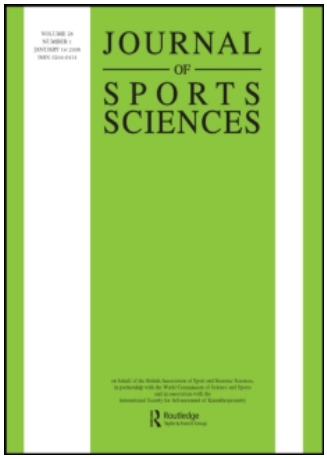

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Effect of antioxidants and exercise on bone metabolism

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Abstract
We examined the effects of antioxidant supplementation in association with progressive aerobic training on the bone metabolism of healthy elderly individuals. For 8 weeks, 13 participants (mean age 74 years) received vitamin C (500 mg) and vitamin E (100 mg) daily and participated in a supervised progressive aerobic training programme. After the 8 weeks, 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D concentrations were increased significantly by 42.8% ($P < 0.001$) and 26.8% ($P < 0.01$) respectively, while parathyroid hormone concentration was decreased by 17.5% ($p < 0.05$). Of the bone markers, only bone alkaline phosphatase decreased, by 14.6% ($P < 0.05$). No variation was observed for ionized calcium, insulin-like growth factor-1 or insulin-like growth factor binding protein-3. Our findings suggest that 8 weeks of combined antioxidant supplementation and aerobic training modified vitamin D metabolism and parathyroid hormone concentration. These adaptations might counterbalance the unfavourable hormonal profile frequently observed in the elderly that predisposes them to accentuated age-related bone loss.

Keywords: *Antioxidant, physical activity, calcium homeostasis, elderly participants, insulin-like growth factor-1*

Introduction

Osteoporosis is defined as a diffuse skeletal disease with reduced bone mineral density and altered bone micro-architecture, leading to increased bone fragility and fracture risk (Consensus Development Conference, 1993). Its pathology is viewed as multi-factorial, with genetic and environmental factors (Fox, Cummings, & Threets, 1994) possibly interacting. Among the environmental influences, nutritional habits (Heanay *et al.*, 1982) and physical inactivity (Nguyen, Center, & Eisman, 2000) seem to play major roles in the development of the disease. Moreover, age-related hormonal deficiencies can be considered aggravating factors (Endres, Morgan, Garry, & Omdahl, 1987).

For the early prevention or inhibition of age-related bone loss, adapted nutritional programmes might be efficient. Various vitamins and minerals play an important role in bone metabolic processes, and calcium and vitamin D supplementation in particular constitutes the most basic preventive strategy (Chapuy *et al.*, 1992). On the other hand, vitamin C, a key antioxidant, was found to be essential for type I collagen matrix synthesis, alkaline phosphatase activity, and matrix mineralization in osteoblast cultures (Franceschi, 1992). Epidemiological studies in adults have reported an association between vitamin C intake or blood levels and self-reported fracture (Simon & Hudes, 2001) and bone mineral density (Morton, Barrett-Connor, & Schneider, 2001; Simon & Hudes, 2001). The same relationship between bone mass and vitamin C was also observed in the forearm of children and adolescents (Gunnes & Lehmann, 1995). Moreover, it has been shown that insufficient dietary intake of vitamins C and E substantially increases the risk of hip fracture in smokers, whereas an adequate intake appears to protect against the adverse effects of smoking on bone health (Melhus, Michaelsson, Holberg, Wolk, & Ljunghall, 1999). More recently, low concentrations of vitamins C and E were reported in elderly osteoporotic women compared with age-matched controls (Maggio *et al.*, 2003).

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These data seem to point out the negative effect of an antioxidant deficit regarding age-related bone loss (Maggio *et al.*, 2003). Although the underlying physiological mechanisms of antioxidant action on bone metabolism are not precisely understood, the data do suggest that an increase in the dietary intake of vitamins C and E may improve bone health in the elderly.

