Current Concepts Review

Oxidative Stress and Osteoporosis

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- Oxidative stress has been implicated as a causative factor in many disease states, possibly including the diminished bone mineral density in osteoporosis.
- Understanding the effects of oxidative stress on the development of osteoporosis may lead to further research improving preventative and therapeutic measures that can combat this important contributor to morbidity and mortality worldwide.
- A diet rich in whole plant foods with high antioxidant content along with antioxidant-preserving lifestyle changes may improve bone mineral density and reduce the risk of fragility-related fractures. While it is not explicitly clear if antioxidant activity is the effector of this change, the current evidence supports this possibility.
- Supplementation with isolated antioxidants may also provide some osteoprotective benefits, but whole plant food-derived antioxidants potentially have more overall benefits. Larger-scale clinical trials are needed to give credence to definitive clinical recommendations.

This review introduces orthopaedic surgeons to the growing body of literature on oxidative stress, which relates not only to osteoporosis but also likely to many other aspects of orthopaedic surgery.

Osteoporosis

The term *osteoporosis* refers to a reduction in bone quantity or mass that typically does not affect the overall composition of bone. The progressive reduction in bone mass leads to a progressive reduction in bone strength. The normal coordinated action of osteoclasts and osteoblasts is uncoupled in osteoporosis and is thought to contribute to the pathophysiology.

Critically low bone content and/or mass leads to a substantial risk of fractures from even very-low-energy trauma. Osteoporotic fragility fractures represent a considerable burden in the United States. In a 10-year study, Singer et al.[1] reported that, in women who are ≥55 years old, osteoporotic fractures are associated with a higher hospitalization rate and cost burden than myocardial infarction, stroke, or breast cancer. Osteoporosis and its complications contribute considerably to morbidity and mortality in the U.S. and remain a substantial global health concern[2]. Worldwide, it is estimated that 1 in 3 women and 1 in 5 men who are >50 years old will experience an osteoporotic fracture in their lifetime[3,4]. With even a modest 10% decrease in bone mass, the risk of vertebral body and hip fractures can more than double[5]. Osteoporosis is often measured by a dual x-ray absorptiometry (DXA) scan, which involves analyzing the bone density of the lumbar spine and proximal part of the femur, the most common areas of osteoporosis-related fragility fractures[2].

Traditional teaching points to a multifactorial imbalance of hormonal changes, calcium and vitamin-D deficiency, and normal aging as the main causative factors of osteoporosis. Oxidative stress as an additional important contributor is also starting to be borne out in the literature. Recent studies have implicated oxidative stress as 1 possible culprit leading to the uncoupling of osteoblast and osteoclast function seen in osteoporosis[6-9].

Oxidative Stress

The term *oxidative stress* most commonly refers to an overabundance of free radicals, namely, reactive oxygen species (ROS) and reactive nitrogen species (RNS). An imbalance between these free radical oxidants and the antioxidant

Disclosure: The authors indicated that no external funding was received for any aspect of this work. The **Disclosure of Potential Conflicts of Interest** forms are provided with the online version of the article (http://links.lww.com/JBJS/G503).

defenses of the cells causes cellular damage. Damage from free radical molecules like superoxide (O_2^-) starts chain reactions affecting cellular contents including DNA, proteins, cell wall lipoproteins, and other structures. In addition, nuclear factor kappa-B (NF-κB) and other pathways are activated, producing downstream cytokines (tumor necrosis factor [TNF]-alpha, interleukin [IL]-6, and others). Single cellular pathways are initially influenced; however, with ongoing or increased oxidants, entire physiologic systems can be affected. Changes in the cell and its organelles alter cellular function through inhibition or activation of various cellular pathways. These changes cause inflammation (via IL-4, IL-6, TNF-alpha, and others), inhibit differentiation of progenitor cells, alter vital cellular functions, and even cause mitochondria-initiated cell death and apoptosis, and when severe, lead to end organ damage[10]. The mitochondrial theory of aging suggests that ongoing cumulative oxidative stress promotes disease states and premature aging[11]. Since the initial presentation of this mitochondrial theory of aging, a huge body of literature has implicated oxidative stress as a causative factor in many chronic diseases, including osteoporosis[10,12-16].

Sources of Free Radicals

The most common sources of ROS are found in the mitochondria of the cell (complexes I and III)[17]. During electron transport in the mitochondria of the cell, an oxygen molecule (O_2) combines with 4 electrons and 4 protons to make 2 water molecules. Approximately 1% to 5% of the time, this transfer produces a superoxide ion (O_2^-), or less commonly a hydroxide (OH^-) or peroxide ion (O_2^{2-}), each a reactive free radical in ROS form. Under normal cellular conditions, these ROS are neutralized by the antioxidant enzymes superoxide dismutase (SOD) and peroxidase into less reactive molecules of hydrogen peroxide (H_2O_2) and then to water (H_2O).

