A 1-y walking program and increased dietary calcium in postmenopausal women: effects on bone[1-4]

Miriam E Nelson, Elizabeth C Fisher, F Avraham Dilmanian, Gerard E Dallal, and William J Evans

ABSTRACT   The effects of a supervised 1-y walking program and increased dietary calcium (milk supplement, 831 mg/d, vs placebo drink, 41 mg/d) on bones were examined in 36 postmenopausal women (60.2 ± 6.5 y). Trabecular bone-mineral density (BMD) of the lumbar spine (L_1–L_3), measured by computed tomography, increased by 0.5% in exercising women ($n = 18$) and decreased by 7.0% in sedentary women ($n = 18$; $P = 0.02$). Femoral-neck BMD measured by dual-photon absorptiometry (DPA) increased by 2.0% in women consuming high dietary calcium ($n = 18$) and decreased by 1.1% in those on moderate calcium intake ($n = 18$; $P = 0.001$). Neither exercise nor dietary calcium had an effect on lumbar spine (L_2–L_4) measured by DPA, distal radius measured by single-photon absorptiometry, or total body calcium measured by in vivo neutron activation. The varying proportions and rates of turnover of trabecular and cortical bone from one site to another suggest that exercise and high dietary calcium may preferentially alter bone density at different skeletal sites.   *Am J Clin Nutr* 1991;53:1304–11.

KEY WORDS   Exercise, diet, osteoporosis, body composition, dual-photon absorptiometry, single-photon absorptiometry, quantitative computed tomography, in vivo neutron-activation analysis

Introduction

Osteoporosis is a major public health problem that is characterized by low bone mass and increased susceptibility to fractures, primarily in the hip, spine, and wrist. It is estimated to cause 1.3 million fractures annually in the United States in people aged ≥ 45 y, with an estimated annual cost between 7 and 10 billion dollars (1). One-third of women aged > 65 y will have one or more vertebral fractures and as women survive into their eighties, one-third will experience hip fractures (2). Therefore, poor bone health is an increasingly common medical, social, and economic problem and its prevention or amelioration is of major importance for maintenance of health in elderly people.

Increased activity and increased dietary calcium have been suggested as two ways of preserving bone health in older women (3–6). Physical activity and weight bearing are important mechanical stimuli for bone gain (7) whereas physical inactivity has been implicated in bone loss (8). A variety of prospective studies have examined the effects of exercise on postmenopausal bone loss (9–12). However, the results of these studies vary, probably because of differences in exercise protocols (either being more vigorous or less vigorous) and measurement techniques used.

Epidemiologic studies demonstrated that populations consuming low amounts of calcium have an increased prevalence of bone fractures and that symptomatic osteoporotic people have lower calcium intakes (13, 14). Although there is a general agreement that lifelong adequate calcium intakes will retard bone loss, evidence for a direct relationship between dietary calcium and bone mineral density (BMD) is inconsistent (15).

The disparities in results obtained from a variety of prospective studies examining the effect of exercise and/or calcium supplementation on BMD may be attributed to the various detection methods employed as well as to the variety in sites where bone mineral status is measured. The purpose of the present study was to examine the effects of a 1-y walking program and increased dietary calcium on postmenopausal bone loss at a variety of skeletal sites.

Methods

Subjects

Forty-one postmenopausal Caucasian women were recruited from the Boston area to participate in this study after giving their informed consent. The protocol was approved by the Tufts University Human Investigation Review Committee. The

women were ≥ 2-y postmenopausal (no menstrual bleeding in past 2 y) but not older than 70 y. They did not engage in any regular exercise program, weighed < 130% of ideal body weight, and were not taking estrogen or any other medication known to interfere with calcium metabolism. In addition, none of the women had a history of amenorrhea during the premenopausal years and all were in good health as determined by a physical examination and urine and blood tests.

Design

To enhance compliance of the exercise training, sedentary and exercise groups were recruited according to preference rather than random assignment. All of the women were randomly assigned in a double-blind fashion to a high-calcium milk drink (831 mg Ca) or a low-calcium placebo drink (41 mg Ca) (**Table 1**). Drinks were developed and prepared in our Metabolic Nutrition Laboratory in batches and frozen until use. The calcium content of the drinks was measured by direct-current plasma-emission spectroscopy after digestion in 50% nitric acid at 85 °C (Beckman Co, Anaheim, CA). Subjects consumed one drink per day for the entire study period. One subject in the low-calcium placebo group could not tolerate the drink and was given a placebo pill in its place. During vacations subjects were given either a calcium carbonate supplement (900 mg Ca) or a placebo pill depending on which group they were in. All subjects were instructed by the dietitian to consume 800 mgCa/d by including in their diet four servings of dairy products daily. In addition, all subjects were weighed every week so that none of the women gained or lost > 2.2 kg body wt over the entire study period. In summary, there were four groups: *1*) exercise, moderate dietary calcium (EXMOD); *2*) exercise, high dietary calcium (EXHI); *3*) sedentary, moderate dietary calcium (SEDMOD); and *4*) sedentary, high dietary calcium (SEDHI). Of the 41 women who started the study, 36 women completed the study. Three women in the exercise group and one woman in the control group dropped out in the first month because of time constraints and one woman in the control group dropped out after 6 mo because of uncontrollable weight gain. All five of these women were on the low-calcium drink.