The contribution of load-bearing exercise to the preservation of bone density, and thus to the prevention of osteoporosis, has also received considerable attention. Epidemiological data have shown that high physical activity levels are associated with high bone mineral density at the weight-bearing bone sites in elderly individuals (Nguyen *et al.*, 2000). The favourable effect of physical activity on bone health has also been observed in many longitudinal studies of supervised training programmes (Keer, Morton, Dick, & Prince, 1996; Menkes *et al.*, 1993), with few studies reporting no significant effect (Prince *et al.*, 1991). Based on experimental research (Rubin & Lanyon, 1984), strength-training programmes have generally been chosen for elderly participants (Ryan *et al.*, 1994; Yarasheski, Campbell, & Kohrt, 1997), and a vigorous aerobic and strength-training regimen has been shown to be the most effective (Gutin & Gasper, 1992). However, strenuous load-bearing exercise may increase the risk of injuries and has the inherent disadvantage of lower long-term compliance, expecially in elderly individuals (Kallinen & Markku, 1995). However, a recent study showed that more appropriate and lower-risk training programmes consisting of only aerobic training improved bone mineral density in this type of population (Hurley & Hagberg, 1998). Moreover, endurance activities improve the participant's hormonal profile, as evidenced by increases in 1,25-dihydroxyvitamin D and serum insulin-like growth factor-1, and this may add to the favourable effect of physical exercise on bone (Maïmoun *et al.*, 2004; Zittermann *et al.*, 2000).

The aim of this study was to assess the cumulative effect of supplementation with the antioxidant vitamins C and E and progressive endurance exercise on calcium homeostasis, bone cell activity using peripheral bone biochemical markers, and bone-related hormones in healthy elderly participants.

Materials and methods

Participants

Thirteen healthy elderly individuals (9 women and 4 men) aged 69 – 79 years (mean 74 years) were recruited for the study, all of whom completed the programme. They first underwent a medical screening procedure that included a medical history, a physical examination, and an electrocardiogram. All participants were free of any limiting orthopaedic conditions. Exclusion criteria included intake of vitamin or mineral supplements or any medication known to affect bone metabolism (e.g. testosterone, cortisol), evidence of cardiovascular disease, and diabetes mellitus. None of the participants had been involved in a regular exercise programme for at least 2 years before the study and their habitual physical activity was evaluated by a questionnaire adapted for elderly people (Voorrips, Ravelli, Dongelmans, Deurenberg, & Van Staveren, 1991). The study was approved by the Regional Research Ethics Committee (Languedoc-Roussillon, France) and all participants provided informed written consent.

Experimental design

Each participant received one capsule daily containing 500 mg of vitamin C and another containing 100 mg of vitamin E for the 8-week experimental period (May and June). The supplements used in this experiment were commercially available vitamin C and E (Laroscorbine for Vitamin C and Ephynal for Vitamin E, both from Roche Nicholas, France). The supplements were authorised by the French Agency for Sanitary Affairs for Health Products (AFSSAPS, Agence Francaise de Securite Sanitaire des Produits de Sante). No independent verification of the composition of these supplements was undertaken. During this period, the antioxidant supplementation was associated with an aerobic training programme consisting of 3 one-hour supervised progressive exercise sessions per week (Monday, Wednesday, Friday). The sessions were held in a gymnasium and started with a 10-min standardized warm-up followed by 40 – 48 min of brisk aerobic walking. The exercise intensity was set according to a specific training concept – that is, at a heart rate corresponding to the ventilatory threshold; this was determined during a maximal exercise test presented in detail elsewhere (Maïmoun *et al.*, 2005). The same maximal exercise test was again performed at the end of the 8-week training programme. The exercises were supervised by a coach who monitored and motivated the participants, and each exercise session ended with 10 min of stretching. During the study period, the participants took part in no additional recreational activities and did not modify their lifestyle in any way.

Sample collection

Blood samples were obtained before and at the end of the study period, after 48 h of rest. All blood samples (20 ml) were collected in chilled sterile tubes.

The samples were allowed to clot at room temperature and were then centrifuged at 3000 rev · min^{-1} for 10 min at 4°C. Serum samples were stored at −80°C until analysis and each individual's pre- and post-training samples were analysed in the same assay to reduce inter-assay variability. Concentrations of ionized calcium, intact parathyroid hormone, 25-hydroxyvitamin D, 1,25-dihydroxyvitamin D, osteocalcin, bone alkaline phosphatase, urinary type I collagen C-telopeptide, and insulin-like growth factor-1 (IGF-1) and its major binding protein, IGFBP-3, were determined.

Biochemical assays

Calcium homeostasis. Ionized calcium was measured by an ion-selective electrode (BGE Electrolytes Instrumentation Laboratory, Lexington, MA). Intact parathyroid hormone was measured by an immunoradiometric assay (N-tact® PTH SP Diasorin, Stillwater, MN). The intra- and inter-assay coefficients of variation were 3.6% and 3.4% respectively. 25-Hydroxyvitamin D was measured by radioimmunoassay (25-Hydroxyvitamin D RIA kit, Nichols Institute Diagnostics, Paris, France). The intra- and inter-assay coefficients of variation were 5% and 8.1% respectively. Serum 1,25-dihydroxyvitamin D was measured by radioimmunoassay (1,25-Dihydroxyvitamin D RIA kit, Nichols Institute Diagnostics, Paris, France). The intra- and inter-assay coefficients of variation were 5% and 10.8% respectively.

