Review

Iron accumulation leads to bone loss

Ye Yuan[1,2,3,4], Xuelong Jin[5], Youjia Xu[1,2]
1. Department of Orthopaedics, The Second Affiliated Hospital of Soochow University, 1055 Sanxiang Road, 215004, Suzhou, China.
2. Osteoprosis Institute of Soochow University, 1055 Sanxiang Road, 215004, Suzhou, China.
3. Division of Endocrinology, Diabetes, and Hypertension, Brigham and Women's Hospital, Boston, MA 02115, USA
4. Harvard Medical School, Boston, MA 02115,USA
5. Department of Physiology, Tianjin Medical University, 300070, Tianjin, China.

Corresponding author: Youjia Xu, Email: xuyoujia@suda.edu.cn

Abstract: Osteoporosis is a disease associated with bone loss and brittle fracture. Osteoporosis is common complication in thalassemia, hereditary hemochromatosis and sickle cell disease, which are combined with iron accumulation. Enhanced iron level is risk factor for postmenopausal osteoporosis. The pathological tendency of iron accumulation induced bone loss may be boost in case of estrogen deficiency. Meanwhile, osteogenesis inhibition is observed in iron accumulation environment. Iron chelator and iron homeostasis regulator hormone alleviate iron accumulation induced bone loss. Reducing iron accumulation may be a new potential target for prevention and treatment of postmenopausal osteoporosis.

Kew words: Iron accumulation; bone loss; postmenopausal osteoporosis.

Osteoporosis, or bone loss, occurs in postmenopausal women and aged people. Osteoporosis is defined by the World Health Organization (WHO) as bone mineral density DXA T value score<-2.5[1] , resulting in increased risk of fracture. More than 40% white women aged over 50 years suffer osteoporotic fracture[2]. In Japan, the estimated osteoporotic related fracture over 50 years is 37.4% of whom 41.5% with multiple fractures[3]. Accordingly, the prevalence of osteoporosis and osteoporotic fracture is major issue for public health management and health care costs. Osteoporosis patients are associated with significant worse outcome and economic burden than other chronic conditions among with hypertension, high cholesterol, and insomnia in Japan[4]. In the United State, annual cost of osteoporotic incident fractures is approximately $17 billion[5].

Bone homeostasis is maintained by osteogenesis and osteoclastogenesis. Osteoblasts and its progenitor cells mesenchymal stem cells are involved in osteogenesis, meanwhile, Osteoclasts and its progenitor cells hematopietic stem cells are invoved in osteoclastogenesis. Osteocytes, which

are differentiated from osteoblasts, have important role in bone homeostasis as well. Several known factors induced osteoporosis, including estrogen deficiency, low level of calcium and vitamin D, aged mesenchymal stem cells[6-8]. Recent studies reveal iron accumulation is risk factor of bone loss, especially in postmenopausal osteoporosis. The aim of this review is to review iron accumulation as a risk factor of bone loss.

Iron accumulation is risk factor of menopausal osteoporosis

Elevated level of serum ferritin is accompanied with estrogen deficiency in menopausal women[9], which means estrogen decreases by 90%, with a concurrent 2-3 folds serum increases in postmenopausal women (figure 1).

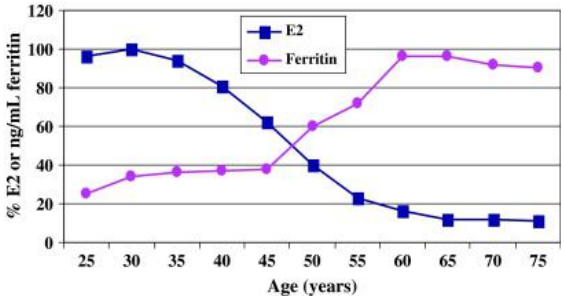

Figure1. Changes of serum ferritin versus estrogen for female. Ferritin amplifies after 45-year-olde. Estrogen decreases in postmenopausal women[9].

Clinical observation showed osteoporosis and iron metablolism markers were significant higher in older women (>75 years) compared with younger women (<45 years). Iron accumulation was risk factor of bone loss in healthy postmenopausal women. Increased serum ferritin is related to bone loss in healthy individuals[10]. In women >45 years, clinical evidence revealed serum ferritin level and low bone mineral density [11, 12]. Retrospective study showed low BMD is more dependent with ferritin level, BMI than sex, age and menopause[13]. In vivo, iron accumulation in ovariectomized rats induced osteopenia. Treated with iron chelator prevented iron accumulation in bone tissue and mitigated bone loss[14]. Iron accumulation induced bone loss in postmenopausal women may be related to degradation of type I collagen. In this regard, iron plays crucial role in female osteoporosis despite of sex hormones[15]. Reducing iron accumulation may be a potential target for postmenopausal osteoporosis women[16].