ROS are also produced in other cellular pathways including the NADP-NADPH (nicotinamide adenine dinucleotide phosphate) cycle used in neutrophils for the destruction of bacteria and viruses, xanthine oxidase reactions during purine catabolism, nitric oxide synthase reactions in nitric oxide production, cyclooxygenases (COX) in the pathway for prostaglandin production, and cytochrome P450 reactions, among others[17].

Despite their destructive potential, ROS and RNS are necessary for normal cell function as they are involved in cellular messaging regulating proliferation, differentiation, apoptosis, repair processes, and gene transcription and transduction[7,17]. Thus, the goal of normal cellular defenses against ROS and RNS is not to completely eliminate them, but rather to maintain a homeostasis preventing the consequences of uncontrolled oxidative stress.

Sources of Oxidative Stress

Anything that increases the metabolic rate in the cell's mitochondria increases oxidative stress. Detoxifying of toxins or ethanol (by cytochrome P450); environments with a high concentration of oxygen (increased rate of O_2^- production); smoke inhalation; increased concentrations of sodium, heavy metals (including iron), and other toxins; and injury from fractures, surgery, trauma, and infections are a few examples. Radiation from radiographs, solar or cosmic sources, or electromagnetic forces also increases oxidative stress[18]. Even eating foods that do not carry naturally occurring antioxidants increases oxidative stress; the most serious such foods include processed sugars and certain saturated fats[19]. Dehydration, stress, anxiety, and possibly lack of sleep all appear to be further mediums for increasing oxidative stress as well[12,20].

Neutralizing Oxidative Stress

ROS and RNS are neutralized by both intrinsic and extrinsic methods. The intrinsic mechanisms include enzymatic and nonenzymatic processes. The intrinsic enzymatic processes involve antioxidant enzymes like SOD, peroxidases, glutathione reductases, and others. These enzymes are located throughout the cell to protect the integrity of DNA, enzymes, hormones, the cell membrane and wall, and other organelles and structures from ROS damage.

The most common protein in the body, albumin, serves as an abundant and important circulating antioxidant and is an example of an intrinsic nonenzymatic antioxidant. Albumin acts as a powerful scavenger neutralizing both ROS and RNS in the forms of hydroxyl radicals and peroxynitrite ($ONOO^-$)[21]. The disulfide bonds in the albumin are readily available to neutralize these dangerous free radicals. There are other valuable intrinsic nonenzymatic antioxidant scavengers. Estrogen, mostly lacking in postmenopausal women, is also a powerful antioxidant through both direct scavenging and stimulating increased expression of antioxidant enzymes[22]. Furthermore, other endogenous nonenzymatic antioxidants include glutathione (GSH) and alpha-lipoic acid[13]. GSH is the most abundant nonenzymatic antioxidant produced in cells. Changes in GSH homeostasis may be an important contributor to uncontrolled oxidative damage[23]. Each of these endogenous antioxidants helps to maintain a healthy balance of oxidative stress.

The other form of protection against oxidative stress is taking in antioxidants through diet, or extrinsic protection. The sources of antioxidants with the highest ability to fight oxidative stress are plant foods[24]. These foods are commonly compared by their ORAC (oxygen radical absorption capacity) values[25]. The higher the ORAC value, the greater the ability to scavenge oxygen free radicals. Unfortunately, the ORAC value is only a measurement of the food item before metabolism and only measures the ability to scavenge free radicals. There are whole plant foods that have a relatively low ORAC value but have a quite large impact on oxidative stress. Broccoli is one of those relatively low ORAC value foods (2,160 compared with 314,446 for cloves) that provide a much higher oxidative stress impact than the ORAC value would imply. After disruption of the cell walls during food preparation or mastication, sulforaphane is enzymatically produced. Sulforaphane has been shown have a multitude of effects. (1) It activates NF-E2-related factor 2 (Nrf2)-mediated phase-II enzymes. Upregulating Nrf2 induces

the antioxidant response element DNA sequence transcription, encoding for increased endogenous antioxidant enzyme production[1,26-29]. (2) It downregulates NF-κB, which is responsible for some of the production of the oxidative stress cytokines[30,31]. (3) It upregulates GSH production (an endogenous nonenzymatic antioxidant)[32,33]. (4) It increases heat shock proteins, further promoting cell stability and decreasing the likelihood of cellular apoptosis[34,35].

