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Review Article

The Anti-Inflammatory Role of Vitamin E in Prevention of Osteoporosis

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There is growing evidence that inflammation may be one of the causal factors of osteoporosis. Several cytokines such as IL-1, IL-6, RANKL, OPG, and M-CSF were implicated in the pathogenesis of osteoporosis. These cytokines are important determinants of osteoclast differentiation and its bone resorptive activity. Anticytokine therapy using cytokine antagonists such as IL-receptor antagonist and TNF-binding protein was able to suppress the activity of the respective cytokines and prevent bone loss. Several animal studies have shown that vitamin E in the forms of palm-derived tocotrienol and α-tocopherol may prevent osteoporosis in rat models by suppressing IL-1 and IL-6. Free radicals are known to activate transcription factor NFκB which leads to the production of bone resorbing cytokines. Vitamin E, a potent antioxidant, may be able to neutralise free radicals before they could activate NFκB, therefore suppressing cytokine production and osteoporosis. Vitamin E has also been shown to inhibit COX-2, the enzyme involved in inflammatory reactions. Of the two types of vitamin E studied, tocotrienol seemed to be better than tocopherol in terms of its ability to suppress bone-resorbing cytokines.

1. Introduction

Osteoporosis is a bone disease, characterized by low bone mass and increased risk of fractures [1]. It is well accepted that osteoporosis can be caused by various endocrine, metabolic, and mechanical factors. However, recently, there are opinions that there may be an inflammatory component in the etiology of osteoporosis [2, 3]. There is plenty of evidence linking inflammation to osteoporosis. Epidemiological studies have identified higher incidence of osteoporosis in various inflammatory conditions such as ankylosing spondylitis, rheumatoid arthritis, and systemic lupus erythematosus [4–7]. This association was also observed clinically whereby the degree of osteoporosis was equivalent to the extent of inflammation. If the inflammation was systemic, bone loss will occur at all skeletal sites, whereas if the inflammation was only restricted to a site, bone loss will only occur locally at that site of inflammation [3]. Elderly patients are more prone to osteoporosis, and this was believed to be connected to the elevated production of proinflammatory cytokines with aging [8, 9].

The occurrence of inflammation is indicated by the presence of inflammatory markers such as cytokines and C-reactive protein. Biochemical studies have demonstrated elevation of proinflammatory cytokines TNF-α and IL-6 in arthritic disease such as gouty arthritis, rheumatoid arthritis, and psoriatic arthritis [10, 11]. An obvious relationship between inflammation and osteoporosis was seen in rheumatoid arthritis, whereby proinflammatory cytokines were released causing bone loss around the affected joints [12]. The level of C-reactive protein, a sensitive marker of systemic inflammation, was also found to be associated with bone mineral density [13]. Inflammation may contribute to bone loss by affecting the bone remodeling process, favouring bone resorption activity by osteoclasts rather than bone formation activity by osteoblasts [14, 15]. Bone resorption is determined by the balance between two cytokines, receptor activator of nuclear factor κB ligand (RANKL), and osteoprotegerin (OPG) [16]. RANKL is crucial for the differentiation and activation of osteoclast [17]. Higher RANKL levels were associated with lower bone mineral density in men [18]. Administration of serum

RANKL to mice promoted osteoclast growth and activation, leading to osteoporosis [19]. On the other hand, OPG antagonizes RANKL by binding with RANKL and preventing it from binding to RANK receptors. By doing that, OPG was able to inhibit osteoclastogenesis and bone resorption [20]. Macrophage colony stimulating factor (MCSF) is another important determinant of osteoclastogenesis, but its mechanism to modulate osteoclastogenesis is still not clear [20].

The "upstream" cytokines such as IL-1, IL-6, and TNF-α [21, 22] and "downstream" cytokines such as RANKL, OPG, and M-CSF [23–25] played an important role in bone remodeling. Imbalance in their bioactivity may lead to bone loss and osteoporosis. Cytokines are small- to medium-sized proteins or glycoproteins with molecular weight ranging from 8 to 40,000 dalton. They act as the biological mediator for most cells and function at low concentrations between 10^{-10} and 10^{-5} molar. They have a short half-life of less than 10 minutes, and their serum level can be as low as 10 pg/mL. The cytokine levels increase dramatically during inflammation and infection. The measurement of cytokine levels in close vicinity to bone such as the bone marrow is important for studies on osteoporosis and other bone diseases. In postmenopausal women, cytokine production by the peripheral monocytes correlated well with cytokines secreted by monocytes in the bone marrow. Therefore, cytokine levels in the serum are representative of the local monocytes [26]. Stromal cells and osteoblasts produce interleukin-1, interleukin-6, and tumor necrosis factor-α. These proinflammatory cytokines are also known as the bone-resorbing cytokines or proosteoclast cytokines as they promote osteoclast differentiation and activity [27–30]. The bone resorption activity of these cytokines in ovariectomised rats was reduced with anticytokine therapy such as IL-1 receptor antagonists and TNF-binding protein [31]. Vitamin E, a potent antioxidant vitamin, was also found to inhibit or suppress cytokine production [32, 33]. This vitamin E action may be responsible for its ability to prevent inflammation and osteoporosis, seen in several studies on osteoporosis using animal models [34].