Bone biochemical markers

Markers of bone formation. Serum osteocalcin was measured by immunoradiometric assay (Elsa-OST-NAT™, CIS Biointernational®, Gif/Yvette, France). The intra- and inter-assay coefficients of variation were below 5%. Serum alkaline phosphatase was measured by immunoradiometric assay (Tandem®-R Ostase® Hybritech, Inc.®, San Diego, CA). The intra- and inter-assay coefficients of variation were less than 7% and 9% respectively.

Marker of bone resorption. Serum urinary type I collagen C-telopeptide was measured by ELISA (CrossLaps™ ELISA, OSTEOMETER A/S®, Rodovre, Denmark). The intra- and inter-assay coefficients of variation were less than 5.7% and 9.4% respectively.

Somatotropic hormones. Insulin-like growth factor-1 was measured by immunoradiometric assay (DSL, Diagnostic Systems Laboratories, Webster, TX), as was insulin-like growth factor binding protein-3 (IGFBP-3 IRMA, Immunotech, Marseille, France).

Statistical analysis

All data are expressed as means and standard deviations (*s*). The distribution of variables was tested by the Shapiro-Wilk statistical method. A non-parametric test for small samples was then used. Differences between baseline data and data after treatment (8 weeks of supplementation and exercise) were tested with the Wilcoxon paired-sample test. Statistical significance was set at $P < 0.05$. SAS software, version 8.2 (SAS Institute, Cary, NC), was used for statistical analysis.

Results

Anthropometric data and parameters of physical fitness

The participants' biometric characteristics and parameters of physical fitness are shown in Table I. Total body mass and the body mass index remained constant throughout the study. No significant changes in maximal oxygen uptake or exercise duration were observed after the study period, although these values tended to increase.

Calcium homeostasis and bone-related hormones

The effects of the supplementation and exercise intervention on the parameters of calcium homeostasis and the bone-related hormones are presented in Table II. 25-Hydroxyvitamin D and 1,25dihydroxyvitamin D increased significantly by 42.8% ($P < 0.001$) and 26.8% ($P < 0.01$) respectively. Concentrations tended towards the upper limit of the normal range after the 8-week intervention. In contrast, intact parathyroid hormone decreased significantly by 17.5% ($P = 0.048$). Antioxidant supplementation associated with exercise had no effect

Table I. Anthropometric and physical fitness parameters at baseline and after 8 weeks of intervention (mean ± *s*).

	Baseline	After 8 weeks of intervention	*P*-value
Anthropometric variables			
Age (years)	73.9 ± 3.8	–	
Height (m)	1.65 ± 0.08	–	
Weight (kg)	66.5 ± 9.7	66.3 ± 10.5	N.S.
BMI (kg · m^{-2})	24.3 ± 2.8	24.3 ± 3.0	N.S.
Parameters of physical fitness			
$\dot{V}O_{2max}$ (ml · min^{-1} · kg^{-1})	27.3 ± 3.6	27.9 ± 3.8	N.S.
Exercise duration (min)	10.7 ± 1.8	11.5 ± 0.5	N.S.

Note: BMI = body mass index; $\dot{V}O_{2max}$ = maximal oxygen uptake; N.S. = no significant difference between baseline values and those after 8 weeks.

on plasma concentration of ionized calcium, IGF-1 or IGFBP-3. The IGF-1/IGFBP-3 ratio was also assessed, but no significant variation was observed. Concentrations of IGF-1 and IGFBP-3 were observed to be close to or below the lower limit of the normal range.

Bone biochemical markers

The effects of vitamin C and E supplementation and physical training on bone biochemical markers are summarized in Table III. Bone alkaline phosphatase decreased after the intervention by 14.6% ($P = 0.012$), whereas no change was observed for osteocalcin or urinary type I collagen C-telopeptide.

Discussion

Our results show that 8 weeks of vitamin C and vitamin E supplementation in combination with a progressive aerobic training programme resulted in noticeable changes in the parameters of calcium homeostasis and bone formation activity, as evaluated by peripheral markers, in healthy elderly individuals.