Fiber may also contribute to antioxidant protection. The gut microbiome is becoming an area of increased interest in its relation to disease. It appears that there are pathologic and beneficial bacteria in the microbiome. Fiber not only stimulates growth of the beneficial bacteria but also provides substrates for the production of beneficial short-chain fatty acids such as butyrate. Butyrate has been shown to be a TNF-alpha immunomodulator[36] (decreasing the production of the inflammatory cytokine from oxidative stress) and has been shown to communicate with mitochondria, decreasing NLRP3 (NOD, LRR, and pyrin domain-containing protein 3) inflammasomes[37], which add to inflammation and likely oxidative stress. Processed foods have most of the antioxidants and fiber removed, resulting in a more pro-oxidant (oxidatively stressed) state[38,39].

Antioxidant supplements have been used clinically to combat oxidative stress. There is a substantial amount of literature describing potential uses of vitamins and minerals and their effects as extrinsic and intrinsic defenses against oxidative stress. The results of these studies have been mixed, with some even suggesting potential harm[40,41]. In contrast, whole plant foods combine complex nutrients, fiber, and antioxidants working synergistically in a way that supplements alone cannot replicate[38,39]. Vitamin D, previously thought to be a benefit in osteoporosis because of its mediation of calcium balance, may actually derive some of its benefits from its immunomodulatory effects (modifying the cytokine messaging initiated from free radicals) in oxidative stress[42].

Lifestyle choices may also be very intertwined, having a profound effect on the aforementioned defenses. Sleep, hydration, whole plant food diets, limiting psychological stressors, meditation, and avoiding smoking, alcohol, and other drugs like methamphetamines all support improved natural antioxidant defenses in the body[12,19,20,43]. Additionally, while exercise initially increases oxidative stress, it also stimulates intrinsic oxidative enzymes, producing a net positive effect[44,45].

Bone Metabolic Pathways and Oxidative Stress

It is proposed that oxidative stress affects bone mineral density (BMD) through the pathways of bone metabolism. Namely, oxidative stress can (1) increase osteoclastogenesis, (2) decrease osteoprogenitor differentiation to the osteoblast cell lineage, (3) decrease osteoblast activity, and (4) increase osteoblast and osteocyte apoptosis (Fig. 1).

Increased osteoclastogenesis in oxidative stress results from the receptor activator of NF-κB ligand (RANKL) upregulation and osteoprotegerin (OPG) downregulation. RANKL, an osteoclastic activity activator, and OPG, an osteoclastic activity inhibitor, are critical factors in bone metabolism[13,16,46]. RANKL upregulation and OPG downregulation occur through the Wnt/β-catenin pathway and are mediated by activation of protein kinases (ERK1/2 and JNK)[13]. Whole plant foods and isolated antioxidants have been proposed to counteract bone loss by reducing the expression of RANKL and increasing the expression of OPG. Animal studies have supported this conclusion, as antioxidants have been shown to increase bone density by mitigating the RANKL activation of osteoclastic activity[47,48].

Decreased osteoprogenitor differentiation to the osteoblast cell lineage has been seen with increasing pro-oxidants. Bai et al.[49] discussed the finding that H_2O_2-induced oxidative stress suppresses the osteoblastic differentiation of rabbit bone marrow stromal cells and calvarial osteoblasts. This is manifested as a reduction of differentiation markers, including alkaline phosphatase, type-I collagen, colony-forming unit-osteoprogenitor, and nuclear phosphorylation of Runx2.

Decreased osteoblast activity leads to decreased OPG production. This reduction further changes the overall RANKL-OPG ratio, which is important in maintaining the balance of osteoblastic and osteoclastic activity.

Increased osteoblast and osteocyte apoptosis occur with oxidative stress. With osteocyte death, the cytokines for osteoblastic activity are decreased, leading to further predominance of osteoclastogenesis[50-55]. Osteoclastogenesis is also stimulated by the dead and dying osteocytes. These effects are mitigated by antioxidants such as GSH, N-acetylcysteine, and alpha-lipoic acid[7,13].

Thus, the loss of coupling of osteoblast and osteoclast function is likely from oxidative stress-induced changes.

Markers of Oxidative Stress and BMD: Population Studies

Numerous clinical studies have supported the theory that oxidative stress affects BMD; these are summarized in Table I. Although confounding variables must be considered, a comparison of blood and urine markers of oxidative stress or markers of endogenous enzyme activity with changes in BMD provides a reasonable tool to evaluate the effects of oxidative stress on bone. Measures of BMD by DXA or quantitative ultrasound have proven to be an effective means to evaluate BMD[56,57]. Markers for oxidative stress include advanced oxidation protein products (AOPP; proteins that have been oxidized from free radicals); malondialdehyde (MDA; a previous marker for osteoporosis); the ratio of reduced GSH to oxidized GSH (GSH/GSSG); total oxidant state; and downstream cytokines, including IL-6, IL-4, TNF-alpha, and many others. Novel methods for the measurement of total sample oxidant and antioxidant status have also been developed, providing a means to measure an oxidative stress index[58]. These studies have provided supporting evidence that oxidative stress is an independent risk factor for osteoporosis and that osteoclastic and osteoblastic activity may be affected by an imbalance in the oxidant and antioxidant status in postmenopausal osteoporosis[7,59-63]. They have also suggested that