Osteoporosis is complication of iron accumulation disease

Thalassemia-associated bone loss and high fracture risk are main complication of thalassemia. Regular blood transfusion therapy is necessary for maintaining hemoglobin that results in iron accumulation[17]. Thalassemia patients with blood transfusion therapy have to reverse accumulated iron level by iron chelation[18]. Thalassemia induced osteoporosis is progressive in pathologic fracture of spine and back pain[19]. Long term analysis male with thalassemia had increased bone loss and prevalence of fracture risk[20]. Osteoclastogenesis was induced in thalassemia iron accumulation environment. Iron accumulation may

affect bone healthy by transient receptor potential vanilloid type 1 (TRPV1) channels, influencing tartrate-resistant acid phosphatase (TRAP) expression and activity[21].

High prevalence of bone metabolism disorder was reported in patient with genetic hemochromatosis[22]. Analysis of 87 patients with hereditary hemochromatosis showed correlation between iron overload and osteoporosis. The association was independent of genetic background[23]. Severe iron overload (serum ferritin level> 1000 ug/l) in hereditary hemochromatosis was associated with not only osteoporosis but also wrist fractures and vertebral fractures[24]. Hfe-/- mice is biological model of hereditary hemochromatosis. Excess iron was observed in Hfe-/- mice, especially excess iron in bone tissue with increased hepatic iron[25]. Bone loss occurred in genetic mouse model of hemochromatosis. Osteoblasts was inhibited and bone microstructure was undermined in Hfe-/- mice[26, 27]. Iron enriched diet led Hfe-KO mice to osteoporosis. Bone formation and osteoblast number decreased in this mice model. Downregulation of bone metabolism markers and upregulation of ferritin heavy polypetide 1 and transferrin receptor 1 were observed[28].

Sickle cell disease is inherited disorder caused by mutation of HBB (Hb b-chain coding gene). Chronic red blood cell transfusion therapy resulted in severity of iron accumulation in sickle cell desease patients[29-31]. Children with sickle cell disease were associated with slight BMD decreased, being more marked in female and starting before puberty[32]. Impaired osteoblasts function was related to bone loss in sickle cell disease[33]. Reduced bone formation in transgenic sickle cell disease mice was due to reduced IGF1 and osteoblasts terminal differentiation[34]. Complication of bone loss in sickle cell disease may link to iron accumulation.

Iron accumulation leads to bone loss in vivo and vitro

Bone marrow mesenchymal stem cells (MSCs), as precusor of osteoblasts, are multiple functions cells, potentially differentiating into osteoblasts in vivo. Osteogenesis relys on MSCs function, quantity, and differentiation. Iron accumulation induced MSCs apoptosis. Activation of caspase3 is crucial for MSCs apoptosis, consequently leading to bone loss in iron accumulation mice model[35].

ROS plays an crucial role in iron accumulation induced bone loss. Iron accumulation induced preosteoblast cells apoptosis in MC3T3-E1 cells. It may be related to more ROS in cells inhibition of AKT kinase and its downstream protein activity including cleaved caspase3, caspase7 and caspase9[36]. Besides, iron accumulation induced ROS blocked the PI3K/AKT and Jak/Stat3 signal pathways to arrest MC3T3-E1 cell G1 phase and autophagy[37]. Iron dextran injection mice model displayed bone loss with cortical thinning and enhanced bone resorption. Treatment with antioxidant N-acetyl-L-cysteine reversed trabecular abnormalities, which demonstrated bone destroyed induced by ROS[38].

Moreover, iron accumulation inhibited

other osteoblastogenic markers such as Runx2, osterix, osteopotin, and osteocalcin, and had minimal effect on osteoblasts differentiation[39]. Iron accumulation also inhibited osteoblast-specific genes including Runx2a, runx2b, osteocalcin, osteopontin, ALP and collagen type I in iron accumulation zebrafish model[40]. Ferric ammonium citrate induced iron accumulation in zebra caused bone calcification decreased. Osteoblasts related mRNA expression was downregulated in iron accumulation zebra[41].

