Zinc Supplements and Bone Health: The Role of the RANKL-RANK Axis as a Therapeutic Target

Amin, N., Clark, C., Taghizadeh, M. & Jafarnejad, S.

Original citation & hyperlink:
Amin, N, Clark, C, Taghizadeh, M & Jafarnejad, S 2020, 'Zinc Supplements and Bone Health: The Role of the RANKL-RANK Axis as a Therapeutic Target', Journal of Trace Elements in Medicine and Biology , vol. 57, 126417.
https://dx.doi.org/10.1016/j.jtemb.2019.126417

DOI 10.1016/j.jtemb.2019.126417
ISSN 0946-672X

Publisher: Elsevier

Zinc Supplements and Bone Health: The Role of the RANKL-RANK Axis as a Therapeutic Target

Negin Amin[1], Cain C.T. Clark[2], Mohsen Taghizadeh[1], and Sadegh Jafarnejad*[1]

1. Research Center for Biochemistry and Nutrition in Metabolic Diseases, Kashan University of Medical Sciences, Kashan, IR Iran

2. Faculty of Health and Life Sciences, Coventry University, Coventry, United Kingdom

A shortened version of the title: *Zinc Supplements and the RANKL-RANK Axis*

* Corresponding Author. Research Center for Biochemistry and Nutrition in Metabolic Diseases, Kashan University of Medical Sciences, Kashan, I.R. Iran. Tel: +98-31-55463378; Fax: +98-31-55463377. E-mail addresses: sjafarnejad@alumnus.tums.ac.ir

Abstract

Background:

To this day, empirical data suggests that zinc has important roles in matrix synthesis, bone turnover, and mineralization and its beneficial effects on bone could be mediated through different mechanisms. The influence of zinc on bone turnover could be facilitated via regulating RANKL/RANK/OPG pathway in bone tissue. Therefore, the aim of the study was to conduct a review to investigate the possible effect of the zinc mediated bone remodeling via RANKL/RANK/OPG pathway.

Methods:

A comprehensive systematic search was performed in MEDLINE/PubMed, Cochrane Library, SCOPUS, and Google Scholar to explore the studies investigating the effect of zinc as a bone remodeling factor via RANKL/RANK/OPG pathway regulation. Subsequently, the details of the pathway and the impact of zinc supplements on RANKL/RANK/OPG pathway regulation were discussed.

Results:

The pathway could play an important role in bone remodeling and any imbalance between RANKL/RANK/OPG components could lead to extreme bone resorption. Although the outcomes of some studies are equivocal, it is evident that zinc possesses protective properties against bone loss by regulating the RANKL/RANK/OPG pathway. There are several experiments where zinc supplementation resulted in upregulation of OPG expression or decreases RANKL level. However, the results of some studies oppose this.

Conclusion:

It is likely that sufficient zinc intake will elicit positive effects on bone health by RANKL/RANK/OPG regulation. Although the outcomes of a few studies are equivocal, it seems that zinc can exert the protective properties against bone loss by suppressing osteoclastogenesis via downregulation of RANKL/RANK. Additionally, there are several experiments where zinc supplementation resulted in upregulation of OPG expression. However, the results of limited studies oppose this. Therefore, aside from the positive role zinc possesses in preserving bone mass, further effects of zinc in RANKL/RANK/OPG system requires further animal/human studies.

Keywords: zinc supplement, bone health, bone remodeling, RANKL/RANK/OPG system

Introduction

The skeleton is responsible for enduring mechanical load in the body and plays some important roles in inflammation, hormonal and mineral challenges in the body. Further, it is suggested that the skeleton may influence the parenchymal function of organs such as the kidney and pancreas by secreting some agents [1]. As a connective tissue, the skeleton has 4 types of cells including osteoblasts, osteoclasts, osteocytes, and bone lining cells. Bone tissue is perpetually in turnover; where osteoblasts control the formation of bone, and osteoclast activities lead to bones resorption. [2]. Bone mass is mainly comprised of minerals in varying types and combinations, such as hydroxyapatite, proteins including collagen and noncollagenous proteins, water and etc. each of these components is influencing by gender, disease, age, and site [3]. There are many risk factors that may deleteriously impact skeleton health, including low calcium level, vitamin D deficiency, sedentary lifestyle, hyperthyroid and hypothyroid, high blood pressure, high-stress level, hysterectomy, and postmenopause [4]. Some of the disorders that

mainly involve the skeleton include osteoporosis, rickets, osteomalacia, renal osteodystrophy, Paget's disease and malignancy of bone [5]. When bone mass reduced, some harmful change appear in bone structure [6]. Further, bone health is weakened as people age, especially in developing countries [7]. In Britain, 1 in 5 men and 1 in 2 women over fifty years old, suffer from at least one fracture during their lifetime [8]. Based on recent findings, over 200 million people suffer from osteoporosis, globally [9], with most fractures occurring in the hips and forearm [10]. Although the actual effects of pharmacological agents on bone disorders is not yet clear; there is a burgeoning field of research dedicated to elucidating potential interactions. Some of the most common drugs in bone diseases include bisphosphonates, denosumab, estrogen, PTH peptide, Strontium ranelate, DKK1, and sclerostin antibodies, and calcium-sensing receptor antagonists [11]. On the other side, there are some medications that may have negative influences on bone health, including thyroid medications [12], anti-diabetic drugs [13], proton pump inhibitors [14], and antidepressants [15]. In order to preserve bone health, sufficient and persistent intake of some nutrients such as vitamin D, vitamin K, vitamin A, calcium, magnesium, fluoride, phosphorus, copper, potassium and zinc are necessary [16]. With a proper nutritional strategy, it is conceivable that reductions in bone loss, especially at early ages when the genome still has time for epigenetic changing, maybe feasible [7].

Zinc is one of the important minerals for growth and bone health preservation; whilst deficiency in zinc has multiple complications, including diarrhea, alopecia immune dysfunction, growth retardation, and cognitive impairment. Aging, bone resorption and post-menopause all act to reduce zinc quantities in bone [17-19]. Zinc is required for protein and DNA synthesis in the skeleton [20, 21], whilst zinc deficiency leads to a major reduction in collagen synthesis and turnover [21]. It is also a stimulant for osteoblastic cells and suppresses osteoclast activity [22]. Empirical data suggests that zinc has important roles in matrix synthesis, mammalian system and probably in bone mineralization [23], and its protective and beneficial effects on bone could be mediated through cell proliferation, increasing collagen production, and stimulation alkaline phosphatase activity [24]. Furthermore, influence on bone turnover could be facilitated via zinc regulating RANKL/RANK/OPG pathway in bone tissue [25]. Zinc could inhibit the osteoclastogenesis which is induced by RANKL (receptor activator of NF-KB ligand) [19]. The pathway and its components are part of the tumor necrosis factor (TNF) superfamily, which could play an important role in bone remodeling. RANKL/RANK axis control osteoclasts formation and activity [26]. Moreover, these are identifying as key indicators on bone turnover in bone-related pathological situations. OPG, as the RANKL`s decoy receptor, plays a bone

protective role by binding to RANKL and prevention of bone resorption [26]. The imbalance between RANKL and OPG could lead to extreme bone resorption [26].