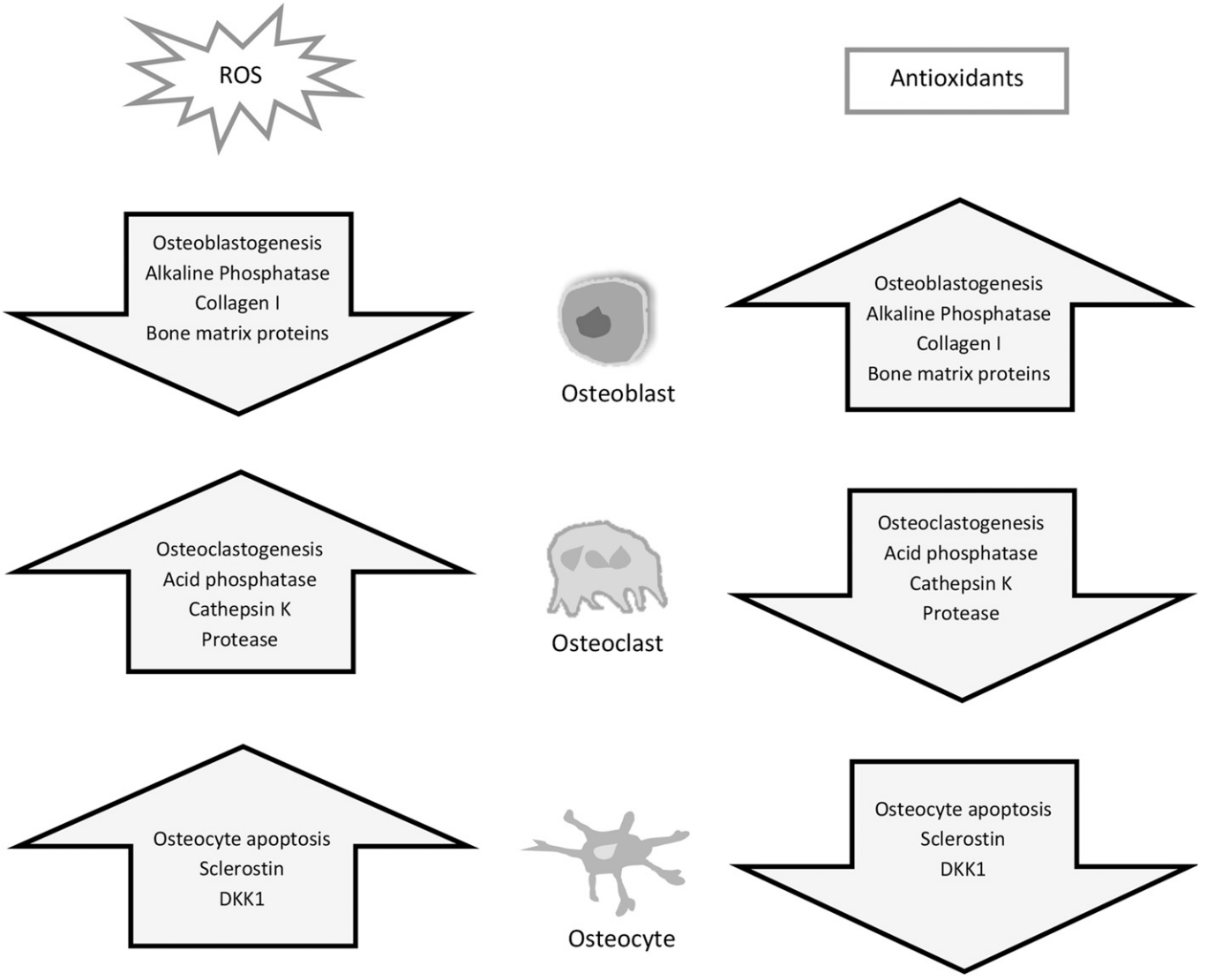

Fig. 1

Oxidative stress in bone remodeling. ROS = reactive oxygen species, and DKK1 = Dickkopf1 gene.

menopause-related estrogen withdrawal may make bone more vulnerable to oxidative injury, increasing the risk of postmenopausal osteoporosis[64].

Increased iron stores have also been linked to oxidative stress through associations with oxidative stress markers, including urinary 8-OHdG (8-hydroxy-2′-deoxyguanosine), AOPPs, and oxidized low-density lipoprotein[26,65,66]. Zwart et al.[27] evaluated blood and urine samples of 23 crew members of the International Space Station. They found that serum ferritin and body iron increased early in flight through the mobilization of iron to storage tissues. There was an inverse relationship between ferritin during flight and BMD in the hip and pelvis after flight. The relationship of the ferritin to the disuse phenomenon associated with the near-zero gravitational stresses on the space crew members is not clear.