Calciotropic hormones

The increases in 25-hydroxyvitamin D and 1, 25dihydroxyvitamin D, and the decrease in intact parathyroid hormone, might counterbalance the unfavourable hormonal profile, very commonly observed in aged populations, that predisposes the elderly to accentuated age-related bone loss. Most studies have demonstrated a gradual reduction in circulating 1,25dihydroxyvitamin D in the elderly, including osteoporotic individuals, of about 30% (Tsai, Heath, Kumar, & Riggs, 1984). The process leading to an alteration in vitamin D status appears to be multi-factorial and related to: (1) a reduction in 1 α-hydroxylase synthesis in association with the reduced sensitivity of this enzyme to parathyroid hormone (Jorde, Bona, & Sundsfjord, 1999); (2) a reduced capacity of the skin to produce vitamin D; and (3) a lack of sufficient sun exposure (Ledger *et al.*, 1994). The increase in circulating 1, 25dihydroxyvitamin D induced by this 8-week intervention might favour certain physiological mechanisms. In particular, it might help to increase intestinal calcium absorption efficiency, which declines with age (Heanay, Recker, Stegman, & Moy, 1989), thus limiting the high parathyroid hormone concentrations and abnormal parathyroid hormone secretory dynamics found in the elderly (McKane *et al.*, 1996). The significantly reduced intact parathyroid hormone observed in our study is consistent with this assumption. This finding is interesting because senile secondary hyperparathyroidism is also a potential pathophysiological factor of senile osteoporosis (Riggs & Melton, 1983). The net

Table II. Parameters of calcium homeostasis and somatotropic hormone values at baseline and after 8 weeks of intervention (mean $\pm$ *s*).

	Baseline	After 8 weeks of intervention	*P*-value	Normal range
Calcium homeostasis				
iCa ($mmol \cdot l^{-1}$)	**1.18 ± 0.06**	**1.19 ± 0.04**	0.31	1.10–1.25
25(OH)D ($ng \cdot ml^{-1}$)	20.1 ± 7.2	28.7 ± 7.8	< 0.001	16–28
$1.25(OH)_2D$ ($ng \cdot ml^{-1}$)	51.4 ± 12.0	65.2 ± 16.0	< 0.01	20–66
IPTH ($pg \cdot ml^{-1}$)	39.9 ± 24.4	32.9 ± 20.8	0.048	10–55
Somatotropic hormones				
IGF-1 ($ng \cdot ml^{-1}$)	114.6 ± 36.0	102.5 ± 37.0	0.094	100–500
IGFBP-3 ($ng \cdot ml^{-1}$)	1631.1 ± 310.0	1601.6 ± 206.4	0.892	2000–4500
BII	7.0 ± 1.4	6.3 ± 1.8	0.168	–

Note: iCa = ionized calcium, 25(OH)D = 25-hydroxyvitamin D, $1,25(OH)_2D$ = 1,25-dihydroxyvitamin D, iPTH = intact parathyroid hormone, BII = bioavailability IGF-1 index ([(IGF-1 = insulin-like growth factor-1/IGFBP-3 = insulin-like growth factor binding protein-3)*1000].

Table III. Bone biochemical marker values at baseline and after 8 weeks of intervention (mean $\pm$ *s*).

	Baseline	After 8 weeks of intervention	*P*-value	Normal range
Formation				
Osteocalcin ($ng \cdot ml^{-1}$)	12.9 ± 4.5	13.2 ± 4.5	0.735	5–20
B-ALP ($ng \cdot ml^{-1}$)	11.7 ± 3.4	10.0 ± 2.8	0.012	4–16
Resorption				
CTX ($pmol \cdot ml^{-1}$)	2522 ± 1320	2744 ± 1648	0.250	< 5500

Note: B-ALP = bone alkaline phosphatase, CTX = urinary type I collagen C-telopeptide.

result of such alteration is generally an increase in bone turnover that leads to bone loss, especially cortical bone loss (Riggs & Melton, 1983). Hypovitaminosis D has also been associated with muscle weakness, limb pain, and impaired muscle function, all of which reduce the ability to counteract falls (Boland, 1986). Moreover, Pasco *et al.* (2004) demonstrated that the reduction in serum 25-hydroxyvitamin D concentrations during winter was accompanied by an increase in the proportion of falls resulting in fracture. Improved vitamin D status should thus contribute to reducing fall-related fractures, most of which involve the hip (Cooper & Melton, 1996). Such an effect of vitamin D on muscle would be mediated by the highly specific receptors of 1,25-dihydroxyvitamin D that are localized in human skeletal muscle and that seem to promote protein synthesis and improve muscle strength (Bischoff *et al.*, 2001).