Vitamin E is a group of potent, lipid-soluble, chain-breaking antioxidants. It can be classified into tocopherol and tocotrienol based on the chemical structure. Palm oil, which is extracted from the pulp of the fruit of the oil palm *Elaeis guineensis*, is abundant in tocotrienols. Tocotrienol has an unsaturated farnesyl (isoprenoid) side-chain, while tocopherol has a saturated phytyl side chain [35].

Vitamin E occurs in eight isoforms of α-, β-, γ-, and δ-tocopherols or tocotrienols. It was thought that both the γ and δ isomers of tocopherol have better antioxidant and anti-inflammatory activities than the α isomer [36, 37]. Once vitamin E is absorbed in the intestine, it will enter the circulation via the lymphatic system and be transported to the liver with the chylomicrons [38]. Vitamin E is metabolized by cytochrome P450 and then excreted in the urine [39].

In human subjects and animal models, high doses of vitamin E were found to exhibit anti-inflammatory effects by decreasing C-reactive protein (CRP) and inhibiting the release of proinflammatory cytokines [40]. These were evident in a study on patients with coronary artery disease, whereby the CRP and tumor necrosis factor-α (TNF-α) concentrations were found to be significantly lowered with α-tocopherol supplementation compared to placebo [41]. Since vitamin E was also found to inhibit cyclooxygenase-2 activities, it was thought to be able to exert anti-inflammatory and anticarcinogenic activities, especially in the colon [42]. This was demonstrated by Yang et al. [43], who found that vitamin E was able to significantly lower colon inflammation index and reduced the number of colon adenomas in mice given azoxymethane.

This paper will focus on the effects of vitamin E on bone-resorbing cytokines with special attention on IL-1 and IL-6.

2. Interleukin-1 (IL-1)

IL-1 plays an important role in various reactions towards infection, inflammation, and immune activation. This cytokine is produced by various cells but the main producer is the monocyte. In the physiological condition, monocytes do not secrete IL-1 but, under pathological conditions such as septic shock, IL-1 is rapidly released and acts directly on the blood vessels. Other cytokines such as TNF-α and interferon, bacterial endotoxin, virus, and antigen can also stimulate the release of IL-1. Reactive oxygen species such as superoxide radicals have been shown to induce IL-1 production [32, 44]. IL-1 is involved in the pathogenesis of various diseases associated with bone loss such as osteoporosis [45, 46], cancer-induced osteolysis [47], rheumatoid arthritis [48], and osteolysis of orthopedic implants [49]. IL-1 is also an important factor in both *in vivo* and *in vitro* bone resorption [50, 51]. It stimulates the formation and activity of osteoclasts, leading to excessive bone resorption. Suda et al. [52] demonstrated that the presence of osteoblast and stromal cells was crucial in the formation of osteoclasts by IL-1. Thomson et al. [53] also reported that osteoblasts secrete a factor that stimulates the bone-resorbing activities of rat osteoclasts. However, Xu et al. [54] demonstrated that rat osteoclasts expressed mRNA to IL-1 receptors, while Yu and Ferrier [55] found that osteoclast is one of the target cells for IL-1. These studies proved that IL-1 can act directly on osteoclasts without the presence of osteoblasts or stromal cells. IL-1 may also promote formation of osteoclasts [56]. It acts by activating nuclear factor κB (NFκB) in osteoclast and prevents its apoptosis [57]. It was found that the estrogen-deficient state in postmenopausal women or ovariectomised rats resulted in increased production of IL-1 by monocyte and other bone marrow cells [58, 59]. Estrogen replacement or IL-receptor antagonist was able to prevent the elevation of IL-1 in ovariectomised rats [60, 61]. Vitamin E was also found to have the ability to suppress IL-1 production by activated monocytes [62]. In a different study, combination of superoxide dismutase and vitamin E was effective in inhibiting IL-1 production by human monocytes [32]. The ability of vitamin E to inhibit IL-1 in the bone environment may have prevented bone loss.

3. Interleukin-6 (IL-6)

IL-6 is another cytokine that is associated with various pathophysiological processes in humans. It is produced by the haematopoetic and nonhaematopoetic cells when they were exposed to various types of stimulation. During bone remodeling, IL-6 is produced in nanomolar concentrations by stromal cells and osteoblasts under the influence of parathyroid hormone, vitamin D_3, growth factor, and other cytokines [63]. IL-6 was also reported to be produced by osteoblasts when stimulated by IL-1, TNF-α, and lipopolysaccharide [64]. McSheeny and Chambers [65] reported that osteoblasts were stimulated by local IL-1 to produce IL-6, which was responsible for the activation of osteoclasts. IL-6 promoted the differentiation of osteoclasts from its precursor and played an important role in the pathogenesis of osteoporosis due to estrogen deficiency [66, 67]. The IL-6 elevation in postmenopausal women was reduced by estrogen replacement therapy [68]. The elevation of IL-6 may be related to free radical activities especially reactive oxygen species. Reactive oxygen species was found to elevate the IL-6 levels directly via activation of nuclear factor κB (NFκB) [69]. High cytokine levels would also result in activation of NFκB and promotion of osteoclastogenesis [70].