The exercise group participated in a supervised walk four times per week for 52 wk. Exercise sessions lasted 50 min. After 4 wk of training the subjects wore leaded belts (3.1 kg) around their waists during each session. Subjects were instructed to exercise at 75–80% of measured maximum heart rate. Heart rate during exercise was measured periodically by the exercise leader and recorded in a log book. During at least six different exercise sessions, heart rates were recorded continuously every 45 s with a heart-rate monitor (UNIQ Heartwatch belt, Kemple, Finland) to assure that the subjects were exercising at the prescribed intensity. During vacations each subject was asked to keep a diary of her training. No subject took > 3 wk vacation. Attendance averaged > 90%.

The women in the sedentary group were told they could engage in occasional weekend tennis, skiing, cycling, swimming, and other recreational activities. They were asked not to engage in these activities on a weekly basis.

Testing protocol

Testing took place at baseline (prestudy) and at 52 wk (poststudy). The same testing protocol was used both times. One to two days before testing, each woman collected a 24-h urine sam-

TABLE 1
Calcium beverage composition

	High-calcium milk drink	Moderate-calcium milk drink
Nuform milk (1% fat) (g)*	369.0	—
Nonfat dry milk powder (g)	30.6	—
Fluid nondairy creamer (g)	19.2	187.5
Heavy cream (g)	—	49.5
Imitation vanilla extract (g)	0.9	3.0
Glucose polymers (g)	3.0	7.5
Ergocalciferol (μg)	—	4.5
Energy (kcal)	295	437
Protein (g)	23.1	3.0
Carbohydrates (g)	37.1	25.9
Fat (g)	6.04	37.0
Calcium (mg)	831	41.0
Vitamin D (μg)	7.1	4.5
Phosphorus (mg)	668	151

* HP Hood, Inc, Somerville, MA.

ple in an acid-free container after eating a meat-free diet for 3 d. The urine samples were stored at −20 °C until analysis. On the first testing day each woman reported to the laboratory at 0700 after an overnight fast. A blood sample was drawn after a 30-min rest in the supine position. The blood samples were allowed to clot and then centrifuged; samples of serum were stored at −20 °C until analyzed. The blood for measuring serum calcium was collected into rubber-stoppered evacuated tubes with minimum exposure to air and analyzed within 2 h. Hydrostatic weight and maximal aerobic capacity were then measured. BMD measurements and muscle-strength measurements were also made. On the second day of testing, subjects flew to Long Island, NY, and reported to Brookhaven National Laboratory (BNL) for the determination of total body calcium (TBCa) and total body potassium (TBK).

Measurements

Body density was estimated by hydrostatic weighing. Underwater weight was measured with a Sauter scale (model Kizo, Denshore Scale, Holbrook, MA). Lung residual volume was estimated (16). Body fat was calculated from total body density by using Siri's equation (17). Fat-free mass was calculated by subtraction.

A multistage continuous exercise test was performed on a treadmill for the determination of maximal aerobic capacity (18). Each test continued until volitional fatigue. Eighty-nine percent of the subjects reached a respiratory quotient of ≥ 1.00. Ninety-two percent of subjects reached a heart rate no < 5 beats/min below the predicted maximum rate. Expired gas was collected and continuously analyzed for carbon dioxide (Beckman LB-2 CO_2 analyzer, Beckman, Anaheim, CA) and oxygen (oxygen analyzer, Applied Electrochemistry Inc, Sunnyvale, CA). The analyzers were connected to a Z-150 PC Series Computer (Zenith Data Systems Corp, St Joseph, MI) for immediate calculation of oxygen consumption and respiratory-exchange ratio. The analyzers were calibrated with gases of known partial pressures of oxygen and carbon dioxide (19). Oxygen consumption was corrected for body size by dividing by body weight (mL $O_2 \cdot kg^{-1} \cdot min^{-1}$).

Isometric and dynamic strength of the knee and elbow extensors and flexors were measured with an isokinetic dynamometer (CYBEX II, Ronkonkoma, NJ). During the test the lever arm was attached to the nondominant limb and its axis of rotation was aligned with the anatomical axis of rotation of the joint. A familiarization period preceded the actual testing. Maximal voluntary isometric contraction was measured in the knee and elbow by having the subject maximally contract the joint five times at 60°/s. Peak torque was defined as the highest torque of five contractions and was measured in Nm.