Increasing Antioxidant Defenses as Preventative and Therapeutic Intervention to Improve BMD: Population Studies

If neutralizing oxidative stress can improve bone density, the prospect of clinical application as preventative and therapeutic interventions is raised to either decrease oxidative stress or increase antioxidant defenses. Clinicians routinely make recommendations that decrease oxidative stress, including the avoidance of tobacco, alcohol, and illicit drugs. They also encourage healthy habits such as maintaining good sleep routines, staying well hydrated, and utilizing safety features such as seatbelts and helmets to avoid trauma. What recommendations can clinicians then make regarding increasing antioxidant defenses?

An obvious answer is to increase antioxidant intake. Given the intrinsic value of antioxidant-rich foods, much

TABLE I Markers of Oxidative Stress and BMD: Population Studies

Study	Type of Study	Level of Evidence	Oxidative stress Marker	No. of Patients			BMD Comparison	P Value
				Case	Control	Total		
Mangiafico et al.[60] (2007)	Case-control	III	Serum 8-iso-PGF(2alpha)	173	152	325	Femoral neck	<0.0001
							Lumbar spine	<0.0001
Altindag et al.[59] (2008)	Case-control	III	Plasma hydrogen peroxide equivalents	39	26	65	Lumbar spine	<0.001
							Femoral neck	0.018
Maggio et al.[61] (2003)	Case-control	III	Plasma vitamins A, C, and E and enzymatic activity of GPx and SOD	75	75	150	Femoral neck	<0.001
Sánchez-Rodríguez et al.[62] (2007)	Case-control	III	Plasma antioxidant activity of SOD and GPx	44	50	94	Calcaneus	0.034
Wu et al.[63] (2015)	Case-control	III	Plasma AOPPs	60	60	120	Lumbar spine	<0.001
Cervellati et al.[64] (2013)	Case-control	III	Serum lipid hydroperoxides	93	98	191	Lumbar spine	0.012
Zwart et al.[27] (2013)	Cross-sectional	IV	Urinary 8OHdG and plasma AOPPs	–	–	23	Total hip	0.031
							Trochanter	0.006
							Femoral neck	0.044
							Pelvis	0.049
Basu et al.[84] (2001)	Cross-sectional	IV	Urinary 8-iso-PGF(2alpha)	–	–	101	Lumbar spine	0.04
							Whole body	0.001

*PGF = prostaglandin, GPx = glutathione peroxidase, SOD = superoxide dismutase, and AOPP = advanced oxidation protein products

research has been done to extract the useful components such as vitamins and minerals to prepare therapeutic and preventative pharmaceuticals to manage oxidative stress and its effects on disease states. There have been mixed results as to the efficacy of vitamin and mineral supplements compared with the consumption of natural antioxidant-rich foods. Disease states associated with oxidative stress, such as cardiovascular disease and diabetes, have, like osteoporosis, had poor results with antioxidant supplements[30]. In contrast, a healthy diet rich in natural antioxidants along with other lifestyle changes like exercise and better sleep hygiene has led to success[32]. This may be due to a synergistic effect when antioxidants are consumed in the form of whole antioxidant-rich plant foods.

There are abundant animal studies supporting the positive effects of various foods on bone density. Some inhibit bone turnover and resorption while others stimulate bone formation[34]. Pomegranate[67,68], legumes[69], and soybeans[70] prevent bone loss, stimulate osteoblastic differentiation, and decrease inflammation. Higher intake of polyphenols found in berries also leads to increased bone mass[71]. Alpha lipoic acid promotes osteoblast formation and improved bone density[72]. Carnosic acid[73] found in sage and rosemary has been shown to reduce RANKL-induced oxidative stress and osteoclastogenesis, and the areca nut[74] has been shown to inhibit release of ROS that stimulate bone resorption. These animal and in vitro studies are encouraging, but not definitive in their findings as an application for human bone density.

Human studies have also provided a litany of research that supports the effects of antioxidants on bone density. Fruit and vegetable consumption is an independent predictor of bone mass in early childhood through the effects that regular consumption has on peak bone mass. Tylavsky et al.[75], when controlling for age, body mass index, and physical activity, found that children who consumed ≥3 servings of fruit and vegetables per day had more whole-body bone area, lower urinary calcium, and lower parathyroid hormone than those consuming <3 servings per day.