4. Vitamin E as Anticytokine Agent

The effects of vitamin E on bone resorbing cytokines for prevention and treatment of osteoporosis have been studied using FeNTA and nicotine rat models [34, 71]. These models represent osteoporosis caused by oxidative stress and smoking, respectively. However, similar studies in humans are still lacking. Ferric nitrilotriacetate (FeNTA) is an oxidizing agent which produces free radicals via the Fenton reaction [72, 73]. Oxidative stress can be induced in rats by injecting them with FeNTA, allowing the hazardous effects of free radicals on various organs and tissues including bone to be studied. The bone resorbing cytokines, IL-1 and IL-6, were found to be elevated in this oxidative stress rat model, indicating inflammation. This was accompanied by osteoporotic changes as indicated by the measurement of bone markers and histomorphometric parameters [34]. The elevation of cytokines was probably achieved through the activation of cytokine-encoding genes like STAT3 or nuclear factor-kappaB by the free radicals [74, 75]. Therefore, there exist relationships between free radicals, inflammation, and bone loss which can lead to osteoporosis. When vitamin E in the form of tocotrienols and α-tocopherol were supplemented to these rats, IL-1 and IL-6 elevations were suppressed. Concurrent with this, the osteoporotic changes were also inhibited [34, 71, 76]. Therefore, there is a possibility that vitamin E, a potent antioxidant, has prevented free radicals from causing inflammation and osteoporosis. Tocotrienols seemed to be more superior than α-tocopherol in suppressing proinflammatory cytokines in the FeNTA rat model and in protecting their bone against osteoporosis [34]. Both the tocopherol and tocotrienol may have achieved this by scavenging the free radicals generated by FeNTA before they could activate the monocytes and osteoblasts, cells that produce IL-1 and IL-6.

Cigarette smoking is a modest risk factor for osteoporosis [77]. Nicotine is among the 4,700 chemicals found in the tar phase of cigarette smoke [78]. Nicotine injected into rats can be used as a model for osteoporosis related to smoking. Various animal studies have confirmed the deleterious effects of nicotine on bone remodeling [79–85]. Nicotine inhibited osteoblast activity and growth [86, 87] but stimulated osteoclast activity [83]. Nicotine has also been shown to induce oxidative stress in both *in vitro* and *in vivo* animal studies [88, 89]. Crowley-Weber et al. [90] had reported that other than oxidative stress, nicotine also activated nuclear transcription factor-κB (NF-κB) in the tissues of smokers. The activation of NF-κB-signaling pathway may be the mechanism for bone loss as it is responsible for osteoclast differentiation [76, 91]. Nicotine has been shown to significantly elevate the proinflammatory cytokines IL-1 and IL-6 in rats. Using the same model, tocotrienol was able to prevent nicotine-induced elevation of IL-1 and IL-6, while tocopherol had no significant effects on both cytokines [71]. Tocotrienol was more effective compared to tocopherol in terms of its action on bone resorbing cytokines and therefore was more effective in reducing inflammation and bone loss.

5. Anti-Inflammatory Action of Vitamin E in Prevention of Osteoporosis

Results from studies on cytokines have given us some insight on the mechanisms involved in the protection of vitamin E against osteoporosis. Free radicals are known to activate transcription factor NFκB which leads to the production of bone resorbing cytokines interleukin-1 and interleukin-6. These proinflammatory cytokines were believed to provide the link between inflammation and osteoporosis. Vitamin E may scavenge and neutralize free radicals before they could activate transcription factor NFκB. This was seen in an oxidative stress model (FeNTA model) in which vitamin E had reduced the levels of bone-resorbing cytokines [34]. Alternatively, vitamin E may have prevented the activation of NFκB by enhancing the internal antioxidative enzymes within the bone. This was demonstrated by Maniam et al. [92], whereby vitamin E supplementation reduced the femoral thiobarbituric acid-reactive substance (TBARS) and increased the glutathione peroxidase activity.

Since osteoporosis is associated with inflammation, there is also a possibility that Vitamin E may have some anti-inflammatory action. Yam et al. [93] found that tocotrienol was able to suppress cyclooxygenase-2 (COX-2) expression in RAW 264.7 cells that were exposed to lipopolysaccharide. COX-2 is an inducible enzyme expressed during inflammation. A RAW cell is a macrophage-like cell which transformed into preosteoclasts when RANKL is added. This suggested that vitamin E may act as anti-inflammatory agent in protecting bone against excessive osteoclastic activity. Previous study has shown that aspirin or other nonsteroidal anti-inflammatory drugs (NSAID) inhibited NFκB [94]. Similar

to tocotrienol, these anti-inflammatory drugs inhibit COX-2. As the activation of NFκB is linked to proinflammatory cytokines and inflammation, it further provides evidence of the anti-inflammatory role of tocotrienol in preventing osteoporosis.