BMDs of the lumbar spine (L_2–L_4) and the nondominant proximal femur (femoral neck) were measured by dual-photon absorptiometry (DPA) (20) with a DP-3 model scanner (Lunar Radiation Corp, Madison, WI). DPA measures all mineral within the scan path. This includes not only the trabecular bone but also the cortical bone. The vertebrae contain ~60% trabecular bone whereas the proximal femur contains ~43% trabecular bone (21). The source was changed once during the study period. DPA measurements of lumbar spine were corrected for variation in source, source strength, and soft-tissue thickness by the use of a bone phantom measured at different acrylic thicknesses (22). The precision of this method in our laboratory was 3%.

Lumbar trabecular BMD was also measured by single-energy quantitative computed tomography (QCT) by using the method of Cann (23). The system was a third-generation Siemens DR3 CT scanner (Somatom-Siemens, Erlanger, FRG). The QCT software package was a commercial product from Image Analysis Inc (Los Angeles) that used a dipotassium hydrogen phosphate reference phantom. QCT is an x-ray imaging technique that volumetrically measures the trabecular BMD of the spine at the midpoint of the L_1–L_3 vertebral bodies. Short-term in vivo reproducibility was obtained on a cadaver with 90 mg/cm³ lumbar trabecular BMD and the precision was assessed at 1.3%. Long-term reproducibility was obtained by using a dipotassium hydrogen phosphate solution in a cylindrical-torso water phantom. This phantom contained 100 mg/cm³ lumbar trabecular BMD and the precision was 1–3%.

The BMD of the shaft of the radius of the nondominant arm was measured by single-photon absorptiometry (24) with an ^{125}I source in conjunction with a scintillation detector (Lunar Radiation Corp). The measurement site was one-third the distance from the ulnar styloid process to the olecranon. This site of the radius contains ~10–15% trabecular bone (25). The precision of this measurement in our laboratory was 2%.

TBCa was measured by the delayed-γ neutron-activation method (26) at BNL. The method used a total-body neutron-activation facility and a whole-body counter. The total body contains ~80% cortical bone (25). The delayed-γ activation technique provides a precision of 0.8% for TBCa in an anthropomorphic phantom and in subjects with no weight change (27).

The whole-body counter at BNL was also used to measure absolute amounts of TBK (27, 28). The method is based on counting the 0.234 pJ γ rays emitted from the endogenous ⁴⁰K (natural abundance − 0.012%) of the body. The γ-attenuation correction described in the previous paragraph was applied to the data. The absolute calibration of the system was obtained with an anthropomorphic phantom. The precision of the method is 1.5% for phantoms and for subjects with no weight change.

Diet records

During the initial physical examination each woman was instructed by a dietitian on how to complete an accurate 7-d food record. The subjects completed the food record before entry into the study and poststudy. The diaries were returned to the dietitian, who reviewed the information with the subject. The diet records were analyzed for energy, protein, fat, carbohydrate, calcium, and vitamin D by using the Grand Forks Data Base (GRAND, Release 8606, USDA-ARS Grand Forks Human Nutrition Research Center, Grand Forks, ND). The poststudy dietary data includes the calcium-beverage dietary data.

Blood and urine analyses

Serum estrone was measured by a previously described radioimmunoassay (29). The respective inter- and intraassay coefficients of variation (CVs) were 9.9% and 4.6% for estrone. Serum 1,25-dihydroxyvitamin D [1,25(OH)$_2$D] and 25-hydroxyvitamin D [25(OH)D] were measured by competitive-protein-bonding analysis (30). The respective inter- and intraassay CVs were 7.7% and 4.9% for 1,25(OH)$_2$D and 7.3% and 5.0% for 25(OH)D. Serum growth hormone (GH), intact parathyroid hormone (iPTH), somatomedin-C (SOM-C), and calcitonin were measured by radioimmunoassay (Nichols Institute, San Juan Capistrano, CA). The respective inter- and intraassay CVs were 7.3% and 4.3% for GH, 5.6% and 3.4% for iPTH, 11.2% and 5.0% for SOM-C, and 11.6% and 8.3% for calcitonin. Serum total calcium was measured by direct potentiometry (Nova 7 Electrolyte Analyzer, Waltham, MA). All samples were done at the same time for each analysis; all analyses were done in duplicate.