The precise biochemical mode of action of antioxidants on the regulation of the vitamin D endocrine system is not completely understood. Nevertheless, in experimental animal studies, it is believed that vitamin C deficiency is associated with decreased bone mineral density, low concentrations of 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D, and a reduced concentration of 1,25-dihydroxyvitamin D receptors in the target tissues (Kipp *et al.*, 1996; Sergeev, Arkhapchev, & Spirichev, 1990). Vitamin C seems to interact with vitamin D by protecting or stimulating renal 1 α-hydroxylase activity (Cantatore, Loperfido, Magli, Mancini, & Carrozzo, 1991; Sergeev *et al.*, 1990). Moreover, adequate vitamin C supply is a critical factor for the complete expression of this enzyme (Sergeev *et al.*, 1990).

Cantatore *et al.* (1991) reported a significant increase in serum 1,25-dihydroxyvitamin D in middle-aged individuals receiving ascorbic acid supplementation. However, in contrast with our results, no significant change in 25-hydroxyvitamin D or intact parathyroid hormone was observed. The short duration of the treatment (10 days) and the dose (150 $mg \cdot day^{-1}$ i.v.) probably explain the contrasting results. In fact, the action of vitamin C on 1,25-dihydroxyvitamin D synthesis is dose-dependent. A physiologic dose promotes synthesis of the molecule, while higher doses inhibit and reduce serum ionized calcium concentrations (Cantatore *et al.*, 1991). In our study, the increase in the synthesis of 1,25-dihydroxyvitamin D, the most biologically active metabolite of vitamin D, had no negative effect on its precursor 25-hydroxyvitamin D, which evaluates the stock of vitamin D. These observations suggest that, in addition to the known action of vitamin C on 1 α-hydroxylase activity, other mechanisms of action should be considered to explain the increase in 25-hydroxyvitamin D, such as a modification in the activity of hepatic 25-hydroxylase and/or an increase in vitamin D concentration.

Aerobic training programmes have also been suggested to modulate vitamin D metabolism, as demonstrated by the higher concentration of 1, 25-dihydroxyvitamin D found in endurance-trained men compared with less active controls (Zittermann *et al.*, 2000) and the increase in this metabolite after intense endurance training in triathletes (Maïmoun *et al.*, 2004). However, a comparison with our results should be made with caution, because the type of training, the duration of the programme, and the population studied were all different. In outdoor sports, the enhanced vitamin D synthesis might be due to the increase in sunlight exposure, in addition to the direct action of exercise itself (Zittermann *et al.*, 2000). However, in our study, this hypothesis did not apply because the physical exercise programme was performed in a gymnasium with no other changes in the participants' lifestyle.

Several studies have reported an annual periodicity in circulating 25-hydroxyvitamin D, with the nadir occurring in winter and the peak in summer, while the seasonal variation in serum parathyroid hormone was represented by an inverse model (Pasco *et al.*, 2004; Woitge *et al.*, 2000). Our study was conducted in May and June, favourable months for ultraviolet exposure, and this may have contributed to the increased synthesis of serum 25-hydroxyvitamin D. Although the effect of season cannot be dismissed entirely without the monitoring of a control group matched for age and sex, two arguments suggest that seasonal factor had a minor effect on the observed hormonal variations. First, despite the study period being relatively short (8 weeks), the variation in 25-hydroxyvitamin D we observed (42%) was similar to or greater than the mean amplitude (range 25 – 50%) of the circannual rhythm reported for 25-hydroxyvitamin D (Rapuri, Kinyamu, Gallagher, & Haynatzka, 2002; Pasco *et al.*, 2004; Woitge *et al.*, 2000). Moreover, the absolute value of 25-hydroxyvitamin D found after 8 weeks of the intervention was, on average, close to the upper limit of the normal range of our laboratory.

The expected favourable effect of the intervention on bone metabolism was not confirmed by the results of the bone biochemical markers. We only found a reduction in bone alkaline phosphatase concentrations, which could suggest an alteration in bone formation. However, it is unlikely that antioxidant supplementation would have a noticeably negative effect on osteoblast activity, since the other marker of bone formation, osteocalcin, was not

affected. Previous experimental studies on growing pigs using various markers of bone formation have shown that only osteocalcin concentrations are increased by vitamin C supplementation (Pointillart, Denis, Colin, & Lacroix, 1997). Cantatore and Carozzo (1990) reported also an increase in osteocalcin in women supplemented with vitamin C for 10 days. However, it is unlikely that the increased plasma osteocalcin reflected a variation in osteoclast activity because the other markers of bone formation were unaffected (Pointillart *et al.*, 1997). Moreover, in our study, the bone degradation marker, urinary type I collagen C-telopeptide, was not modified by the supplementation, indicating that bone resorption activity was unaffected; this was in line with previous results (Pointillart *et al.*, 1997).