An epidemiologic study by Warensjö Lemming et al.[76] that evaluated 56,736 women found that a healthy diet consisting of more vegetables, fruits, cereals, fish, and fermented milk than that in a Western or convenience diet consisting of a high intake of sweet snacks, bakery products, sodas, savory snacks, and white bread led to a lower rate of hip fracture. They found that the rate of hip fracture was 31% lower in the highest compared with lowest quartile of those with a healthy dietary pattern. In contrast to the healthy diet, those in the highest quartile compared with the lowest quartile with respect to following the Western or convenience dietary pattern had a 50% higher rate of hip fracture.

Additional human studies have shown lower rates of osteoporosis with antioxidant-rich dietary intake. Regu et al., in a study of 8,022 Korean adults, showed that postmenopausal women in the highest quintile of daily beta-carotene intake had a lower risk of osteopenia at the lumbar spine[77]. They also found that β-cryptoxanthin intake was significantly associated

TABLE II Controlling Oxidative Stress as Preventative and Therapeutic Intervention for Loss of BMD: Whole Foods Population Studies

Study	Type of Study	Level of Evidence	Total No. of Patients	Dietary Whole Food Consumed	Antioxidant Evaluated	Duration	Dependent Variable	P Value
Kaume et al.[85] (2012)	RCT	I	49*	45 g of blackberries or blueberries	Whole foods	9 mo	Whole body BMD	0.0284
Yang et al.[86] (2008)	Case-Control	III	60†	Whole diet	Whole foods and lycopene	72-hr diet recall questionnaire	Lumbar spine BMD and total hip BMD	<0.03
Regu et al.[77] (2017)	Cross-sectional	IV	8,022	Whole diet	Whole foods beta-carotene or beta-cryptoxanthin	24-hr diet recall questionnaire	Total hip BMD	0.036† and 0.026§
Correa Rodríguez et al.[87] (2017)	Cross-sectional	IV	605	Whole diet	Whole diet	72-hr diet recall interview	Calcaneal BMD vs. dietary antioxidant quality	0.035
Rivas et al.[88] (2012)	Cross-sectional	IV	280	Whole diet	Whole foods with vitamin C, selenium, and zinc	24-hr diet recall questionnaire	Calcaneal BMD vs. dietary antioxidant quality	<0.05
Zalloua et al.[89] (2007)	Cross-sectional	IV	12,055	Whole diet	Whole foods with >250 g/wk of seafood	1-page nutritional questionnaire	Whole body BMD	<0.001
					Whole foods with >250 g/wk of fruit	1-page nutritional questionnaire	Whole body BMD	<0.05
New et al.[90] (2000)	Cross-sectional	IV	62	Whole diet	Whole foods rich in fruit	Food frequency questionnaire	Femoral neck BMD	<0.01
Wattanapenpaiboon et al.[91] (2003)	Cross-sectional	IV	205	Whole diet	Whole foods with lycopene	Food frequency questionnaire	Whole body BMD and lumbar spine BMD	<0.05# and <0.05**
Wolf et al.[92] (2005)	Cross-sectional	IV	11,068	Whole diet	Whole foods with vitamins A, C, and E; beta-carotene; and selenium	Food frequency questionnaire	Whole body BMD, hip BMD, and lumbar spine BMD	>0.05
Kim et al.[93] (2016)	Cross-sectional	IV	189	Whole diet	Whole foods with beta-carotene	Food frequency questionnaire	Lumbar spine BMD	<0.001
					Whole foods with vitamin C	Food frequency questionnaire	Femoral neck BMD	0.001
					Whole foods with zinc	Food frequency questionnaire	Total hip BMD	0.035
Kim et al.[94] (2015)	Cross-sectional	IV	1,196	Whole diet	Whole foods, vitamin C	24-hr diet recall questionnaire	Lumbar spine BMD	<0.05
							Total hip BMD	<0.05
Hall and Greendale[95] (1998)	Cross-sectional	IV	775	Whole diet	Whole foods with vitamin C	Food frequency questionnaire	Total hip BMD	0.005††
							Femoral neck BMD	0.002†† and 0.002‡‡

continued

TABLE II (*continued*)

Study	Type of Study	Level of Evidence	Total No. of Patients	Dietary Whole Food Consumed	Antioxidant Evaluated	Duration	Dependent Variable	P Value
Leveille et al.[96] (1997)	Cross-sectional	IV	1,892	Whole diet with or without vitamin-C supplementation	Whole foods with or without vitamin-C supplementation	Food frequency questionnaire and vitamin frequency questionnaire	Total hip BMD	>0.05§§ and 0.01##
Kaptoge et al.[97] (2003)	Cross-sectional	IV	944	Whole diet	Whole foods, vitamin C	7-day food diary, at 2 time points 3 years apart	Total hip BMD	0.016***
Simon and Hudes[98] (2001)	Cross-sectional	IV	13,080	Whole diet	Whole foods, vitamin C	24-hr diet recall questionnaire	Proximal femural BMD	0.001†††
Odai et al.[99] (2019)	Cross-sectional	IV	157	Whole diet	Whole foods with vitamin E	Food frequency questionnaire	Lumbar spine BMD	0.022†‡†
Zhang et al.[100] (2017)	Cross-sectional	IV	989	Whole diet	Whole foods, vitamin E	Food frequency questionnaire	Lumbar spine BMD	0.002§§§
							Femoral neck BMD	0.001§§§
Shi et al.[101] (2016)	Cross-sectional	IV	3,203	Whole diet	Whole foods, vitamin E	Food frequency questionnaire	Lumbar spine BMD	0.022###