Urinary creatinine was measured by colorimetric reaction (31) by using a COBAS FARA analyzer (Roche Diagnostic Systems Inc, Nutley, NJ). Urinary calcium was measured by direct-current plasma-emission spectroscopy (Beckman Co). 3-Methylhistidine was measured by high-pressure liquid chromatography (HPLC) by the method of Wassner et al (32). An HPLC method was used to determine γ-carboxyglutamate (33). Hydroxyproline in hydrolyzed urine was measured colorimetrically with an amino acid analyzer (Beckman 6300).

Statistical analysis

The results are reported for the four groups EXMOD, EXHI, SEDMOD, and SEDHI. There were nine subjects in each group. Differences at baseline were determined by using Tukey's honestly significant differences or two-factor (calcium and exercise) analysis of variance. Because there was no calcium-by-exercise interaction for any of the baseline variables, calcium and exercise main effects were examined. This is equivalent to dividing the entire group of women into exercise (EXER) and sedentary (SED) groups or moderate dietary calcium (MODCA) and high dietary calcium (HICA) groups, with 18 subjects in each of these groups.

Associations between variables and initial bone status and potential predictor variables were examined by using forward-selection and backward-elimination regression analysis. Repeated-measures analysis of variance was used when pre- and posttreatment measurements were compared. All analyses were done with the SAS statistical package (34). Results were considered significant if $P < 0.05$. Data are reported as mean ± SEM.

Results

The physical characteristics of the women at entry into the study are shown in **Table 2.** No differences were seen between

TABLE 2
Baseline physical characteristics*

Age (y)	60.2 ± 1.1
Years postmenopause (y)	10.8 ± 1.2
Parity	3.4 ± 0.3
Weight (kg)	64.03 ± 1.4
Height (cm)	161.9 ± 1.0
Body mass index†	24.4 ± 0.5
Fat (%)	42.1 ± 0.9
Fat-free mass (kg)	36.6 ± 0.6
Total body potassium (mol)	2.21 ± 0.04
Urinary creatinine (mmol/d)	8.0 ± 0.4

* $\bar{x}$ ± SEM.
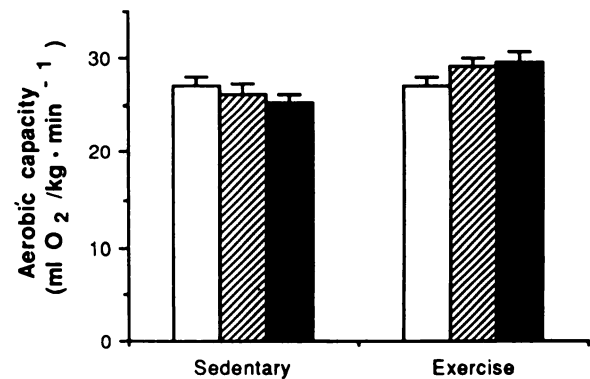† Expressed in kg/m^2.

FIG 1. Changes in aerobic capacity over the 12-mo study period between the exercise group and the sedentary group (□ prestudy, ▨ midstudy, ■ poststudy). $\bar{x}$ ± SEM. The change in aerobic capacity of the exercise group was different from the change observed in the sedentary group ($P < 0.001$).

the groups for any of the indices measured except for parity. EXER averaged more births than did SED (4.3 vs 2.4, $P = 0.003$). None of the groups changed differently from the others in any of the body-composition measures and these data are therefore not reported.

Before training there were no significant differences in aerobic capacity between EXER and SED (**Fig 1**). However, by 6 mo aerobic capacity had increased by 7.3% in EXER and had decreased by 3.3% in SED. By 12 mo SED values declined even further, with a total decline of 7.0%, whereas EXER values increased slightly further, with a total increase of 8.8%. The difference in change in aerobic capacity over 12 mo between EXER and SED, as measured by the exercise group-by-time interaction in a repeated-measures analysis of variance is highly significant ($P < 0.001$). Calcium supplementation did not influence the change in aerobic capacity.

Strength testing showed no differences between groups at entry into study and no changes over time or correlation with bone status in any groups. Therefore, these data are not reported.

The dietary intakes of various nutrients are shown in **Table 3**. There were no significant differences between groups at entry into the study in any of the nutrients measured. The changes in nutrient intake were due to intake of the calcium beverages. All groups reported an increase in calorie intake over time, which corresponds closely to the caloric content of the beverages.