The effects of a short-term training programme on bone turnover in elderly participants remain controversial (Ryan *et al.*, 1994; Sartorio *et al.*, 2001; Yaresheski *et al.*, 1997). The discrepancies might be principally related to the heterogeneous features of the training programmes. Nevertheless, it is worth noting that a reduction in bone alkaline phosphatase was reported in a recent study in young participants undergoing endurance training, without modification of osteocalcin or urinary type I collagen C-telopeptide (Maïmoun *et al.*, 2004). This was interpreted as an adaptation of bone formation activity to training rather than an expression of a bone alteration process. In the present study, changes observed in physical fitness were rather small, which was probably due to the insufficiency, in terms of intensity and duration, of the training programme performed by the elderly participants. This could be confirmed by unchanged IGF-1 or IGFBP-3 concentrations at the end of the intervention, in so far as somatotropic parameters are known to be modified by physical activity. Horber and colleagues (Horber, Kohler, Lippuner, & Jaeger, 1996) reported higher IGF-1 concentrations in male joggers with a mean age of 67 years than age-matched sedentary men. A similar endocrine profile was reported in trained individuals aged 50–74 years (Ravaglia *et al.*, 2001; Tissandier, Peres, Fiet, & Piette, 2001). Such an adaptation probably requires long and regular physical training (Horber *et al.*, 1996; Ravaglia *et al.*, 2001; Tissandier *et al.*, 2001) as shown in several studies in accordance with our results, where no IGF-1 variation was found in elderly men and post-menopausal women after strength training of short duration (Ryan *et al.*, 1994; Sartorio *et al.*, 2001; Yaresheski *et al.*, 1997). Nevertheless, it is likely that our measurements of serum IGF-1 concentration underestimated the changes in local IGF-1 production within the bones. This limitation is inherent to all studies using similar biochemical assays.

Conclusion

Fractures related to osteoporosis are an important cause of morbidity and mortality and represent a major socio-economic problem. Strategies to both prevent falls and limit bone loss must be developed. Our findings suggest that an intervention combining vitamin C and E supplementation together with aerobic training might improve the calciotropic hormone profile generally altered in the elderly, associated with bone loss. As our participants showed good compliance, investigations using programmes of longer duration should be undertaken to evaluate the effect on bone mineral density and fracture risk.

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References

Bischoff, H. A., Borchers, M., Gudat, F., Duermeller, U., Theiler, R., Stakelin, H. B. *et al.* (2001). *In situ* detection of 1,25-dihydroxyvitamin D3 receptor in human skeletal muscle tissue. *Histochemical Journal*, *33*, 19–24.

Boland, R. (1986). Role of vitamin D in skeletal muscle function. *Endocrine Reviews*, *7*, 434–438.

Cantatore, F. P., & Carozzo, M. (1990). The action of ascorbic acid in increasing serum 1,25 $(OH)_2$D3 and osteocalcin in human. *Calcified Tissue International*, *46* (suppl. 2), A16.

Cantatore, F. P., Loperfido, M. C., Magli, D. M., Mancini, L., & Carrozzo, M. (1991). The importance of vitamin C for hydroxylation of vitamin D3 to 1,25(OH)2D3 in man. Clinical. *Rheumatology*, *10*, 162–167.

Chapuy, M. C., Arlot, M. E., Duboeuf, F., Brun, J., Crouzet, B., Arnaud, S. *et al.* (1992). Vitamin D3 and calcium to prevent hip fractures in elderly women. *New England Journal of Medicine*, *327*, 1637–1642.

Consensus Development Conference (1993). Diagnosis, prophylaxis and treatment of osteoporosis. *American Journal of Medicine*, *94*, 646–650.

Cooper, C., & Melton, L. J. (1996). Magnitude and impact of osteoporosis and fractures. In R. Marcus, D. Felman, & J. Kelsey (Eds.), *Osteoporosis* (pp. 419–434). London: Academic Press.

Endres, D. B., Morgan, C. H., Garry, P. J., & Omdahl, J. L. (1987). Age-related changes in serum immunoreactive parathyroid hormone and its biological action in healthy men and women. *Journal of Clinical Endocrinology and Metabolism*, *65*, 724–731.

Fox, K. M., Cummings, S. R., & Threets, K. (1994). Family history and risk of osteoporotic fracture. *Journal of Bone and Mineral Research*, *9* (suppl. 1), S153.

Franceschi, R. T. (1992). The role of ascorbic acid in mesenchymal differentiation. *Nutrition Reviews*, *50*, 65–70.

Gunnes, M., & Lehmann, E. H. (1995). Dietary calcium, saturated fat, fiber and vitamin C as predictors of forearm cortical and trabecular bone mineral density in healthy children and adolescents. *Acta Paediatrica*, *84*, 388–392.