Although, there are several studies depicting the beneficial effects of zinc on bone health; the current evidence is not enough to approve the bone protective role of zinc supplementation in respect to RANKL/RANK/OPG balance. Therefore, the aim of this study was to review the effect of zinc supplements on regulation of RANKL/RANK/OPG axis which will help the future studies to focus and assist in developing potent strategies for treating patients with bone disorders.

A comprehensive systematic search was performed in MEDLINE/PubMed, Cochrane Library, SCOPUS, and Google Scholar to explore the studies investigating the effect of zinc as a bone remodeling factor via RANKL/RANK/OPG pathway regulation. We comprehensively searched through the databases for in vitro and in vivo studies that investigated the effect of zinc on bone health via regulation of RANKL/RANK/OPG pathway. Additionally, we tried to find more related studies with manual reference list checking. We used both MeSH term and free-text in titles/abstracts as follow: ("Zinc"[Mesh] OR "Zinc supplement*") AND (""Bone and Bones"[Mesh]) AND ("RANK Ligand"[Mesh] OR "RANKL" OR "RANK" OR "Osteoprotegrin" OR "OPG". We did not restrict the searches according to their languages or the type of study. The studies for literature review were selected if they met the following criteria: 1)

using any form of zinc as single or multi-component supplement, 2) studies in which they investigated the role of zinc in RANKL/RANK/OPG modulation regarding the bone cells. We primarily searched and identified the eligible in-vivo and in-vitro studies by electronic searching and manual searching of reference lists of eligible studies. By secondary screening, ineligible articles were excluded due to duplication in investigated databases, and being irrelevant to the purpose of the current review. Subsequently, the details of the pathway and the impact of zinc supplements on RANKL/RANK/OPG pathway regulation were discussed.

Zinc and bone health

There are multiple trace minerals which participate in bone metabolism, such as copper, magnesium, fluoride, and zinc [27]. Low level of copper and magnesium in serum can lead to depletion in BMD [28, 29]. Zinc is one of the most essential trace elements for the body [30], is an important cofactor for DNA and RNA synthesize enzymes, and is a major stimulant for growth [31], and could interfere in chemical catalysis or indirectly participate in protection of protein structure [32]. Zinc food sources include; red meat, poultry, shellfish, seeds, nuts, dairy, and beans [33], and a reported 2 billion people suffer from zinc deficiency, necessitating supplementation with zinc [34]. Zinc supplements have varying side

effects and different absorption rates based on their composition, with zinc sulfate, zinc picolinate, zinc acetate, zinc gluconate, zinc orotate, and zinc citrate are available zin supplements in market [35-37]. Zinc deficiency can result in, for example, alopecia, dermatitis, loss of appetite, immune system dysfunction, and growth retardation [38, 39]. Conversely, zinc toxicity includes issues such as nausea, vomiting, lethargy, fatigue and epigastric pain [40]. There is about 2-3 gr zinc in the human body and approximately 0.1% of this amount is excreted which and thus must be replenished through dietary intake [31]. A great amount of total body zinc esides in the skeleton [41]. It has been shown that zinc has an essential role in bone metabolism and mineralization, for instance, in osteoblasts, zinc could activate aminoacyl-tRNA synthetase and also prohibit osteoclasts from bone resorption. Zinc increases protein synthetase which helps in the protection of bone health [42]. It has been suggested that urinary zinc level may be employed as a convenient marker for bone loss recognition [43]. Zinc could result in significant upregulation in alkaline phosphatase activity which is important for bone calcification and plays an important role as a biochemical marker for bone development [44, 45], zinc finger transcription factors, TRAF6-inhibitory zinc finger protein and Schnurri-3 [46]. Aging, bone unloading and post-menopause may facilitate a reduction in bone zinc quantities [25]. Since almost 800 microgram per gram of creatinine is expelled in urine in women with osteoporosis, urinary

zinc is utilized as a marker for bone resorption in postmenopausal women [47]. It has been shown that zinc receptors are necessary for the normal matrix and cells in bone. Antecedent work has indicated that zinc and its receptor could change the expression of some enzymes and adjust collagen production in the skeleton [48]. Furthermore, zinc activity in the bone could affect both proliferation and differentiation of osteoblast-like cells [49]. Oral intake of zinc supplements could help to protect bone from resorption in various conditions, such as aluminum toxicity, Ca deficiency, vitamin D deficiency, estrogen deficiency, diabetes, and arthritis. So, administration of zinc compounds could add to bone loss protection and prevention protocols [50]. Zinc deficiency contributes to many types of abnormalities in bones in fetal and postnatal terms [50]. In young animals, where zinc deficiency is identified, they face to reduction in somatomedin (IGF-I) synthesis and growth problem [51, 52]. A zinc finger transcription factor, Osterix (Osx), is expressed in, almost, all growing bones; the genetical role of Osx in bone formation and bone homeostasis is well known and operates gene collection for differentiation of pre-osteoblasts to osteoblasts and osteocytes [53]. Zinc chelated with β-alanyl-L-histidine results in the formation of β-Alanyl-L-histidine zinc (AHZ), which can stimulate bone formation more than zinc sulfate. In addition, zinc acexamate is another compound which has identical effects to AHZ in bone formation, and is comparable with other bone regulator factors [25]. Some studies

indicate zinc may elicit positive influences on the healing of bone fractures [54, 55]. The ability of zinc to suppress osteoclastogenesis is likely because of its inhibitory effects on RANKL. Moreover, it may conceivably prevent the pre-osteoclast signaling pathway which is related to the RANK/RANKL system [50].

RANKL/RANK/OPG pathway

Nf-kB is a superfamily, where constituent proteins take part in the signaling pathways [56]. Some of the Nf-kB family members are controlled in inflammatory and neoplastic conditions that might be motivated by proinflammatory factors [57]. Also, elimination of some of these agents may leads to unwanted bone development [58]. One of the most important systems of this family is RANKL/RANK/OPG signaling pathway, is regarded an important system for immunity and is essential for bone homeostasis [59]. RANK or receptor activator of NF-kB is a transmembrane protein contain 3 subunits. It originally discovered in mature osteoblasts, dendric cells and osteoclast precursors (OCP) [60]. A deletion mutation in RANK which reported in transgenic mice identified the significance of RANK in osteoclastogenesis [61]. RANKL (receptor activator of NF-kB ligand also known as osteoclast differentiation factor) is a homotrimeric protein, part of TNF superfamily, and is encoded by TNFSF11 gene [61]. RANKL is mainly

expressed by activated T cells, osteoblasts, fibroblasts and stromal cells [62], and expressed in mammary gland epithelial cells during pregnancy, causing hyperplasia during lactation [63]; in addition, it is expressed in some tumor cells and may control their proliferation [64]. Overexpression of RANKL has been observed in various diseases such as arthritis rheumatoid and psoriatic arthritis [65, 66]. RANKL is regulatable by different agents such as glucocorticoids, TNF-a, TGF-b, IL-1 LSP. this protein has the facility to bind to both RANK and OPG

[59]. OPG (the soluble decoy receptor osteoprotegerin) encoded by TNF receptor family member 11B gene (TNFRSF11B) [67], and has been detected during an investigation designed for TNFR (tumor necrosis factor receptor) related molecules [68]. OPG mRNA has been found in many cell types, including skin, liver, heart, lung and bone marrow stromal cells [69, 70]. The main responsibility of OPG is inhibition of RANK matching with RANKL [67]; moreover, OPG could bind to TRAIL (TNF-related apoptosis-inducing ligand) to prevent TRAIL-induced apoptosis [71]. In addition to RANKL and TRAIL, OPG could bind to some other agents like glycosaminoglycans (GAGs), von Willebrand Factor, Factor VIII-von Willebrand Factor complex, and syndecan-1 [72]. The RANKL/RANK/OPG system has important roles in various organs and conditions such as bone modeling and remodeling [61], cancer cells [73], pregnancy [63] , immune system [74], and cardiovascular disease [75].