The bone-status data for all groups are shown in **Table 4**. There were no significant differences between groups at entry into the study. To determine which factors were most related to bone status, forward-selection and backwards-elimination regression analyses were performed on prestudy bone-status measurements by using body mass, age, fat-free mass, aerobic capacity ($\dot{V}O_2max$), and years postmenopause as potential predictors. TBCa (in moles) was positively related to fat-free mass (FFM) and negatively related to $\dot{V}O_2max$ and age

$$TBCa = 22.937 + 0.372 \text{ (FFM)} - 0.240(\dot{V}O_2max)$$
$$- 0.160(AGE), R^2 = 0.553$$

Femoral BMD (FBMD) was positively related to FFM

$$FMBD = 0.297 + 0.014(FFM), R^2 = 0.207$$

BMD of the trabecular bone in the spine measured by QCT (TMBD) was negatively related to age

$$TMBD = 224.611 - 2.130(AGE), R^2 = 0.259$$

Over the 12-mo period the trabecular BMD of the spine, as measured by QCT, decreased by 7.0% in SED whereas there was a 0.5% increase in EXER (**Fig 2**). The difference in change in TBMD was judged significant as measured by the time-by-exercise interaction in a repeated-measures analysis of variance ($P = 0.028$). There was no significant effect involving calcium. The femoral-neck BMD as measured by DPA decreased 1.1% in MODCA and increased 2.0% in HICA (**Fig 3**). Again, as measured by using the time-by-calcium interaction in a repeated-measures analysis of variance, the difference in change in FBMD was judged significant ($P = 0.014$). There was no significant effect involving EXER.

Hormones and urinary metabolites showed no differences between groups nor changes over time within in any group. The vitamin D status of the groups is reported in **Table 5**.

TABLE 3
Dietary intake by drink group*

	Moderate calcium (n = 18)		High calcium (n = 18)	
Calcium (mg/d)				
Prestudy	869	± 76	889	± 101
Poststudy	761	± 51	1462	± 53†‡
Energy (kcal/d)				
Prestudy	1650	± 68	1553	± 73
Poststudy	1986	± 97‡	1744	± 53†‡
Protein (g/d)				
Prestudy	71.1 ±	3.8	68.5 ±	4.1
Poststudy	67.3 ±	3.3	81.1 ±	2.3†‡
Fat (g/d)				
Prestudy	65.6 ±	4.3	59.2 ±	3.2
Poststudy	95.3 ±	6.2‡	61.9 ±	3.1†
Carbohydrate (g/d)				
Prestudy	192	± 9	189	± 11
Poststudy	203	± 9	215	± 10‡
Vitamin D (μg/d)				
Prestudy	2.9 ±	0.5	3.5 ±	0.8
Poststudy	6.3 ±	0.3‡	8.9 ±	0.3†‡

* $\bar{x}$ ± SEM.
† Significantly different from moderate calcium, $P < 0.05$.
‡ Significantly different from prestudy, $P < 0.05$.

TABLE 4
Bone status*

	Exercise		Sedentary	
	Moderate calcium ($n = 9$)	High calcium ($n = 9$)	Moderate calcium ($n = 9$)	High calcium ($n = 9$)
Trabecular BMD (L_1-L_3) (mg/cm³)				
Prestudy†	101.2 ± 8.3	96.4 ± 13.3	79.3 ± 8.2	107.0 ± 7.7
Poststudy†	101.1 ± 9.4	97.5 ± 14.2	74.1 ± 7.5	99.1 ± 6.5
Difference‡	−0.1 ± 3.5	1.1 ± 2.1§	−5.2 ± 2.7§	−7.9 ± 3.4‖
Spine BMD (L_2-L_4) (g/cm²)				
Prestudy	1.181 ± 0.069	1.081 ± 0.067	1.065 ± 0.051	1.138 ± 0.037
Poststudy	1.170 ± 0.069	1.107 ± 0.075	1.082 ± 0.042	1.143 ± 0.038
Difference	−0.010 ± 0.016	0.026 ± 0.025§	0.017 ± 0.024	0.005 ± 0.020
Femur BMD (g/cm²)				
Prestudy	0.837 ± 0.054	0.811 ± 0.055	0.804 ± 0.025	0.803 ± 0.025
Poststudy	0.827 ± 0.053	0.836 ± 0.051	0.796 ± 0.026	0.810 ± 0.020
Difference	−0.010 ± 0.012‖	0.024 ± 0.007	−0.008 ± 0.011§	0.007 ± 0.007
Radius BMD (g/cm²)				
Prestudy	0.645 ± 0.042	0.580 ± 0.022	0.608 ± 0.040	0.652 ± 0.023
Poststudy	0.637 ± 0.039	0.597 ± 0.023	0.612 ± 0.038	0.644 ± 0.020
Difference	−0.008 ± 0.016	0.018 ± 0.018	0.004 ± 0.010	−0.008 ± 0.006
Total body calcium (mol)				
Prestudy	21.0 ± 1.3	20.2 ± 0.9	21.7 ± 0.7	19.8 ± 0.4
Poststudy	20.5 ± 1.1	20.0 ± 0.9	21.0 ± 0.8	19.1 ± 0.6
Difference	−0.5 ± 0.3	−0.2 ± 0.3	−0.7 ± 0.3§	−0.7 ± 0.3

* BMD, bone mineral density.
† Prestudy and poststudy values, $\bar{x}$ ± SEM.
‡ (Poststudy − prestudy values) ± SE of the difference.
§ $n = 8$.
‖ $n = 7$.