Osteoclastogenesis is enhanced in postmenopausal osteoporosis women, and the pathological tendency of iron accumulation induced osteoporosis may be boost in case of gonadal hormone deficiency. Iron accumulation significantly increased osteoclastogenesis when absent of estrogen[42]. In vitro, ferric ion induced iron accumulation amplified both RAW264.7 and bone marrow derived macrophages. ROS may be the key point in the process, which could be attenuated by antioxidant N-acetyl-L-cysteine (NAC)[43]. Iron promoted osteoclasts by stimulating ROS, sequentially activiting JNK, ERK and NF-kB signaling pathways[44].

Hepcidin, a hormone produced by liver, is crucial for iron homeostasis in vivo. Hepcidin 1 knockout mice, contributing to iron accumulation, had low bone mass and undermine bone microstructure[45]. Similarly, hepcidin deficient mice amplified C-telopetide of type I collagen (CTX-1) and reduced bone load-bearing capacity, which induced bone loss likely through enhancing bone resorption[46]. Treated with hepcidin in hFOB 1.19 osteoblasts downregulated ferroportin 1, suggested osteoblasts may be target cells for hepcidin[47].

Ferroportin(FPN) is cell membrane protein regulating trasfering intracellular and extracellular iron. Deletion of ferroportin in myeloid cells resulted in iron overload in osteoclast precursors, increased osteoclastogenesis and, of note, bone loss in mice. Increased intracellular iron stimulated expression of nuclear factor of activated T cells 1 (Nfatc1) and PPARG coactivator 1β (Pgc-1β), which is critical for osteoclast differentiation[48].

Under static magnetic fields, iron environment also cause bone disorder. Hypomagnetic field with high iron level inhibited MC3T3-E1 cells differentiation potential, as well as alkaline phosphatase (ALP) activity, mineralization and calcium deposition[49].

Iron accumulation may also have indirect role on bone. Male iron accumulation rats femoral bone mineral density decreased combined with renal dysfunction and liver iron overload syndrome[50]. Meaningfully, bone loss was found in renal failure patients treated with iron[51]. The evidence of indirect effect between iron accumulation and bone deformity need further investigation.

Treatment for iron accumulation osteoporosis

Bisphosphonates are effective and safe anti-resorption drugs used for

osteroporosis. Clinical evidence suggested bisphosphonates were able to increase BMD and serum bone turnover[52]. Icariin had the ability to protect against iron accumulation induced bone loss by reversing mitochondrial membrane potential dysfunction and ROS level[53]. Inhibitor of mTOR, Rapamycin, was able to improve bone mass in high turnover osteoporosis with iron accumulation[54]. Rapamycin had positive effect on angiogenesis, which was crucial for osteogenesis. Treated with oral chelator [1-N-docosyl-triethylenetetraminepentaacetic acid (DoTTPA)] prevented iron accumulation induced oxidative damage in skeletal tissue[55].

Hepcidin, as endogenous hormone, regulates iron metabolism. Hepcidin overexpression in ovariectomy induced or iron accumulation induced animal osteoporosis model showed that hepcidin level had negative correlation with bone loss. In addition, hepcidin inhibited iron accumulation by reversing ROS in vivo[56]. Hepcidin actived extracellular calcium transport in hFOB 1.19 osteoblast. Increasing extracellular benefited for mineralization and osteogenesis[57].

Transferrin receptor 2 (Tfr2) controls iron metabolism. Tfr2 knockout mice increased bone mass and mineralization independent of iron metabolism. Tfr2 controled bone mass by BMP and Wnt signaling, suggesting Tfr2 was a potential therapeutic target for iron disorder induced bone loss[58].

Iron binding agents, or iron chelator, might have therapeutic utility to treat iron accumulation induced bone loss. Clinical observation of oral iron chelation deferasirox seemed to be low rate of endocrine disorders in thalassemia major patients[59]. Five consecutive years of iron chelation therapy with deferasirox decreased thalassemia dedocrinopathy including osteoporosis[60]. Iron chelating properties of Eltrombopag could amelliorate bone loss in thalassemia induced osteoporosis[61]. Type H vessel, positive for CD31 and Endomucin, is age-related marker in osteoporosis. Recent evidence suggested desferrioxamine contributed to bone mass by enhancing type H vessels in osteoporosis mice model[62]. Deferoxamine is effective iron chelator. Treated with deferoxamine alleviated bone loss in iron accumulation zebrafish model, attenuating ROS level[40]. Reducing iron accumulation may be a new potential target for prevention and treatment of postmenopausal osteoporosis[16].

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