RANKL/RANK/OPG in bone

Osteoclasts are regarded the most important cells in bone resorption, where osteoclast precursors have the potential to maturate via stimulation of some factors. Among these factors, there are 2 cytokines; M-CSF, a cytokine that secreted by several types of cells like stromal cells and osteoblast, and receptor activator of NF-kB, part of RANKL/RANK/OPG system [76]. RANKL/RANK/OPG system is active in various conditions including vascular calcification [77], cancer [78], bone modeling a remodeling, and some genetic disorders [79]. The discovery of RANKL/RANK/OPG system in bone turnover in the late 1990s is regarded as an important finding for developing bone health [80].

The interaction between RANK and its ligand (RANKL), on the external layer of OPCs (osteoclast precursor cells), results in maturation of these cells and turning to osteoclasts. Osteoclasts which migrated to the bone surface, secretion of some enzyme like cathepsin K and tartrate-resistant acid phosphatase, and acceleration of bone resorption. Osteoprotegerin expressed by bone stromal cells and osteoblasts, play role as decoy receptor for RANKL and prevents the binding of

RANK and RANKL with its higher affinity to RANKL [81]. The OPG-RANKL ratio is less than 1 in postmenopausal women [49], with some studies reported that blocking RANKL leads to an increase in OPG level, and could noticeably reduce bone loss during lactation [82]. The timing of RANK expression and its link to RANKL is a serious issue in osteoclastogenesis [83]. Both RANKL and RANK must be present in order for osteoclastogenesis to happen, where, in mice unable to express RANK and RANKL, they suffered from osteopetrosis [84]. However, in mice with OPG deficiency, osteoporosis occurred earlier than normal mice, attributed to the high number of osteoclasts differentiation [85]. OPG overexpression in animal models has resulted in deep osteoporosis to the extent that a total loss in osteoblasts happen [86]. Rheumatoid arthritis is an inflammatory disease that can weaken bone in numerous ways. RANKL is one of the factors that have expected to take part in RA bone problems. Suppressing RANKL and other agents like TNF represent a promising treatment for minimizing RA Complications

[87]. Paget disease is another bone disorder with the Interference of RANKL-RANK in its development [88]. Paget disease of bone is mainly identified by central bone resorption and increased osteoclast activity. In PDB, RANKL expression and its sensitivity increases which leads to more bone problems [89]. Recent studies regarding bone cancer have investigated signaling systems in this disease; RANKL and RANK most probably have developing effects on bone

tumors while OPG could be decreasing cancer-induced bone pain [73, 90]. Tumor cells upregulating RANKL expression in bone stromal cells through increasing IL-6, IL-1B, TNF, epidermal growth factor and PTH-related peptide (PTH-rP), so osteoclastogenesis increased [91]. In this case, using the anti-RANKL treatment such as anti-RANKL medication (like denosumab) or gene therapy might have significant effects in cancer development [92]. Furthermore, recent investigations has suggested that mechanical pressure could lead osteocytes to recall osteoclasts to the bone resorption sites via regulating RANKL synthesis in osteoblasts [61].

Zinc and RANKL/RANK/OPG in bone

The aim of this study was to review the effect of zinc on RANKL/RANK/OPG pathway in the skeleton. In a recent animal study, zinc supplementation in ovariectomized (OVX) and diabetic (T1DM) rats resulted in increases in OPG expression that led to marked decreases in RANKL/OPG ratio [93]. Another study indicated zinc antioxidant property might prevent RANKL/RANK/OPG disbalance in Cd-induced rats [94], whilst an in vitro study on mouse marrow cells indicated that adding zinc sulfate led to inhibition in RANKL-induced osteoclast-like cells

[95]. Additionally, zinc sulfate could increase OPG mRNA expression after 24-48 hours in cultured cells, based on an in-vitro study [96]. The reports of a study regarding the effects of zinc demonstrated that as a result of using a zinc-free diet, the expression of RANK reduced and the level of RANKL and OPG did not change. Overall, [97] indicated that zinc deficiency could decrease both osteoclastogenesis and osteoblastogenesis [97]. The result of an in vitro investigation regarding M-CSF and RANKL highlighted that zinc can prevent osteoclasts differentiation by the dose-depending reduction in RANKL [98]. In a recent study, the effects of Puerarin and zinc on ovariectomized rat (OVX) bones demonstrated co-supplementation decreased RANKL and increased OPG and OPN (osteopontin) [99]. It was further demonstrated that RANKL worked through indirect Ca2+ signaling mediation [100]. Based on a rat study, adequate consumption of zinc likely does not affect osteoblastogenesis, but it may reduce osteoclastogenesis via suppression of RANK expression through prevention of ROS (reactive oxygen species) production and ERK (extracellular signal-regulated kinase) activation [101]. A recent study has shown, in the group of mice in the TNF inflammatory environment, with a higher concentration on Zn, RANKL expression is meaningfully diminished [102]. The effect of zinc supplementation on diabetic induced bone loss has been investigated through an animal study, demonstrating that supplementation can reduce chronic T1DM-induced bone loss

by increasing OC (osteocalcin) and decreasing RANKL and OPG [103]. Also, another experiment on diabetic rat demonstrated a reduction in RANK expression via zinc supplementation [104]. In Fong et. al, a higher ratio of RANKL/OPG was reported in a metallothionein knockout (MT -/-) group and received a lower amount of zinc compared to others [105]; whilst, according to Liang et. al, zinc could affect OPG gene expression in osteoblasts and increasing it [106].

In contrast to these studies, there are some studies which show distinct consequences. In a study on male rats that received zinc supplements, it was reported that dietary zinc supplementation upregulated RANKL expression in bone by TNF-α and IL-1β; this resulted in bone loss without any specific change in the size of bones [107]. It was shown in another animal study that suppressing ZIP4 (Zinc transporter ZIP4) could up-regulate bone mineral density and down-regulate bone turnover in mice with pancreatic cancer via RANKL-RANK system [108]; whilst some work has reported that zinc-modified titanium (EZ) affects osteoblasts differentiation but no significant changes in OPG and RANKL levels are observed [109] (Table1)(figure1).