Discussion

The past 20 y have seen the development of several noninvasive measurements of bone status. These techniques are capable of quantitating bone mass at axial, appendicular, and total skeletal sites. However, controversy exists over the relationship between measurements (35). Measured sites contain various proportions of cortical and trabecular bone. The greater surface area of trabecular bone is correlated with a greater metabolic activity, and 25% of all trabecular bone is remodeled annually compared with 2–3% of cortical bone (36). These factors make it very difficult for bone-mass changes at one site to predict a
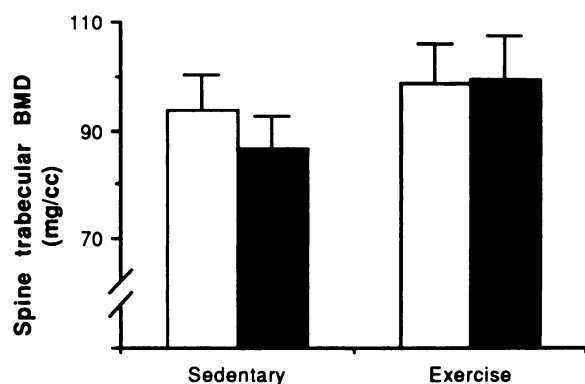

FIG 2. Changes in spine (L_1-L_3) trabecular bone mineral density (BMD) (measured by quantitative computed tomography) over the 12-mo study between the exercise group and the sedentary group (□ prestudy, ■ poststudy). $\bar{x}$ ± SEM. The change in spine trabecular BMD of the exercise group was different from the change observed in the sedentary group ($P = 0.028$).
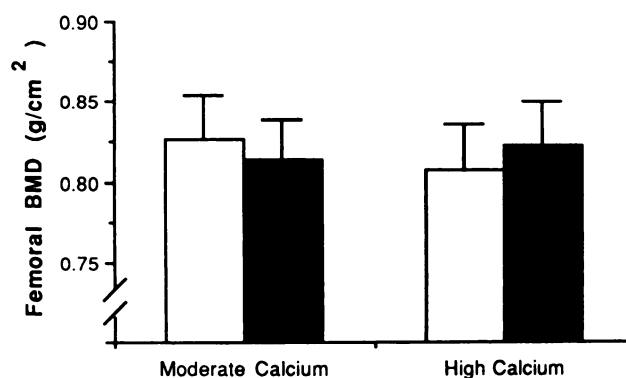

FIG 3. Changes in femoral-neck BMD (measured by dual-photon absorptiometry) over the 12-mo study between the high-dietary-calcium group and the moderate-dietary-calcium group (□ prestudy, ■ poststudy). $\bar{x}$ ± SEM. The change in femoral BMD of the high-calcium group was different from the change observed in the moderate-calcium group ($P = 0.014$).

TABLE 5
Hormonal status*

	Exercise		Sedentary	
	Moderate calcium (n = 9)	High calcium (n = 9)	Moderate calcium (n = 9)	High calcium (n = 9)
1,25(OH)$_2$D (pmol/L)				
Prestudy	67.9 ± 7.2	81.6 ± 8.4	70.8 ± 6.0	77.1 ± 4.6
Poststudy	59.5 ± 5.5	73.0 ± 7.9	60.0 ± 6.2	74.4 ± 8.9
25(OH)D (nmol/L)				
Prestudy	75.1 ± 8.0	69.6 ± 10.7	65.1 ± 9.7	71.9 ± 4.7
Poststudy	72.1 ± 8.5	73.6 ± 5.7	74.4 ± 7.2	77.6 ± 5.5

* $\bar{x}$ ± SEM.

change at another site (35). The present study is the first to address these concerns in an exercise and dietary-calcium intervention study of postmenopausal women. This study evaluates TBCa and measures BMD in the forearm, spine, and hip, the three sites most likely to fracture in old age.

A moderate exercise protocol was chosen for this study because it is an exercise prescription that could be easily followed by a healthy individual over a lifetime and it is frequently recommended by health professionals (4, 6). A dietary milk supplement was chosen to increase calcium in the diet because dietary calcium, with its lactose, may improve the bioavailability of calcium (37).

The results of this study suggest that moderate exercise and dietary calcium had different effects at various skeletal sites. The women who exercised maintained trabecular BMD of the spine whereas the women who remained sedentary lost bone at this site, with dietary calcium having no effect. However, calcium intake did have an effect on the BMD of the femur. The women who consumed high dietary calcium maintained femoral BMD whereas those on moderate amounts showed a decrease. There was no effect of either intervention on the BMD of the distal radius or on TBCa.