Based on the results from the studies above, tocotrienol was more superior than tocopherol in terms of its ability to suppress bone resorbing cytokines. The more superior tocotrienol action may be contributed by its more potent antioxidant property. It has better interaction with lipoprotein in membrane lipids and is uniformly distributed in the membrane layer compared to tocopherol [35, 95]. Tocotrienol was also better at maintaining the antioxidant status within the rat bone compared to tocopherol [92]. Thus, the antiosteoporotic effect of tocotrienol may be partly explained by its anti-inflammatory as well as antioxidative effects.

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References

[1] P. D. Delmas, "Treatment of postmenopausal osteoporosis," *Lancet*, vol. 359, no. 9322, pp. 2018–2026, 2002.

[2] A. J. Yun and P. Y. Lee, "Maldaptation of the link between inflammation and bone turnover may be a key determinant of osteoporosis," *Medical Hypotheses*, vol. 63, no. 3, pp. 532–537, 2004.

[3] D. Mitra, D. M. Elvins, D. J. Speden, and A. J. Collins, "The prevalence of vertebral fractures in mild ankylosing spondylitis and their relationship to bone mineral density," *Rheumatology*, vol. 39, no. 1, pp. 85–89, 2000.

[4] G. Hougeberg, M. C. Lodder, W. F. Lems et al., "Hand cortical bone mass and its associations with radiographic joint damage and fractures in 50–70 year old female patients with rheumatoid arthritis: cross sectional Oslo-Truro-Amsterdam (OSTRA) collaborative study," *Annals of the Rheumatic Diseases*, vol. 63, no. 10, pp. 1331–1334, 2004.

[5] I. E. M. Bultink, W. F. Lems, P. J. Kostense, B. A. C. Dijkmans, and A. E. Voskuyl, "Prevalence of and risk factors for low bone mineral density and vertebral fractures in patients with systemic lupus erythematosus," *Arthritis and Rheumatism*, vol. 52, no. 7, pp. 2044–2050, 2005.

[6] T. R. Mikuls, K. G. Saag, J. Curtis et al., "Prevalence of osteoporosis and osteopenia among African Americans with early rheumatoid arthritis: the impact of ethnic-specific normative data," *Journal of the National Medical Association*, vol. 97, no. 8, pp. 1155–1160, 2005.

[7] C. Franceschi, M. Bonafè, S. Valensin et al., "Inflamm-aging. An evolutionary perspective on immunosenescence," *Annals of the New York Academy of Sciences*, vol. 908, pp. 244–254, 2000.

[8] J. K. Kiecolt-Glaser, K. J. Preacher, R. C. MacCallum, C. Atkinson, W. B. Malarkey, and R. Glaser, "Chronic stress and age-related increases in the proinflammatory cytokine IL-6," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 100, no. 15, pp. 9090–9095, 2003.

[9] K. Ishihara and T. Hirano, "IL-6 in autoimmune disease and chronic inflammatory proliferative disease," *Cytokine and Growth Factor Reviews*, vol. 13, no. 4-5, pp. 357–368, 2002.

[10] A. R. Moschen, A. Kaser, B. Enrich et al., "The RANKL/OPG system is activated in inflammatory bowel diseases and relates to the state or bone loss," *Gut*, vol. 54, no. 4, pp. 479–487, 2005.

[11] N. Saidenberg-Kermanac'h, M. Cohen-Solal, N. Bessis, M. C. De Vernejoul, and M. C. Boissier, "Role for osteoprotegerin in rheumatoid inflammation," *Joint Bone Spine*, vol. 71, no. 1, pp. 9–13, 2004.

[12] K. Ganesan, S. Teklehaimanot, T. H. Tran, M. Asuncion, and K. Norris, "Relationship of C-reactive protein and bone mineral density in community-dwelling elderly females," *Journal of the National Medical Association*, vol. 97, no. 3, pp. 329–333, 2005.

[13] J. R. Arron and Y. Choi, "Bone versus immune system," *Nature*, vol. 408, no. 6812, pp. 535–536, 2000.

[14] J. Lorenzo, "Interactions between immune and bone cells: new insights with many remaining questions," *Journal of Clinical Investigation*, vol. 106, no. 6, pp. 749–752, 2000.

[15] S. L. Teitelbaum, "Bone resorption by osteoclasts," *Science*, vol. 289, no. 5484, pp. 1504–1508, 2000.

[16] D. L. Lacey, E. Timms, H. L. Tan et al., "Osteoprotegerin ligand is a cytokine that regulates osteoclast differentiation and activation," *Cell*, vol. 93, no. 2, pp. 165–176, 1998.

[17] A. Stern, G. A. Laughlin, J. Bergstrom, and E. Barrett-Connor, "The sex-specific association of serum osteoprotegerin and receptor activator of nuclear factor κB legend with bone mineral density in older adults: the Rancho Bernardo study," *European Journal of Endocrinology*, vol. 156, no. 5, pp. 555–562, 2007.

[18] S. A. J. Lloyd, Y. Y. Yuan, P. J. Kostenuik et al., "Soluble RANKL induces high bone turnover and decreases bone volume, density, and strength in mice," *Calcified Tissue International*, vol. 82, no. 5, pp. 361–372, 2008.

[19] W. J. Boyle, W. S. Simonet, and D. L. Lacey, "Osteoclast differentiation and activation," *Nature*, vol. 423, no. 6937, pp. 337–342, 2003.