Conclusion

In conclusion, it is likely that sufficient zinc intake will elicit positive effects on bone health by RANKL/RANK/OPG regulation. Although the outcomes of a few

studies are equivocal, it seems that zinc can exert the protective properties against bone loss by suppressing osteoclastogenesis via downregulation of RANKL/RANK. Additionally, there are several experiments where zinc supplementation resulted in upregulation of OPG expression. However, the results of limited studies oppose this. Therefore, aside from the positive role zinc possesses in preserving bone mass, further effects of zinc in RANKL/RANK/OPG system requires further animal/human studies.

Conflict of Interest:

The authors declare no conflict of interest regarding the present article.

References:

[1] F. Elefteriou, P. Campbell, Y. Ma, Control of bone remodeling by the peripheral sympathetic nervous system, Calcified tissue international 94(1) (2014) 140-51.

[2] R. Florencio-Silva, G.R. Sasso, E. Sasso-Cerri, M.J. Simoes, P.S. Cerri, Biology of Bone Tissue: Structure, Function, and Factors That Influence Bone Cells, BioMed research international 2015 (2015) 421746.

[3] A.L. Boskey, Bone composition: relationship to bone fragility and antiosteoporotic drug effects, BoneKEy reports 2 (2013) 447.

[4] S. Shenoy, J.K. Chawla, S. Gupta, J.S. Sandhu, Prevalence of low bone health using quantitative ultrasound in Indian women aged 41-60 years: Its association with nutrition and other related risk factors, Journal of women & aging 29(4) (2017) 334-347.

[5] G. Office of the Surgeon, Reports of the Surgeon General, Bone Health and Osteoporosis: A Report of the Surgeon General, Office of the Surgeon General (US), Rockville (MD), 2004.

[6] A.S. Karlamangla, S.M. Burnett-Bowie, C.J. Crandall, Bone Health During the Menopause Transition and Beyond, Obstetrics and gynecology clinics of North America 45(4) (2018) 695-708.

[7] W.E. Ward, J. Kaludjerovic, E.C. Dinsdale, A Mouse Model for Studying Nutritional Programming: Effects of Early Life Exposure to Soy Isoflavones on Bone and Reproductive Health, International journal of environmental research and public health 13(5) (2016).

[8] J. Zhang, K. Jameson, A.A. Sayer, S. Robinson, C. Cooper, E. Dennison, Accumulation of risk factors associated with poor bone health in older adults, Archives of osteoporosis 11 (2016) 3.

[9] T. Sozen, L. Ozisik, N.C. Basaran, An overview and management of osteoporosis, European journal of rheumatology 4(1) (2017) 46-56.
[10] L. Tian, R. Yang, L. Wei, J. Liu, Y. Yang, F. Shao, W. Ma, T. Li, Y. Wang, T. Guo, Prevalence of osteoporosis and related lifestyle and metabolic factors of postmenopausal women and elderly men: A cross-sectional study in Gansu province, Northwestern of China, Medicine 96(43) (2017) e8294.
[11] M.L. Brandi, Drugs for bone healing, Expert opinion on investigational drugs 21(8) (2012) 1169-76.
[12] M. Karimifar, F. Esmaili, A. Salari, A. Kachuei, Z. Faragzadegan, M. Karimifar, Effects of Levothyroxine and thyroid stimulating hormone on bone loss in patients with primary hypothyroidism, Journal of research in pharmacy practice 3(3) (2014) 83-7.
[13] P. Jackuliak, M. Kuzma, J. Payer, [Antidiabetic drugs and their effect on bone], Vnitrni lekarstvi 63(9) 609-616.
[14] T. Ito, R.T. Jensen, Association of long-term proton pump inhibitor therapy with bone fractures and effects on absorption of calcium, vitamin B12, iron, and magnesium, Current gastroenterology reports 12(6) (2010) 448-57.
[15] M.A. Gebara, M.L. Shea, K.L. Lipsey, S.L. Teitelbaum, R. Civitelli, D.J. Muller, C.F. Reynolds, 3rd, B.H. Mulsant, E.J. Lenze, Depression, antidepressants, and bone health in older adults: a systematic review, Journal of the American Geriatrics Society 62(8) (2014) 1434-41.
[16] C. Palacios, The role of nutrients in bone health, from A to Z, Critical reviews in food science and nutrition 46(8) (2006) 621-8.
[17] A.S. Prasad, Discovery of human zinc deficiency: its impact on human health and disease, Advances in nutrition (Bethesda, Md.) 4(2) (2013) 176-90.
[18] R.B. Saper, R. Rash, Zinc: an essential micronutrient, American family physician 79(9) (2009) 768-72.
[19] M. Yamaguchi, Role of nutritional zinc in the prevention of osteoporosis, Molecular and cellular biochemistry 338(1-2) (2010) 241-54.
[20] A.S. Prasad, Clinical manifestations of zinc deficiency, Annual review of nutrition 5 (1985) 341-63.
[21] B.C. Starcher, C.H. Hill, J.G. Madaras, Effect of zinc deficiency on bone collagenase and collagen turnover, The Journal of nutrition 110(10) (1980) 2095-102.
[22] S.A. Seyedmajidi, M. Seyedmajidi, A. Moghadamnia, S. Haghanifar, R. Ziaei, S. Zahedpasha, V. Arash, G. Jorsaraei, S. Halalkhor, Effect of zinc-deficient nutrition on craniofacial bone growth in rats, Dental research journal 11(4) (2014) 475-80.
[23] M. Mahdavi-Roshan, M. Ebrahimi, A. Ebrahimi, Copper, magnesium, zinc and calcium status in osteopenic and osteoporotic post-menopausal women, Clinical cases in mineral and bone metabolism : the official journal of the Italian Society of Osteoporosis, Mineral Metabolism, and Skeletal Diseases 12(1) (2015) 18-21.
[24] H.J. Seo, Y.E. Cho, T. Kim, H.I. Shin, I.S. Kwun, Zinc may increase bone formation through stimulating cell proliferation, alkaline phosphatase activity and collagen synthesis in osteoblastic MC3T3-E1 cells, Nutrition research and practice 4(5) (2010) 356-61.
[25] M. Yamaguchi, Role of zinc in bone metabolism and preventive effect on bone disorder, Biomedical Research on Trace Elements 18(4) (2007) 346-366.
[26] N. Amin, V. Boccardi, M. Taghizadeh, S. Jafarnejad, Probiotics and bone disorders: the role of RANKL/RANK/OPG pathway, Aging clinical and experimental research (2019).
[27] P.D. Saltman, L.G. Strause, The role of trace minerals in osteoporosis, Journal of the American College of Nutrition 12(4) (1993) 384-9.
[28] X. Qu, Z. He, H. Qiao, Z. Zhai, Z. Mao, Z. Yu, K. Dai, Serum copper levels are associated with bone mineral density and total fracture, Journal of orthopaedic translation 14 (2018) 34-44.
[29] T.S. Orchard, J.C. Larson, N. Alghothani, S. Bout-Tabaku, J.A. Cauley, Z. Chen, A.Z. LaCroix, J. Wactawski-Wende, R.D. Jackson, Magnesium intake, bone mineral density, and fractures: results from