Several prospective exercise studies showed various results depending on the study group chosen, exercise intervention, and measurement site (9–12). Dalsky et al (9), in a study involving postmenopausal women aged 55–70 y, found that weight-bearing exercises, such as walking, jogging, or stair climbing, over a period of 9 or 20 mo led to significant increases in lumbar spine BMD (measured by DPA). By 9 mo the exercise group had a 5.2% increase in spine BMD and the sedentary control group had a 1.4% decrease for a total 6.6% change between the two groups. This change was similar to the effects seen between the exercise group (1.2% increase) and the sedentary group (6.0% decrease). However, in the present study this change was only seen in the trabecular bone of the spine measured by QCT and not in measurement of the spine by DPA. In addition, we did not see an increase in bone mass, only a preservation of bone mass in the exercise group.

The differences in the effect of exercise between the two studies may be due to the intensity of the exercise and estrogen therapy. The exercise protocol used in the study by Dalsky et al (9) was more varied and more intense than our exercise protocol. Our exercise included only walking at 75–80% of maximum heart rate, whereas the Dalsky et al study included jogging, stair climbing, weight training (bench press), rowing, and cycling in addition to walking. Also, Dalsky et al did not exclude women who were on estrogen therapy, which may be why they saw a large increase in the exercise group and only mild decreases in the sedentary group.

The only other prospective walking study that used QCT of the spine to measure changes over time was a study by Cavanaugh and Cann (10). In this study a group of postmenopausal women aged 49–64 y walked briskly for 15–40 min/d for 3 d/wk for 12 mo. However, this study failed to show a difference in the rate of bone loss between exercising and control subjects. In the present study the sample size was larger and the exercise was greater in intensity, duration, and frequency. This may account for some of the differences between studies.

There have been no prospective studies investigating the influence of exercise on femoral BMD. We initially hypothesized that exercise would influence the femoral BMD on the basis of the high amount of mechanical stress in this region. The spine data suggest that there is a specific effect of exercise on trabecular bone. It is possible that exercise had an effect on trabecular bone of the femur; however, this study would not have been able to detect this effect because DPA does not provide an independent measure of trabecular bone. Thus, the disparity between our exercise study and the studies of others (9–12) may be due to the exercise protocol employed, duration of the study, age group chosen, and the method and site used to determine bone status.

Low dietary calcium intake was suggested as a major factor in the development of bone loss in older women (3, 5, 38). However, the balance of current evidence suggests that in the postmenopausal years calcium alone has only a small effect on cortical bone and no effect on trabecular bone (39). Indeed, the present study did detect a positive effect of high dietary calcium on the femoral neck, a site of high proportion of cortical bone, and there was no interaction with exercise. We did not, however, detect any effect of high dietary calcium on TBCa or distal radial BMD, two other sites with a high proportion of cortical bone.

In a 2-y prospective study, Riis et al (40) found that calcium supplementation (2000 mg CaCO$_3$/d) in early postmenopausal women (51 y) decreased bone loss in the proximal forearm and there was a trend towards decreased bone loss from the entire skeleton as measured by DPA. The rate of bone loss from the distal forearm and lumbar spine (L$_2$–L$_4$), measured by DPA, was the same for the calcium-supplemented group as for the placebo group. They did not measure the BMD of the femur. The results of our study are similar in that we did not show an effect of calcium on the distal forearm or spine. However, we did not show any tendency of dietary calcium to slow the rate of bone loss from the entire skeleton. This may be due to the older age of our subjects. Furthermore, the 2000 mg/d of supplementation over a 2-y period was in addition to the normal calcium intake in their diet.

From the present study we conclude that high dietary calcium (~1450 mg/d) resulted in reduced bone loss from the femoral neck. The disparity between our study and the studies of others (41) is most likely a result of the sample size used, the duration of the study, the age of the subjects, and the method and site used to determine bone status.

Developing a high-dietary-calcium drink whose calcium source was milk and a placebo drink with little calcium that

tasted and looked similar to the milk drink was difficult. Consequently, the two drinks did not match in protein, fat, and energy content. Of greatest concern was the difference in protein content. Very high dietary protein may effect calcium metabolism and requirements (42). However, protein and phosphorus have opposing effects on urinary calcium and calcium retention. A simultaneous increase in the intake of both, a pattern characterized by high milk consumption, is thought to have little effect on calcium balance at recommended amounts of calcium intake (43). Another nutrient known to affect calcium metabolism is vitamin D. The results from the poststudy food records indicated that the women in HICA had more vitamin D in their diets than did the women in MODCA. If the vitamin D in the diet affected calcium metabolism, changes in serum 25(OH)D or 1,25(OH)$_2$D concentrations would have been observed; however, this did not occur.