*Treatment groups included 6 subjects who received 45 g of blackberries and 13 who received 45 g of blueberries. The control groups included 12 smokers and 18 nonsmokers. Total body BMD increased only in the nonsmoker control group. †There were 31 patients in the treatment group and 29 in the control group. ‡Postmenopausal women who had beta-carotene. §Premenopausal women who had beta-cryptoxanthin. #Men. **Premenopausal women. ††Each 100-mg increment in dietary vitamin-C intake was associated with 0.017-g/cm^2 increment in BMD. ‡‡Assessing the effect modification of dietary calcium, the authors found no relation between BMD and vitamin C at calcium intakes of <500 mg/day; however, for intake of >500 mg, there was an increment of 0.0190 g/cm^2 in BMD per 100 mg of vitamin C consumed. §§No difference between groups. ##Longer duration of vitamin-C supplement use was associated with high BMD in women from 55 to 64 years old. ***Women who consumed <58 mg/day of vitamin C lost bone mass at a faster rate than those who had >58 mg/day. †††In postmenopausal women with a history of smoking and estrogen use, increased serum ascorbic acid was associated with a decreased fracture prevalence. ‡‡‡Premenopausal women only. §§§Significant inverse relationship suggesting harmful effect of vitamin E. ###Women only.

with a lower risk of osteopenia in the total hip. A cross-sectional study of >2,800 Chinese men and women also showed a dose-response positive correlation between circulating levels of the carotenoid β-cryptoxanthin, lycopene, and α-carotene and BMD[78]. Furthermore, a study by Costa-Rodrigues et al.[79] suggested that lycopene may promote an anabolic state of bone metabolism through stimulation of osteoblastogenesis and inhibition of osteoclastogenesis. Similarly, dried plums and almonds appear to improve bone health[80,81].

Population studies evaluating the effects of whole-food diets rich in antioxidants on BMD are summarized in Table II. The evaluation of a whole plant food diet as a variable affecting BMD is wrought with difficulty and potential confounding variables. Most of these studies are limited by cross-sectional design with data collection based on food frequency and diet recall questionnaires. In light of these deficiencies, there remains a strong trend supporting a healthy diet rich in antioxidants as a therapeutic strategy to improve BMD. Large-scale randomized controlled trials (RCTs) are necessary to confirm these results to a higher degree of certainty; however, the challenge and practicality of such a study have thus far limited its realization. Despite the difficulty in evaluating the effect of a whole plant food diet on BMD, it is reasonable to conclude that an antioxidant-rich diet may reduce the risk of osteoporosis.

Antioxidant Supplementation

Notable population studies evaluating the effects of isolated antioxidant supplementation on BMD are listed in Table III. Most of the listed studies evaluate vitamins C and E or isoflavones (found in legumes). All except 1 supported the use of supplementation to improve BMD. Two of the noted studies were large systematic reviews. Wei et al.[82] evaluated the effect of soy isoflavone supplementation on osteoporosis in women. As part of their review, they performed a meta-analysis of 7 RCTs comprising 793 patients. They found that daily ingestion of soy isoflavones for 1 month to 2 years significantly increased BMD. Subgroup analysis revealed a significantly higher weighted mean difference in BMD change for patients who had an isoflavone dose of >75 mg/day.

More recently, in 2017, Lambert et al.[83] performed a systematic review of RCTs evaluating isoflavone supplemental therapies for treating BMD loss in postmenopausal women. Twenty-six total RCTs, comprising 2,652 patients, were

TABLE III Controlling Oxidative Stress as Preventative and Therapeutic Intervention for Loss of BMD: Isolated Antioxidant Supplementation Population Studies*