the Women's Health Initiative Observational Study, The American journal of clinical nutrition 99(4) (2014) 926-33.
[30] K.H. Lim, L.J. Riddell, C.A. Nowson, A.O. Booth, E.A. Szymlek-Gay, Iron and zinc nutrition in the economically-developed world: a review, Nutrients 5(8) (2013) 3184-211.
[31] I. Zofkova, P. Nemcikova, P. Matucha, Trace elements and bone health, Clinical chemistry and laboratory medicine 51(8) (2013) 1555-61.
[32] K.A. McCall, C.-c. Huang, C.A. Fierke, Function and mechanism of zinc metalloenzymes, The Journal of nutrition 130(5) (2000) 1437S-1446S.
[33] G.D. Pepa, M.L. Brandi, Microelements for bone boost: the last but not the least, Clinical cases in mineral and bone metabolism : the official journal of the Italian Society of Osteoporosis, Mineral Metabolism, and Skeletal Diseases 13(3) (2016) 181-185.
[34] K. Jurowski, B. Szewczyk, G. Nowak, W. Piekoszewski, Biological consequences of zinc deficiency in the pathomechanisms of selected diseases, Journal of biological inorganic chemistry : JBIC : a publication of the Society of Biological Inorganic Chemistry 19(7) (2014) 1069-79.
[35] S.A. Barrie, J.V. Wright, J.E. Pizzorno, E. Kutter, P.C. Barron, Comparative absorption of zinc picolinate, zinc citrate and zinc gluconate in humans, Agents and actions 21(1-2) (1987) 223-8.
[36] M. Gupta, V.K. Mahajan, K.S. Mehta, P.S. Chauhan, Zinc therapy in dermatology: a review, Dermatology research and practice 2014 (2014) 709152.
[37] H. Hemila, J.T. Fitzgerald, E.J. Petrus, A. Prasad, Zinc Acetate Lozenges May Improve the Recovery Rate of Common Cold Patients: An Individual Patient Data Meta-Analysis, Open forum infectious diseases 4(2) (2017) ofx059.
[38] T. Kambe, K. Fukue, R. Ishida, S. Miyazaki, Overview of Inherited Zinc Deficiency in Infants and Children, Journal of nutritional science and vitaminology 61 Suppl (2015) S44-6.
[39] L. Rossi, S. Migliaccio, A. Corsi, M. Marzia, P. Bianco, A. Teti, L. Gambelli, S. Cianfarani, F. Paoletti, F. Branca, Reduced growth and skeletal changes in zinc-deficient growing rats are due to impaired growth plate activity and inanition, The Journal of nutrition 131(4) (2001) 1142-6.
[40] G.J. Fosmire, Zinc toxicity, The American journal of clinical nutrition 51(2) (1990) 225-7.
[41] D.G. Masters, C.L. Keen, B. Lönnerdal, L.S. Hurley, Release of zinc from maternal tissues during zinc deficiency or simultaneous zinc and calcium deficiency in the pregnant rat, The Journal of nutrition 116(11) (1986) 2148-2154.
[42] M. Yamaguchi, Role of zinc in bone formation and bone resorption, The Journal of Trace Elements in Experimental Medicine: The Official Publication of the International Society for Trace Element Research in Humans 11(2-3) (1998) 119-135.
[43] R. Razmandeh, E. Nasli-Esfahani, R. Heydarpour, F. Faridbod, M.R. Ganjali, P. Norouzi, B. Larijani, D. Khoda-Amorzideh, Association of Zinc, Copper and Magnesium with bone mineral density in Iranian postmenopausal women - a case control study, Journal of diabetes and metabolic disorders 13(1) (2014) 43.
[44] G.J. Atkins, D.M. Findlay, P.H. Anderson, H.A. Morris, Target genes: bone proteins, Vitamin D, Elsevier2011, pp. 411-424.
[45] P. Bhardwaj, D.V. Rai, M.L. Garg, Zinc as a nutritional approach to bone loss prevention in an ovariectomized rat model, Menopause (New York, N.Y.) 20(11) (2013) 1184-93.
[46] M. Yamaguchi, Nutritional zinc plays a pivotal role in bone health and osteoporosis prevention, aging 6 (2015) 8.
[47] M. Herzberg, J. Foldes, R. Steinberg, J. Menczel, Zinc excretion in osteoporotic women, Journal of Bone and Mineral Research 5(3) (1990) 251-257.
[48] M. Jovanovic, F.N. Schmidt, G. Guterman-Ram, H. Khayyeri, S. Hiram-Bab, A. Orenbuch, S. Katchkovsky, A. Aflalo, H. Isaksson, B. Busse, K. Jahn, N. Levaot, Perturbed bone composition and integrity with disorganized osteoblast function in zinc receptor/Gpr39-deficient mice, FASEB journal :

official publication of the Federation of American Societies for Experimental Biology 32(5) (2018) 2507-2518.
[49] C.V. Gurban, O. Mederle, The OPG/RANKL system and zinc ions are promoters of bone remodeling by osteoblast proliferation in postmenopausal osteoporosis, Romanian journal of morphology and embryology = Revue roumaine de morphologie et embryologie 52(3 Suppl) (2011) 1113-9.
[50] M. Yamaguchi, Role of nutritional zinc in the prevention of osteoporosis, Molecular and cellular biochemistry 338(1-2) (2010) 241-254.
[51] M.S. Bolze, R.D. Reeves, F.E. Lindbeck, M.J. Elders, Influence of zinc on growth, somatomedin, and glycosaminoglycan metabolism in rats, The American journal of physiology 252(1 Pt 1) (1987) E21-6.
[52] J. Kalinowski, E.R. Chavez, Tissue composition and trace mineral content of the dam and litter under low dietary zinc intake during gestation and lactation of first-litter gilts, Journal of trace elements and electrolytes in health and disease 5(1) (1991) 35-46.
[53] K.M. Sinha, X. Zhou, Genetic and molecular control of osterix in skeletal formation, Journal of cellular biochemistry 114(5) (2013) 975-84.
[54] A. Igarashi, M. Yamaguchi, Increase in bone growth factors with healing rat fractures: the enhancing effect of zinc, International journal of molecular medicine 8(4) (2001) 433-8.
[55] A. Sadighi, M.M. Roshan, A. Moradi, A. Ostadrahimi, The effects of zinc supplementation on serum zinc, alkaline phosphatase activity and fracture healing of bones, Saudi medical journal 29(9) (2008) 1276-9.
[56] Y. Abu-Amer, NF-κB signaling and bone resorption, Osteoporosis international 24(9) (2013) 2377-2386.
[57] B.F. Boyce, Z. Yao, L. Xing, Functions of nuclear factor κB in bone, Annals of the New York Academy of Sciences 1192(1) (2010) 367-375.
[58] Y. Abu-Amer, NF-kappaB signaling and bone resorption, Osteoporosis international : a journal established as result of cooperation between the European Foundation for Osteoporosis and the National Osteoporosis Foundation of the USA 24(9) (2013) 2377-86.
[59] M.C. Walsh, Y. Choi, Biology of the RANKL-RANK-OPG System in Immunity, Bone, and Beyond, Frontiers in immunology 5 (2014) 511.
[60] B.F. Boyce, L. Xing, Biology of RANK, RANKL, and osteoprotegerin, Arthritis research & therapy 9(1) (2007) S1.
[61] B.F. Boyce, L. Xing, Functions of RANKL/RANK/OPG in bone modeling and remodeling, Archives of biochemistry and biophysics 473(2) (2008) 139-46.
[62] C.M. Wang, S.C. Tsai, J.C. Lin, Y.J. Wu, J. Wu, J.Y. Chen, Association of Genetic Variants of RANK, RANKL, and OPG with Ankylosing Spondylitis Clinical Features in Taiwanese, Mediators of inflammation 2019 (2019) 8029863.
[63] J.E. Fata, Y.Y. Kong, J. Li, T. Sasaki, J. Irie-Sasaki, R.A. Moorehead, R. Elliott, S. Scully, E.B. Voura, D.L. Lacey, W.J. Boyle, R. Khokha, J.M. Penninger, The osteoclast differentiation factor osteoprotegerin-ligand is essential for mammary gland development, Cell 103(1) (2000) 41-50.
[64] N.S. Kim, H.J. Kim, B.K. Koo, M.C. Kwon, Y.W. Kim, Y. Cho, Y. Yokota, J.M. Penninger, Y.Y. Kong, Receptor activator of NF-kappaB ligand regulates the proliferation of mammary epithelial cells via Id2, Molecular and cellular biology 26(3) (2006) 1002-13.
[65] M. Bezerra, J. Carvalho, A. Prokopowitsch, R. Pereira, RANK, RANKL and osteoprotegerin in arthritic bone loss, Brazilian Journal of Medical and Biological Research 38(2) (2005) 161-170.
[66] S. Colucci, G. Brunetti, F.P. Cantatore, A. Oranger, G. Mori, L. Quarta, N. Cirulli, L. Mancini, A. Corrado, F. Grassi, Lymphocytes and synovial fluid fibroblasts support osteoclastogenesis through RANKL, TNFα, and IL-7 in an in vitro model derived from human psoriatic arthritis, The Journal of Pathology: A Journal of the Pathological Society of Great Britain and Ireland 212(1) (2007) 47-55.