[20] R. Pacifici, "Estrogen, cytokines, and pathogenesis of postmenopausal osteoporosis," *Journal of Bone and Mineral Research*, vol. 11, no. 8, pp. 1043–1051, 1996.

[21] S. C. Manolagas and R. L. Jilka, "Mechanisms of disease: bone marrow, cytokines, and bone remodeling - Emerging insights into the pathophysiology of osteoporosis," *New England Journal of Medicine*, vol. 332, no. 5, pp. 305–311, 1995.

[22] K. Fuller, B. Wong, S. Fox, Y. Choi, and T. J. Chambers, "TRANCE is necessary and sufficient for osteoblast-mediated activation of bone resorption in osteoclasts," *Journal of Experimental Medicine*, vol. 188, no. 5, pp. 997–1001, 1998.

[23] R. Pacifici, "Cytokines and osteoclast activity," *Calcified Tissue International*, vol. 56, no. 1, pp. S27–S28, 1995.

[24] R. L. Jilka, "Cytokines, bone remodeling, and estrogen deficiency: a 1998 update," *Bone*, vol. 23, no. 2, supplement 1, pp. 75–81, 1998.

[25] K. Matsuzaki, N. Udagawa, N. Takahashi et al., "Osteoclast differentiation factor (ODF) induces osteoclast-like cell formation in human peripheral blood mononuclear cell cultures," *Biochemical and Biophysical Research Communications*, vol. 246, no. 1, pp. 199–204, 1998.

[26] M. E. Cohen-Solal, F. Boitte, O. Bernard-Poenaru et al., "Increased bone resorbing activity of peripheral monocyte culture supernatants in elderly women," *Journal of Clinical*

Endocrinology and Metabolism, vol. 83, no. 5, pp. 1687–1690, 1998.

[27] J. Pfeilschifter, C. Chenu, A. Bird, G. R. Mundy, and G. D. Roodman, "Interleukin-1 and tumor necrosis factor stimulate the formation of human osteoclastlike cells in vitro," *Journal of Bone and Mineral Research*, vol. 4, no. 1, pp. 113–118, 1989.

[28] M. Kanatani, T. Sugimoto, M. Fukase, and K. Chihara, "Role of interleukin-6 and prostaglandins in the effect of monocyte-conditioned medium on osteoclast formation," *American Journal of Physiology*, vol. 267, no. 6, pp. E868–E876, 1994.

[29] R. Pacifici, "Cytokines and osteoclast activity," *Calcified Tissue International*, vol. 56, no. 1, supplement 1, pp. S27–S28, 1995.

[30] T. Suda, N. Udagawa, I. Nakamura, C. Miyaura, and N. Takahashi, "Modulation of osteoclast differentiation by local factors," *Bone*, vol. 17, no. 2, pp. S87–S91, 1995.

[31] R. Kitazawa, R. B. Kimble, J. L. Vannice, V. T. Kung, and R. Pacifici, "Interleukin-1 receptor antagonist and tumor necrosis factor binding protein decrease osteoclast formation and bone resorption in ovariectomized mice," *Journal of Clinical Investigation*, vol. 94, no. 6, pp. 2397–2406, 1994.

[32] T. Kasama, K. Kobayashi, T. Fukushima et al., "Production of interleukin 1-like factor from human peripheral blood monocytes and polymorphonuclear leukocytes by superoxide anion: the role of interleukin 1 and reactive oxygen species in inflamed sites," *Clinical Immunology and Immunopathology*, vol. 53, no. 3, pp. 439–448, 1989.

[33] S. Devaraj, D. Li, and I. Jialal, "The effects of alpha tocopherol supplementation on monocyte function: decreased lipid oxidation, interleukin 1β secretion, and monocyte adhesion to endothelium," *Journal of Clinical Investigation*, vol. 98, no. 3, pp. 756–763, 1996.

[34] N. S. Ahmad, B. A. K. Khalid, D. A. Luke, and S. I. Nirwana, "Tocotrienol offers better protection than tocopherol from free radical-induced damage of rat bone," *Clinical and Experimental Pharmacology and Physiology*, vol. 32, no. 9, pp. 761–770, 2005.

[35] E. Serbinova, V. Kagan, D. Han, and L. Packer, "Free radical recycling and intramembrane mobility in the antioxidant properties of alpha-tocopherol and alpha-tocotrienol," *Free Radical Biology and Medicine*, vol. 10, no. 5, pp. 263–275, 1991.

[36] Q. Jiang and B. N. Ames, "γ-tocopherol, but not α-tocopherol, decreases proinflammatory eicosanoids and inflammation damage in rats," *FASEB Journal*, vol. 17, no. 8, pp. 816–822, 2003.

[37] A. Patel, F. Liebner, T. Netscher, K. Mereiter, and T. Rosenau, "Vitamin E chemistry. Nitration of non-α-tocopherols: products and mechanistic considerations," *Journal of Organic Chemistry*, vol. 72, no. 17, pp. 6504–6512, 2007.

[38] M. G. Traber and H. Sies, "Vitamin E in humans: demand and delivery," *Annual Review of Nutrition*, vol. 16, pp. 321–347, 1996.

[39] R. Brigelius-Flohé, "Vitamin E and drug metabolism," *Biochemical and Biophysical Research Communications*, vol. 305, no. 3, pp. 737–740, 2003.

[40] U. Singh and S. Devaraj, "Vitamin E: inflammation and atherosclerosis," *Vitamins and Hormones*, vol. 76, pp. 519–549, 2007.

[41] S. Devaraj, R. Tang, B. Adams-Huet et al., "Effect of high-dose α-tocopherol supplementation on biomarkers of oxidative stress and inflammation and carotid atherosclerosis in patients with coronary artery disease," *American Journal of Clinical Nutrition*, vol. 86, no. 5, pp. 1392–1398, 2007.

[42] Q. Jiang, X. Yin, M. A. Lil, M. L. Danielson, H. Freiser, and J. Huang, "Long-chain carboxychromanols, metabolites of vitamin E, are potent inhibitors of cyclooxygenases," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 51, pp. 20464–20469, 2008.

[43] C. S. Yang, G. Lu, J. Ju, and G. X. Li, "Inhibition of inflammation and carcinogenesis in the lung and colon by tocopherols," *Annals of the New York Academy of Sciences*, vol. 1203, pp. 29–34, 2010.

[44] S. K. Clinton and P. Libby, "Cytokines and growth factors in atherogenesis," *Archives of Pathology and Laboratory Medicine*, vol. 116, no. 12, pp. 1292–1300, 1992.

[45] R. Pacifici, L. Rifas, and S. Teitelbaum, "Spontaneous release of interleukin 1 from human blood monocytes reflects bone formation in idiopathic osteoporosis," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 84, no. 13, pp. 4616–4620, 1987.

[46] S. H. Ralston, "Analysis of gene expression in human bone biopsies by polymerase chain reaction: evidence for enhanced cytokine expression in postmenopausal osteoporosis," *Journal of Bone and Mineral Research*, vol. 9, no. 6, pp. 883–890, 1994.

[47] M. Kawano, I. Yamamoto, K. Iwato et al., "Interleukin-1 beta rather than lymphotoxin as the major bone resorbing activity in human multiple myeloma," *Blood*, vol. 73, no. 6, pp. 1646–1649, 1989.

[48] S. J. Hopkins, M. Humphreys, and M. I. V. Jayson, "Cytokines in synovial fluid. I. The presence of biologically active and immunoreactive IL-1," *Clinical and Experimental Immunology*, vol. 72, no. 3, pp. 422–427, 1988.

[49] N. Al Saffar and P. A. Revell, "Interleukin-1 production by activated macrophages surrounding loosened orthopaedic implants: a potential role in osteolysis," *British Journal of Rheumatology*, vol. 33, no. 4, pp. 309–316, 1994.

[50] B. F. Boyce, T. B. Aufdemorte, I. R. Garrett, A. J. P. Yates, and G. R. Mundy, "Effects of interleukin-1 on bone turnover in normal mice," *Endocrinology*, vol. 125, no. 3, pp. 1142–1150, 1989.

[51] M. Gowen, D. D. Wood, and E. J. Ihrie, "An interleukin 1 like factor stimulates bone resorption in vitro," *Nature*, vol. 306, no. 5941, pp. 378–380, 1983.

[52] T. Suda, N. Takahashi, and T. J. Martin, "Modulation of osteoclast differentiation," *Endocrine Reviews*, vol. 13, no. 1, pp. 66–80, 1992.

[53] B. M. Thomson, J. Saklatvala, and T. J. Chambers, "Osteoblasts mediate interleukin 1 stimulation of bone resorption by rat osteoclasts," *Journal of Experimental Medicine*, vol. 164, no. 1, pp. 104–112, 1986.

[54] L. X. Xu, T. Kukita, Y. Nakano et al., "Osteoclasts in normal and adjuvant arthritis bone tissues express the mRNA for both type I and II interleukin-1 receptors," *Laboratory Investigation*, vol. 75, no. 5, pp. 677–687, 1996.

[55] H. Yu and J. Ferrier, "Interleukin-1 alpha induces a sustained increase in cytosolic free calcium in cultured rabbit osteoclasts," *Biochemical and Biophysical Research Communications*, vol. 191, no. 2, pp. 343–350, 1993.

[56] E. Jimi, T. Ikebe, N. Takahashi, M. Hirata, T. Suda, and T. Koga, "Interleukin-1α activates an NF-κB-like factor in osteoclast-like cells," *Journal of Biological Chemistry*, vol. 271, no. 9, pp. 4605–4608, 1996.

[57] E. Jimi, I. Nakamura, T. Ikebe, S. Akiyama, N. Takahashi, and T. Suda, "Activation of NF-κB is involved in the survival of osteoclasts promoted by interleukin-1," *Journal of Biological Chemistry*, vol. 273, no. 15, pp. 8799–8805, 1998.

[58] R. Pacifici, C. Brown, E. Puscheck et al., "Effect of surgical menopause and estrogen replacement on cytokine release from human blood mononuclear cells," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 88, no. 12, pp. 5134–5138, 1991.