Gutin, B., & Gasper, M. J. (1992). Can vigorous exercise play a role in osteoporosis prevention? A review. *Osteoporosis International*, *2*, 55–69.

Heanay, R. P., Gallagher, J. C., Johnsson, C. C., Neer, R., Parfitt, A. M., & Whedon, G. D. (1982). Calcium nutrition and bone health in the elderly. *American Journal of Clinical Nutrition*, *36*, 986–1013.

Heanay, R. P., Recker, R. R., Stegman, M. R., & Moy, A. J. (1989). Calcium absorption in women: Relationships to calcium intake, estrogen status, and age. *Journal of Bone and Mineral Research*, *4*, 469–475.

Horber, F. F., Kohler, S. A., Lippuner, K., & Jaeger, P. (1996). Effect of regular physical training on age-associated alteration of body composition in men. *European Journal of Clinical Investigation*, *26*, 279–285.

Hurley, B. F., & Hagberg, J. M. (1998). Optimising health in order persons: Aerobic or strength training? *Exercise and Sport Sciences Reviews*, *26*, 61–89.

Jorde, J., Bona, K. H., & Sundsfjord, J. (1999). Population-based study on serum ionised calcium, serum parathyroid hormone, and blood pressure: The Tromso study. *European Journal of Endocrinology*, *141*, 350–357.

Kallinen, M., & Markku, A. (1995). Aging, physical activity and sports injuries: An overview of common sports injuries in the elderly. *Sports Medicine*, *20*, 41–52.

Keer, D., Morton, A., Dick, I., & Prince, R. (1996). Exercise effects on bone mass in postmenopausal women are site-specific and load-dependent. *Journal of Bone and Mineral Research*, *11*, 218–225.

Kipp, D. E., McElvain, M., Kimmel, D. B., Akhter, M. P., Robinson, R. G., & Lukert, B. P. (1996). Scurvy results in decreased collagen synthesis and bone density in the guinea pig animal model. *Bone*, *18*, 281–288.

Ledger, G. A., Burrit, M. F., Kao, P. C., O'Fallon, W. M., Riggs, B. L., & Khosla, S. (1994). Abnormalities of parathyroid hormone secretion in elderly women that are reversible by short term therapy with 1,25-dihydroxy-vitamin D_3. *Journal of Clinical Endocrinology and Metabolism*, *79*, 211–216.

Maggio, D., Barabani, M., Pierandrei, M., Polidori, M. C., Catani, M., Mecocci, P. *et al.* (2003). Marked decrease in plasma antioxidants in aged osteoporotic women: Results of a cross-sectional study. *Journal of Clinical Endocrinology and Metabolism*, *88*, 1523–1527.

Maïmoun, L., Galy, O., Manetta, J., Coste, O., Peruchon, E., Micallef, J. P. *et al.* (2004). Competitive season of triathlon does not alter bone metabolism and bone mineral status in male triathletes. *International Journal of Sports and Medicine*, *25*, 230–234.

Maïmoun, L., Simar, D., Malatesta, D., Caillaud, C., Peruchon, E., Couret, I. *et al.* (2005). Response of bone metabolism-related hormones to a single session of strenuous exercise in active elderly subjects. *British Journal of Sports Medicine*, *39*, 497–502.

McKane, W. R., Khosla, S., Egan, K. S., Robins, S. P., Burritt, M. F., & Riggs, B. L. (1996). Role of calcium intake in modulating age-related increases in parathyroid function and bone resorption. *Journal of Clinical Endocrinology and Metabolism*, *81*, 1699–1703.

Melhus, H., Michaelsson, K., Holberg, L., Wolk, A., & Ljunghall, S. (1999). Smoking, antioxidant vitamins, and the risk of hip fracture. *Journal of Bone and Mineral Research*, *14*, 129–135.

Menkes, A., Mazels, S., Redmond, R. A., Koffler, K., Libanati, C. R., Gunberg, C. M. *et al.* (1993). Strength training increases regional bone mineral density and bone remodelling in middle-aged and older men. *Journal of Applied Physiology*, *74*, 2478–2484.

Morton, D. J., Barrett-Connor, E. L., & Schneider, D. L. (2001). Vitamin C supplement use and bone mineral density in postmenopausal women. *Journal of Bone and Mineral Research*, *16*, 135–140.

Nguyen, T. V., Center, J. R., & Eisman, J. A. (2000). Osteoporosis in elderly men and women: Effects of dietary calcium, physical activity, and body mass index. *Journal of Bone and Mineral Research*, *15*, 322–331.