[67] B. Mrozikiewicz-Rakowska, P. Nehring, K. Szymanski, A. Sobczyk-Kopciol, R. Ploski, W. Drygas, J. Krzymien, N.A. Acharya, L. Czupryniak, A. Przybylkowski, Selected RANKL/RANK/OPG system genetic variants in diabetic foot patients, Journal of diabetes and metabolic disorders 17(2) (2018) 287-296.
[68] B.F. Boyce, L. Xing, The Rankl/Rank/Opg pathway, Current osteoporosis reports 5(3) (2007) 98-104.
[69] W.S. Simonet, D.L. Lacey, C.R. Dunstan, M. Kelley, M.S. Chang, R. Luthy, H.Q. Nguyen, S. Wooden, L. Bennett, T. Boone, G. Shimamoto, M. DeRose, R. Elliott, A. Colombero, H.L. Tan, G. Trail, J. Sullivan, E. Davy, N. Bucay, L. Renshaw-Gegg, T.M. Hughes, D. Hill, W. Pattison, P. Campbell, S. Sander, G. Van, J. Tarpley, P. Derby, R. Lee, W.J. Boyle, Osteoprotegerin: a novel secreted protein involved in the regulation of bone density, Cell 89(2) (1997) 309-19.
[70] T.J. Yun, P.M. Chaudhary, G.L. Shu, J.K. Frazer, M.K. Ewings, S.M. Schwartz, V. Pascual, L.E. Hood, E.A. Clark, OPG/FDCR-1, a TNF receptor family member, is expressed in lymphoid cells and is up-regulated by ligating CD40, Journal of immunology (Baltimore, Md. : 1950) 161(11) (1998) 6113-21.
[71] P.E. Reid, N.J. Brown, I. Holen, Breast cancer cells stimulate osteoprotegerin (OPG) production by endothelial cells through direct cell contact, Molecular cancer 8 (2009) 49.
[72] M. Baud'huin, L. Duplomb, S. Teletchea, F. Lamoureux, C. Ruiz-Velasco, M. Maillasson, F. Redini, M.F. Heymann, D. Heymann, Osteoprotegerin: multiple partners for multiple functions, Cytokine & growth factor reviews 24(5) (2013) 401-9.
[73] M. Sisay, G. Mengistu, D. Edessa, The RANK/RANKL/OPG system in tumorigenesis and metastasis of cancer stem cell: potential targets for anticancer therapy, OncoTargets and therapy 10 (2017) 3801-3810.
[74] R. Hanada, T. Hanada, V. Sigl, D. Schramek, J.M. Penninger, RANKL/RANK—beyond bones, Journal of Molecular Medicine 89(7) (2011) 647-656.
[75] O.R. Tamtaji, S. Borzabadi, M. Ghayour-Mobarhan, G. Ferns, Z. Asemi, The effects of fatty acids consumption on OPG/RANKL/RANK system in cardiovascular diseases: Current status and future perspectives for the impact of diet-gene interaction, Journal of cellular biochemistry 120(3) (2019) 2774-2781.
[76] S.-I. Hayashi, T. Yamada, M. Tsuneto, T. Yamane, M. Takahashi, L.D. Shultz, H. Yamazaki, Distinct osteoclast precursors in the bone marrow and extramedullary organs characterized by responsiveness to Toll-like receptor ligands and TNF-α, The Journal of Immunology 171(10) (2003) 5130-5139.
[77] L. Rochette, A. Meloux, E. Rigal, M. Zeller, Y. Cottin, C. Vergely, The Role of Osteoprotegerin and Its Ligands in Vascular Function, International journal of molecular sciences 20(3) (2019) 705.
[78] N. Renema, B. Navet, M.F. Heymann, F. Lezot, D. Heymann, RANK-RANKL signalling in cancer, Bioscience reports 36(4) (2016).
[79] D. Vega, N.M. Maalouf, K. Sakhaee, The role of receptor activator of nuclear factor-κB (RANK)/RANK ligand/osteoprotegerin: clinical implications, The Journal of Clinical Endocrinology & Metabolism 92(12) (2007) 4514-4521.
[80] M.C. Walsh, Y. Choi, Biology of the RANKL–RANK–OPG system in immunity, bone, and beyond, Frontiers in immunology 5 (2014) 511.
[81] M. Infante, A. Fabi, F. Cognetti, S. Gorini, M. Caprio, A. Fabbri, RANKL/RANK/OPG system beyond bone remodeling: involvement in breast cancer and clinical perspectives, Journal of Experimental & Clinical Cancer Research 38(1) (2019) 12.
[82] L. Ardeshirpour, C. Dumitru, P. Dann, J. Sterpka, J. VanHouten, W. Kim, P. Kostenuik, J. Wysolmerski, OPG Treatment Prevents Bone Loss During Lactation But Does Not Affect Milk Production or Maternal Calcium Metabolism, Endocrinology 156(8) (2015) 2762-73.
[83] F. Arai, T. Miyamoto, O. Ohneda, T. Inada, T. Sudo, K. Brasel, T. Miyata, D.M. Anderson, T. Suda, Commitment and differentiation of osteoclast precursor cells by the sequential expression of c-Fms and receptor activator of nuclear factor kappaB (RANK) receptors, The Journal of experimental medicine 190(12) (1999) 1741-54.