[59] F. Sato, Y. Ouchi, A. Masuyama et al., "Effects of estrogen replacement on insulin-like growth factor I concentrations in serum and bone tissue and on interleukin 1 secretion from spleen macrophages in oophorectomized rats," *Calcified Tissue International*, vol. 53, no. 2, pp. 111–116, 1993.

[60] R. Pacifici, L. Rifas, R. McCracken et al., "Ovarian steroid treatment blocks a postmenopausal increase in blood monocyte interleukin 1 release," *Proceedings of the National Academy of Sciences of the United States of America*, vol. 86, no. 7, pp. 2398–2402, 1989.

[61] R. B. Kimble, J. L. Vannice, D. C. Bloedow et al., "Interleukin-1 receptor antagonist decreases bone loss and bone resorption in ovariectomized rats," *Journal of Clinical Investigation*, vol. 93, no. 5, pp. 1959–1967, 1994.

[62] S. Devaraj and I. Jialal, "The effects of alpha-tocopherol on critical cells in atherogenesis," *Current Opinion in Lipidology*, vol. 9, no. 1, pp. 11–15, 1998.

[63] S. C. Manolagas, "Role of cytokines in bone resorption," *Bone*, vol. 17, no. 2, pp. 635–675, 1995.

[64] G. Girasole, R. L. Jilka, G. Passeri et al., "17β-Estradiol inhibits interleukin-6 production by bone marrow-derived stromal cells and osteoblasts in vitro: a potential mechanism for the antiosteoporotic effect of estrogens," *Journal of Clinical Investigation*, vol. 89, no. 3, pp. 883–891, 1992.

[65] P. M. J. McSheehy and T. J. Chambers, "Osteoblast-like cells in the presence of parathyroid hormone release soluble factor that stimulates osteoclastic bone resorption," *Endocrinology*, vol. 119, no. 4, pp. 1654–1659, 1986.

[66] N. Kurihara, D. Bertolini, T. Suda, Y. Akiyama, and G. D. Roodman, "IL-6 stimulates osteoclast-like multinucleated cell formation in long term human marrow cultures by inducing IL-1 release," *Journal of Immunology*, vol. 144, no. 11, pp. 4226–4230, 1990.

[67] D. A. Papanicolaou and A. N. Vgontzas, "Interleukin-6: the endocrine cytokine," *Journal of Clinical Endocrinology and Metabolism*, vol. 85, no. 3, pp. 1331–1333, 2000.

[68] R. H. Straub, H. W. Hense, T. Andus, J. Schölmerich, G. A. J. Riegger, and H. Schunkert, "Hormone replacement therapy and interrelation between serum interleukin-6 and body mass index in postmenopausal women: a population-based study," *Journal of Clinical Endocrinology and Metabolism*, vol. 85, no. 3, pp. 1340–1344, 2000.

[69] A. S. Baldwin, "Control of oncogenesis and cancer therapy resistance by the transcription factor NF-κB," *Journal of Clinical Investigation*, vol. 107, no. 3, pp. 241–246, 2001.

[70] D. Maggio, M. Barabani, M. Pierandrei et al., "Marked decrease in plasma antioxidants in aged osteoporotic women: results of a cross-sectional study," *Journal of Clinical Endocrinology and Metabolism*, vol. 88, no. 4, pp. 1523–1527, 2003.

[71] M. Norazlina, Y. M. K. Nik-Farideh, A. Arizi, A. Faisal, and S. Ima-Nirwana, "Effects of nicotine on bone resorbing cytokines in male rats," *International Medical Journal*, vol. 3, no. 2, pp. 1–9, 2004.

[72] M. Awai, M. Narasaki, Y. Yamanoi, and S. Seno, "Induction of diabetes in animals by parenteral administration of ferric nitrilotriacetate A model of experimental hemochromatosis," *American Journal of Pathology*, vol. 95, no. 3, pp. 663–673, 1976.

[73] T. Kawabata, M. Awai, and M. Kohno, "Generation of active oxygen species by iron nitrilotriacetate (Fe-NTA)," *Acta Medica Okayama*, vol. 40, no. 3, pp. 163–173, 1986.

[74] M. Carballo, M. Conde, R. El Bekay et al., "Oxidative stress triggers STAT3 tyrosine phosphorylation and nuclear translocation in human lymphocytes," *Journal of Biological Chemistry*, vol. 274, no. 25, pp. 17580–17586, 1999.

[75] F. L. J. Visseren, M. S. A. Verkerk, T. Van Der Bruggen, J. J. M. Marx, B. S. Van Asbeck, and R. J. A. Diepersloot, "Iron chelation and hydroxyl radical scavenging reduce the inflammatory response of endothelial cells after infection with Chlamydia pneumoniae or influenza A," *European Journal of Clinical Investigation*, vol. 32, no. 1, pp. 84–90, 2002.

[76] H. Hermizi, O. Faizah, S. I. Nirwana, S. A. Nazrun, D. A. Luke, and M. Norazlina, "Nicotine impaired bone histomorphometric parameters and bone remodeling biomarkers in Sprague–Dawley male rats," *Annals of Microscopy*, vol. 7, pp. 10–24, 2007.