Study	Type of Study	Level of Evidence	Antioxidant Supplement	Daily Dose *(mean)*	Duration *(mean)*	Dependent Variable	No. of Patients			P Value
							Treatment	Control)	Total	
Ruiz-Ramos et al.[102] (2010)	RCT	I	Vitamin C and vitamin E	500/1,000 mg and 400 IU, respectively	12 mo	Lumbar spine BMD	30/30	30	90	<0.05
Mazzanti et al.[103] (2015)	RCT	I	Vitamins D3, K1, and B6	10 μg, 0.14 mg, and 1.2 mg, respectively	12 mo	BMD in proximal phalanges 2-5 in right hand	30	30	60	<0.05
Mainini et al.[104] (2012)	RCT	I	Alpha-lipoic acid, vitamin C, vitamin E, and selenium	–	12 mo	Heel quantitative ultrasonometry	23	21	44	0.048
Chuin et al.[105] (2009)	RCT	I	Vitamin C and vitamin E	1,000 mg and 600 mg, respectively	6 m	Lumbar spine BMD	8, 11, and 8†	7	34	<0.05
Stunes et al.[106] (2017)	RCT	I	Vitamin C and vitamin E	1,000 mg and 235 mg, respectively	3 mo	Whole body BMD and hip BMD	16	17	33	0.481 and 0.336, respectively
Wei et al.[82] (2012)	Systematic review (7 studies)	I	Isoflavone	47-126 mg	1-24 mo	BMD	399	394	793	<0.001
Lambert et al.[83] (2017)	Systematic review (26 studies)	I	Isoflavone	–	3-24 mo	Lumbar spine BMD and femoral neck BMD	–	–	2,652	<0.00001 and <0.01, respectively
Lambert et al.[107] (2017)	RCT	I	Isoflavone	60 mg	12 mo	Lumbar spine BMD and femoral neck BMD	38	40	78	<0.05 and <0.01, respectively
Hsu et al.[108] (2001)	Cohort study	II	Isoflavone	300 mg	6 mo	Calcaneal BMD	–	–	37	>0.05
Morton et al.[109] (2001)	Cross-sectional	IV	Vitamin C	100-5,000 mg (mean, 745 mg)	Men, 12.4 yr	Femoral neck BMD	277	717	994	<0.02

*RCT = randomized controlled trial. †Eight patients received antioxidants, 11 had exercise and placebo, and 8 had exercise and antioxidants.

TABLE IV Grades of Recommendation for Preventative and Therapeutic Measures

Recommendation	Grade*
A whole-food diet rich in antioxidants including vitamins C and E, beta-carotene, and lycopene to improve BMD	C
Lifestyle changes including regular exercise, good sleep hygiene, staying well hydrated, and avoidance of drug and alcohol use improve BMD	C

*According to Wright[110], grade A indicates good evidence (Level-I studies with consistent findings) for or against recommending intervention; grade B, fair evidence (Level-II or III studies with consistent findings) for or against recommending intervention; grade C, poor-quality evidence (Level-IV or V studies with consistent findings) for or against recommending intervention; and grade I, insufficient or conflicting evidence not allowing a recommendation for or against intervention.

included in the review and meta-analysis. They found that isoflavone therapies were associated with higher BMD in the lumbar spine and the femoral neck. A subset analysis further demonstrated that the osteoprotective effect of isoflavone therapy resulted from aglycones rather than glycosides.

Overview

From the population studies and bone metabolism studies, it is probable that oxidative stress plays a role in the development of postmenopausal osteoporosis. The question of how best to manage the free radical imbalance and the associated downstream cytokines, to normalize the oxidative state and potentially improve BMD and lower the fracture risk, remains.

The data suggest that diets emphasizing whole plant foods in the form of fruits, vegetables, and possibly whole grains, nuts, and legumes, with avoidance of foods that increase oxidative stress, such as processed sugars and some saturated fats, can improve bone density. This gives rise to the therapeutic strategy that adherence to certain dietary guidelines, along with lifestyle choices of exercise, staying well hydrated, good sleep hygiene, and avoidance of drug and alcohol use, may very well have a substantial impact on improving BMD and reducing the risk of related fragility fractures. While these lifestyle choices have been shown to improve general health, many of them have not been adequately studied to prove their use in improving bone health.

The noted studies in Table III support the use of vitamin C and isoflavone supplementation and possibly vitamin E to improve BMD. At present, the data suggest that supplementation with >500 mg daily of vitamin C and >50 mg daily of isoflavones can improve bone density. Further investigations are warranted to conclude that an evidence-based clinical recommendation for a specific therapeutic regimen can be offered.

Should the current methods of managing osteoporosis be abandoned? We think not. The above recommendations are not based on studies that definitively prove that oxidative stress is responsible for osteoporosis, only that it is likely associated. The current scientifically prudent measures would likely include encouraging a diet emphasizing antioxidant-rich whole plant foods and an antioxidant-preserving lifestyle (for general health and likely bone health) along with other traditional therapeutic regimens. Larger-scale clinical trials are needed to give credence to definitive clinical recommendations. The grades of recommendation for preventative and therapeutic measures are summarized in Table IV. ■

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