[84] F. Xu, S.L. Teitelbaum, Osteoclasts: new insights, Bone research 1 (2013) 11.
[85] N. Bucay, I. Sarosi, C.R. Dunstan, S. Morony, J. Tarpley, C. Capparelli, S. Scully, H.L. Tan, W. Xu, D.L. Lacey, W.J. Boyle, W.S. Simonet, osteoprotegerin-deficient mice develop early onset osteoporosis and arterial calcification, Genes & development 12(9) (1998) 1260-8.
[86] P.J. Kostenuik, V. Shalhoub, Osteoprotegerin: a physiological and pharmacological inhibitor of bone resorption, Current pharmaceutical design 7(8) (2001) 613-35.
[87] M. Papadaki, V. Rinotas, F. Violitzi, T. Thireou, G. Panayotou, M. Samiotaki, E. Douni, New insights for RANKL as a proinflammatory modulator in modeled inflammatory arthritis, Frontiers in immunology 10 (2019).
[88] G.D. Roodman, J.J. Windle, Paget disease of bone, The Journal of clinical investigation 115(2) (2005) 200-8.
[89] C. Menaa, S.V. Reddy, N. Kurihara, H. Maeda, D. Anderson, T. Cundy, J. Cornish, F.R. Singer, J.M. Bruder, G.D. Roodman, Enhanced RANK ligand expression and responsivity of bone marrow cells in Paget's disease of bone, The Journal of clinical investigation 105(12) (2000) 1833-8.
[90] D.R. Clohisy, P.W. Mantyh, Bone cancer pain and the role of RANKL/OPG, Journal of musculoskeletal & neuronal interactions 4(3) (2004) 293-300.
[91] Y. Nakai, K. Okamoto, A. Terashima, S. Ehata, J. Nishida, T. Imamura, T. Ono, H. Takayanagi, Efficacy of an orally active small-molecule inhibitor of RANKL in bone metastasis, Bone research 7(1) (2019) 1.
[92] D. Santini, G. Schiavon, B. Vincenzi, L. Gaeta, F. Pantano, A. Russo, C. Ortega, C. Porta, S. Galluzzo, G. Armento, N. La Verde, C. Caroti, I. Treilleux, A. Ruggiero, G. Perrone, R. Addeo, P. Clezardin, A.O. Muda, G. Tonini, Receptor activator of NF-kB (RANK) expression in primary tumors associates with bone metastasis occurrence in breast cancer patients, PloS one 6(4) (2011) e19234.
[93] E.C.S. Ferreira, R.H. Bortolin, F.P. Freire-Neto, K.S.C. Souza, J.F. Bezerra, M.A.G. Ururahy, A.M.O. Ramos, S.T. Himelfarb, B.J. Abreu, T.V.N. Didone, L.F.C. Pedrosa, A.C. Medeiros, S.Q. Doi, J. Brandao-Neto, R.D.C. Hirata, L.A. Rezende, M.G. Almeida, M.H. Hirata, A.A. Rezende, Zinc supplementation reduces RANKL/OPG ratio and prevents bone architecture alterations in ovariectomized and type 1 diabetic rats, Nutrition research (New York, N.Y.) 40 (2017) 48-56.
[94] M.M. Brzoska, J. Rogalska, Protective effect of zinc supplementation against cadmium-induced oxidative stress and the RANK/RANKL/OPG system imbalance in the bone tissue of rats, Toxicology and applied pharmacology 272(1) (2013) 208-20.
[95] M. Yamaguchi, S. Uchiyama, Receptor activator of NF-κB ligand-stimulated osteoclastogenesis in mouse marrow culture is suppressed by zinc in vitro, International journal of molecular medicine 14(1) (2004) 81-85.
[96] M. Yamaguchi, M. Goto, S. Uchiyama, T. Nakagawa, Effect of zinc on gene expression in osteoblastic MC3T3-E1 cells: enhancement of Runx2, OPG, and regucalcin mRNA expressions, Molecular and cellular biochemistry 312(1-2) (2008) 157-66.
[97] M. Hie, N. Iitsuka, T. Otsuka, A. Nakanishi, I. Tsukamoto, Zinc deficiency decreases osteoblasts and osteoclasts associated with the reduced expression of Runx2 and RANK, Bone 49(6) (2011) 1152-9.
[98] K.H. Park, B. Park, D.S. Yoon, S.H. Kwon, D.M. Shin, J.W. Lee, H.G. Lee, J.H. Shim, J.H. Park, J.M. Lee, Zinc inhibits osteoclast differentiation by suppression of Ca2+-Calcineurin-NFATc1 signaling pathway, Cell communication and signaling : CCS 11 (2013) 74.
[99] H. Liu, W. Li, S. Jia, B. Li, Puerarin and zinc additively prevent mandibular bone loss through inhibiting osteoclastogenesis in ovariectomized rats, Histology and histopathology 32(8) (2017) 851-860.
[100] M. Yamaguchi, Role of zinc in regulation of osteoclastogenesis, Biomedical Research on Trace Elements 15(1) (2004) 9-14.
[101] M. Hie, I. Tsukamoto, Administration of zinc inhibits osteoclastogenesis through the suppression of RANK expression in bone, European journal of pharmacology 668(1-2) (2011) 140-6.

[102] J.Z. Tan, E.M. Nie, C.Y. Zhang, R. Jiang, [Effect of zinc ion on the expression of osteoblastic proteins in MC3T3-E1 cells in inflammatory environment], Zhonghua kou qiang yi xue za zhi = Zhonghua kouqiang yixue zazhi = Chinese journal of stomatology 51(8) (2016) 486-90.
[103] R.H. Bortolin, B.J. da Graca Azevedo Abreu, M. Abbott Galvao Ururahy, K.S. Costa de Souza, J.F. Bezerra, M.B. Loureiro, F.S. da Silva, D.E. Marques, A.A. Batista, G. Oliveira, A.D. Luchessi, V.M. Lima, C.E. Miranda, M.V. Lia Fook, M. Almeida, L.A. de Rezende, A.A. de Rezende, Protection against T1DM-Induced Bone Loss by Zinc Supplementation: Biomechanical, Histomorphometric, and Molecular Analyses in STZ-Induced Diabetic Rats, PloS one 10(5) (2015) e0125349.
[104] N. Iitsuka, M. Hie, I. Tsukamoto, Zinc supplementation inhibits the increase in osteoclastogenesis and decrease in osteoblastogenesis in streptozotocin-induced diabetic rats, European journal of pharmacology 714(1-3) (2013) 41-7.
[105] L. Fong, K. Tan, C. Tran, J. Cool, M.A. Scherer, R. Elovaris, P. Coyle, B.K. Foster, A.M. Rofe, C.J. Xian, Interaction of dietary zinc and intracellular binding protein metallothionein in postnatal bone growth, Bone 44(6) (2009) 1151-62.
[106] D. Liang, M. Yang, B. Guo, J. Cao, L. Yang, X. Guo, Zinc upregulates the expression of osteoprotegerin in mouse osteoblasts MC3T3-E1 through PKC/MAPK pathways, Biological trace element research 146(3) (2012) 340-8.
[107] T. Suzuki, S. Katsumata, H. Matsuzaki, K. Suzuki, Dietary zinc supplementation increased TNFalpha and IL1beta-induced RANKL expression, resulting in a decrease in bone mineral density in rats, Journal of clinical biochemistry and nutrition 58(1) (2016) 48-55.
[108] Q. Zhang, X. Sun, J. Yang, H. Ding, D. LeBrun, K. Ding, C.W. Houchen, R.G. Postier, C.G. Ambrose, Z. Li, X. Bi, M. Li, ZIP4 silencing improves bone loss in pancreatic cancer, Oncotarget 6(28) (2015) 26041-51.
[109] K. Yusa, O. Yamamoto, M. Iino, H. Takano, M. Fukuda, Z. Qiao, T. Sugiyama, Eluted zinc ions stimulate osteoblast differentiation and mineralization in human dental pulp stem cells for bone tissue engineering, Archives of oral biology 71 (2016) 162-169.