[77] P. O. Ill and C. Alexandre, "Tobacco as risk factor of osteoporosis, myth or reality?" *Revue du Rhumatisme*, vol. 60, no. 4, pp. 280–286, 1993.

[78] D. Hoffmann, I. Hoffmann, and K. El-Bayoumy, "The less harmful cigarette: a controversial issue. A tribute to Ernst L. Wynder," *Chemical Research in Toxicology*, vol. 14, no. 7, pp. 767–790, 2001.

[79] P. D. Broulik, J. Rosenkrancová, P. Růžička, R. Sedláček, and I. Kurcová, "The effect of chronic nicotine administration on bone mineral content and bone strength in normal and castrated male rats," *Hormone and Metabolic Research*, vol. 39, no. 1, pp. 20–24, 2007.

[80] A. Riesenfeld, "Growth-depressing effects of alcohol and nicotine in two strains of rats," *Acta Anatomica*, vol. 122, no. 1, pp. 18–24, 1985.

[81] P. D. Broulik and J. Jarab, "The effect of chronic nicotine administration on bone mineral content in mice," *Hormone and Metabolic Research*, vol. 25, no. 4, pp. 219–221, 1993.

[82] J. A. Yee, L. Yan, D. M. Cullen, M. P. Akhter, and R. R. Recker, "Nicotine inhibits osteoblast differentiation in cultures of neonatal rat calvarial cells," *Journal of Bone and Mineral Research*, vol. 14, supplement 1, pp. S23–S29, 1999.

[83] C. L. Henemyre, D. K. Scales, S. D. Hokett et al., "Nicotine stimulates osteoclast resorption in a porcine marrow cell model," *Journal of Periodontology*, vol. 74, no. 10, pp. 1440–1446, 2003.

[84] A. R. Kamer, N. El-Ghorab, N. Marzec, J. E. Margarone, and R. Dziak, "Nicotine induced proliferation and cytokine release in osteoblastic cells," *International Journal of Molecular Medicine*, vol. 17, no. 1, pp. 121–127, 2006.

[85] L.W. Zheng, L. K. Ma, and L. K. Cheung, "Effects of nicotine on mandibular distraction osteogenesis: a radiological and immunohistochemical study," *European Cellular Material*, vol. 13, supplement 2, p. 54, 2007.

[86] M. A. Fang, P. J. Frost, A. Iida-Klein, and T. J. Hahn, "Effects of nicotine on cellular function in UMR 106-01 osteoblast-like cells," *Bone*, vol. 12, no. 4, pp. 283–286, 1991.

[87] W. K. Ramp, L. G. Lenz, and R. J. S. Galvin, "Nicotine inhibits collagen synthesis and alkaline phosphatase activity, but stimulates DNA synthesis in osteoblast-like cells," *Proceedings of the Society for Experimental Biology and Medicine*, vol. 197, no. 1, pp. 36–43, 1991.

[88] G. J. Wetscher, M. Bagchi, D. Bagchi et al., "Free radical production in nicotine treated pancreatic tissue," *Free Radical Biology and Medicine*, vol. 18, no. 5, pp. 877–882, 1995.

[89] C. Kalpana and V. P. Menon, "Protective effect of curcumin on circulatory lipid peroxidation and antioxidant status during nicotine-induced toxicity," *Toxicology Mechanisms and Methods*, vol. 14, no. 6, pp. 339–343, 2004.

[90] C. L. Crowley-Weber, K. Dvorakova, C. Crowley et al., "Nicotine increases oxidative stress, activates NF-κB and GRP78, induces apoptosis and sensitizes cells to genotoxic/xenobiotic stresses by a multiple stress inducer, deoxycholate: relevance to colon carcinogenesis," *Chemico-Biological Interactions*, vol. 145, no. 1, pp. 53–66, 2003.

[91] E. Jimi and S. Ghosh, "Role of nuclear factor-κB in the immune system and bone," *Immunological Reviews*, vol. 208, pp. 80–87, 2005.

[92] S. Maniam, N. Mohamed, A. N. Shuid, and I. N. Soelaiman, "Palm tocotrienol exerted better antioxidant activities in bone than α-tocopherol," *Basic and Clinical Pharmacology and Toxicology*, vol. 103, no. 1, pp. 55–60, 2008.

[93] M. L. Yam, S. R. Abdul Hafid, H. M. Cheng, and K. Nesaretnam, "Tocotrienols suppress proinflammatory markers and cyclooxygenase-2 expression in RAW264.7 macrophages," *Lipids*, vol. 44, no. 9, pp. 787–797, 2009.

[94] Y. Yamamoto and R. B. Gaynor, "Therapeutic potential of inhibition of the NF-κB pathway in the treatment of inflammation and cancer," *Journal of Clinical Investigation*, vol. 107, no. 2, pp. 135–142, 2001.

[95] C. Suarna, R. L. Hood, R. T. Dean, and R. Stocker, "Comparative antioxidant activity of tocotrienols and other natural lipid-soluble antioxidants in a homogeneous system, and in rat and human lipoproteins," *Biochimica et Biophysica Acta*, vol. 1166, no. 2-3, pp. 163–170, 1993.