Pasco, J. A., Henry, M. J., Kotowicz, M. A., Sanders, K. M., Seeman, E., Pasco, J. R. *et al.* (2004). Seasonal periodicity of serum vitamin D and parathyroid hormone, bone resorption and fractures: The Geelong Osteoporosis Study. *Journal of Bone and Mineral Research*, *19*, 752–758.

Pointillart, A., Denis, I., Colin, C., & Lacroix, H. (1997). Vitamin C supplementation does not modify bone mineral content or mineral absorption in growing pigs. *Journal of Nutrition*, *127*, 1514–1518.

Prince, R. L., Smith, M., Dick, I. M., Price, R. I., Webb, P. G., Henderson, N. K. *et al.* (1991). Prevention of postmenopausal osteoporosis: A comparative study of exercise, calcium supplementation, and hormone replacement therapy. *New England Journal of Medicine*, *325*, 1189–1195.

Rapuri, P. B., Kinyamu, H. K., Gallagher, J. C., & Haynatzka, V. (2002). Seasonal changes in calciotropic hormones, bone markers, and bone mineral density in elderly women. *Journal of Clinical Endocrinology and Metabolism*, *87*, 2024–2032.

Ravaglia, G., Forti, P., Maioli, F., Pratelli, L., Vettori, C., Bastagli, L. *et al.* (2001). Regular moderate intensity physical activity and blood concentrations of endogenous anabolic hormones and thyroid hormones in aging men. *Mechanisms of Ageing and Development*, *122*, 191–203.

Riggs, B. L., & Melton, L. J. (1983). Evidence for two distinct syndromes of involutional osteoporosis. *American Journal of Medicine*, *75*, 899–901.

Rubin, C. T., & Lanyon, L. E. (1984). Regulation of bone formation by applied dynamic loads. *Journal of Bone and Joint Surgery*, *66A*, 397–402.

Ryan, A. S., Treuth, M. S., Rubin, M. A., Miller, J. P., Nicklas, B. J., Landis, D. M. *et al.* (1994). Effects of strength training on bone mineral density: Hormonal and bone turnover relationships. *Journal of Applied Physiology*, *77*, 1678–1684.

Sartorio, A., Lafortuna, C., Capodaglio, P., Vangeli, V., Narici, M. V., & Faglia, G. (2001). Effects of a 16 week progressive high-intensity strength training (HIST) on indexes of bone turnover in men over 65 years: A randomised controlled study. *Journal of Endocrinology and Investigation*, *24*, 882–886.

Sergeev, I. N., Arkhapchev, J. P., & Spirichev, V. B. (1990). Ascorbic acid effects in vitamin D hormone metabolism and bindings in guinea pigs. *Journal of Nutrition*, *120*, 1185–1190.

Simon, J. A., & Hudes, E. S. (2001). Relation of ascorbic acid to bone mineral density and self-reported fractures among US adults. *American Journal of Epidemiology*, *154*, 427–433.

Tissandier, O., Peres, G., Fiet, J., & Piette, F. (2001). Testosterone, dehydroepiandrosterone, insulin-like growth factor 1, and insulin in sedentary and physically trained aged men. *European Journal of Applied Physiology*, *85*, 177–184.

Tsai, K. S., Heath, H., Kumar, R., & Riggs, B. L. (1984). Impaired vitamin D metabolism with aging in women: Possible role in pathogenesis of senile osteoporosis. *Journal of Clinical Investigation*, *73*, 1668–1672.

Voorrips, L. E., Ravelli, A. C., Dongelmans, P. C., Deurenberg, P., & Van Staveren, W. A. (1991). A physical activity questionnaire for the elderly. *Medicine and Science in Sports and Exercise*, *23*, 974–979.

Woitge, H. W., Knothe, A., Witte, K., Schmidt-Gayk, H., Ziegler, R., Lemmer, B. *et al.* (2000). Circannual rhythms and interactions of vitamin D metabolites, parathyroid hormone, and biochemical markers of skeletal homeostasis: A prospective study. *Journal of Clinical Endocrinology and Metabolism*, *15*, 2443–2450.

Yaresheski, K. E., Campbell, J. A., & Kohrt, W. M. (1997). Effect of resistance exercise and growth hormone on bone density in older men. *Clinical Endocrinology*, *47*, 223–229.

Zittermann, A., Sabatschus, O., Jantzen, S., Platen, P., Danz, A., Dimitriou, T. *et al.* (2000). Exercise-trained young men have higher calcium absorption rates and plasmacalcitriol levels compared with age-matched sedentary controls. *Calcified Tissue International*, *60*, 332–337.