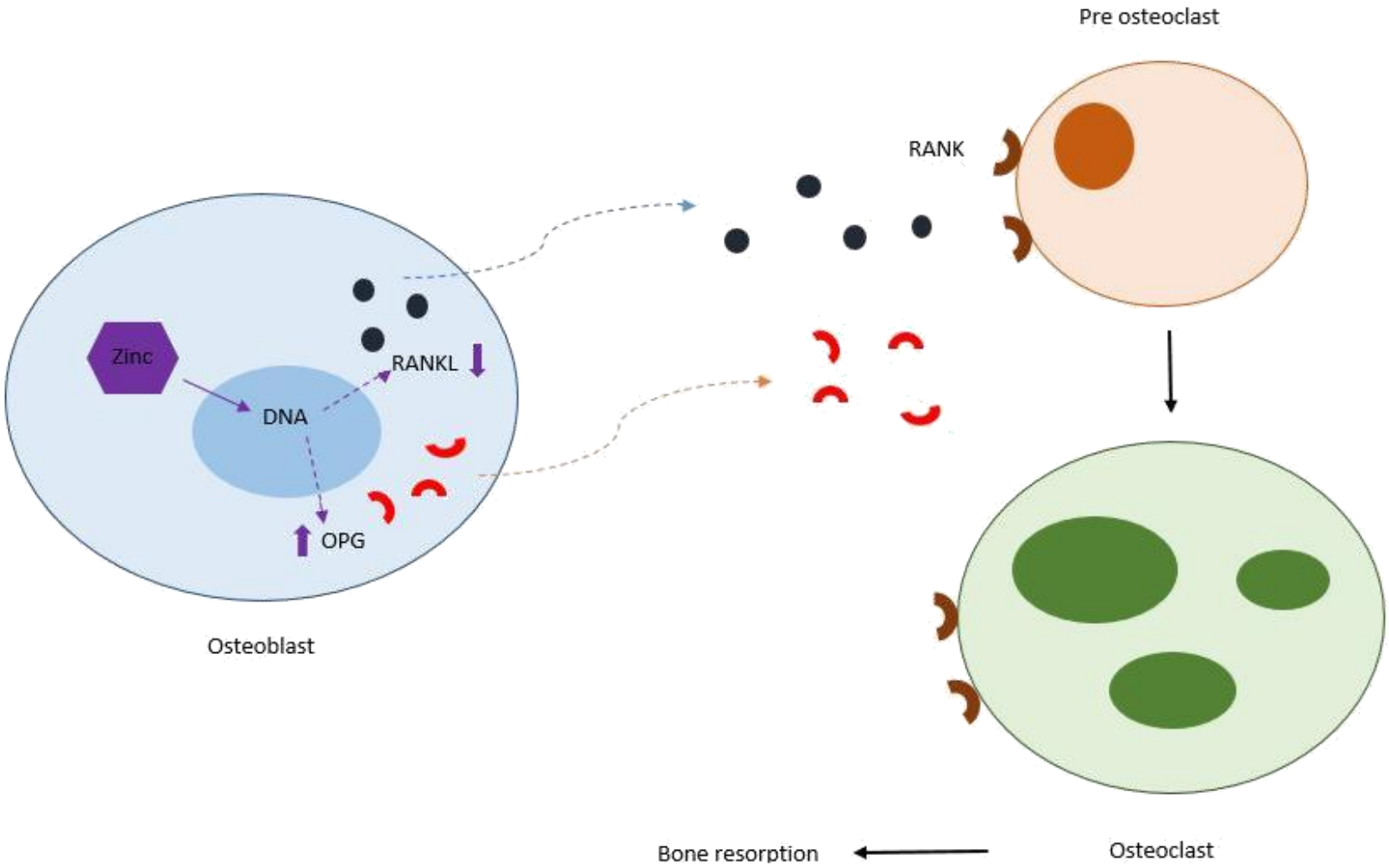

Figure 1: Binding between RANKL and RANK lead to maturation in osteoclasts and enhance bone resorption. OPG, as RANKL decoy receptor, would prevent of RANKL/RANK binding and down-regulating bone loss. Most of the studies have reviewed, indicated a significant reduction in RANKL and increment in OPG. RANKL (receptor activator of NF-kB ligand), RANK (receptor activator of NF-kB), OPG (osteoprotegerin).

intervention	time	species	outcome	reference
Zinc supplement	90 days	OVX/T1DM rats	↓ RANKL/OPG	[93]
Zinc supplement	6 months	Cadmium induced male rats	↓ sRANKL ↓ RANKL/OPG	[94]
Zinc sulfate	7 days	Mouse marrow cells (in vitro)	↓ RANKL	[95]
Zinc sulfate	3 days	Osteoblastic cells (in vitro)	↑ OPG	[96]
Zinc deficiency	3 weeks	Female rats	↓ RANK	[97]
zinc	4 days	Mice (in vitro)	↓ RANKL	[98]
Zinc + puerarin	12 weeks	OVX rats	↓ RANKL ↑ OPG	[99]
zinc	1 week	Female rats	↓ RANK	[101]
zinc	3s days	Mice osteoblasts cell (in vitro/inflammatory condition)	↓ RANKL	[102]

Zinc supplement	90 days	Diabetic male rats	↓ RANKL ↓ OPG	[103]
Zinc supplement	1 week	Diabetic rats (STZ-induced)	↓ RANK	[104]
zinc	2 days	Osteoblastic cells (in vitro)	↑ OPG	[106]
Zinc supplement	4 weeks	Male rats	↓ RANKL	[107]
ZIP4 suppression	-	Mouse (pancreatic cancer)	↓ RANKL	[108]
Zinc-modified titanium	28 days	Human stem cells (in vitro)	No change in OPG RANKL	[109]

Table 1: Summary of the reviewed studies, describing the effects of Zn on RANKL/RANK/OPG pathway in skeleton. RANK (receptor activator of Nf-kB), RANKL (receptor activator of NF-kB ligand), OPG (osteoprotegerin) and OVX (ovariectomized), STZ (streptozotocine).

Conflict of Interest:

The authors declare no conflict of interest in carrying out and reporting this research.

Corresponding author: