factor NF-κB on osteoclasts. NF- κB increases the transcription of genes for activation of osteoclasts. RANK activation results in activation of a signaling cascade, such as nuclear factor (N)-B and nitrogen- activated protein kinases (MAPKs), including p38 MAPK, C-jun N-terminal kinases extracellular signal-regulated kinases signal via multiple adapter proteins and cofactors.

ROS activated FoxO in early mesenchymal progenitors also divert β-catenin away from Wnt signaling, leading to decreased osteoblastogenesis.

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