For the reasons outlined above, we are confident that the dietary changes (other than calcium) that were created by the drinks had little effect on overall calcium metabolism.

No changes over time between the exercise and dietary calcium groups were observed for any hormone or urinary metabolite. This may be a consequence of the small study size, short study duration, and large variability in the different measurements. In a previous study in our laboratory, Nelson et al (44) showed that postmenopausal masters athletes ($\bar{x}$ age 62y) had higher 1,25(OH)$_2$D and SOM-C concentrations than did sedentary control subjects. Both factors favor calcium absorption and bone formation (45, 46). The women in this cross-sectional study were highly trained, had lower body weights and lower body fat, had started training before menopause, and for the most part were competitive masters athletes with an average aerobic capacity of 38.8 ± 1.7 mL O$_2 \cdot$ kg$^{-1} \cdot$ min^{-1}. Although the exercisers in the present study did increase their aerobic fitness by 8.8% by walking, they did not attain the high aerobic fitness of the long-term masters athletes.

These results suggest that exercise and high dietary calcium may have beneficial effects at various skeletal sites. Specifically, exercise by walking in this age group of women decreased bone loss from the trabecular bone of the spine whereas high dietary calcium decreased bone loss from the femoral neck. Sedentary women should be encouraged to adopt a calcium-rich diet and weight-bearing exercise to maintain the health of their skeleton. Both of these interventions are practical and feasible for healthy women and should be encouraged at the earliest possible age.

References

1. Peck WA, Riggs BL, Bell NH, eds. Physicians' resource manual on osteoporosis: a decision making guide. Washington, DC: National Osteoporosis Foundation, 1987.

2. Holbrook TL, Grazier K, Kelsey JL, Stauffer RN. Frequency of occurrence, impact and cost of selected musculoskeletal conditions in United States. Chicago: American Academy of Orthopaedic Surgeons, 1984.

3. National Research Council. Diet and health: implications for reducing chronic disease risk. Washington, DC: National Academy Press, 1980.

4. Marcus R. Understanding and preventing osteoporosis. Hosp Prac [off] 1989:189–218.

5. Walden O. The relationship of dietary and supplemental calcium intake to bone loss and osteoporosis. J Am Diet Assoc 1989;89:397–400.

6. Sinaki M. Exercise and osteoporosis. Arch Phys Med Rehabil 1989;70:220–9.

7. Wolff J. The law of transformation of bones. Berlin: Hirshwald, 1892 (in German).

8. Issekutz B, Blizzard JJ, Birkhead NC, Rodahl K. Effect of prolonged bed rest in urinary calcium output. J Appl Physiol 1966;21:1013–20.

9. Dalsky GP, Stocke KS, Ehsani AA, et al. Weight-bearing exercise training and lumbar bone mineral content in postmenopausal women. Ann Intern Med 1988;108:824–8.

10. Cavanaugh DJ, Cann CE. Brisk walking does not stop bone loss in postmenopausal women. Bone 1988;9:201–4.

11. Smith EL, Gilligan C, McAdam M, Ensign CP, Smith PE. Deterring bone loss by exercise intervention in premenopausal and postmenopausal women. Calcif Tissue Int 1989;44:312–21.

12. Aloia JF, Cohn SH, Ostuni JA, Cane R, Ellis K. Prevention of involutional bone loss by exercise. Ann Intern Med 1978;89:356–8.

13. Matkovic V, Kostial K, Simonvic I, Buzina R, Brodarec A, Nordin BE. Bone status and fracture rates in two regions of Yugoslavia. Am J Clin Nutr 1979;32:540–9.

14. Heaney RP, Gallagher JC, Johnston CC, Near R, Parfitt AM, Whedon GP. Calcium nutrition and bone health in the elderly. Am J Clin Nutr 1982;36:986–1013.

15. Riggs BL, Wahner HW, Melton LJ, O'Fallon WM, Judd HL, Richelson LS. In women dietary calcium intake and rates of bone loss from midradius and lumbar spine are not related. J Bone Miner Res 1986;1(suppl 1)96(abstr).

16. Boren HG, Kory RC, Syner JC. The veterans administration-army cooperative study of pulmonary function. Am J Med 1966;41:96–114.

17. Siri WE. Body composition from fluid spaces and density: analysis of methods. In: Brozek J, Henschel A, eds. Techniques for Measuring Body Composition. Washington, DC: National Academy of Sciences, 1961:223–44.

18. Bruce RA. Exercise testing of patients with coronary heart disease. Ann Clin Res 1971;3:323–32.

19. Scholander PF. Analyzer for accurate estimation of respiratory gases in one-half cubic centimeter samples.

20. Peppler WW, Mazess RB. Total body bone mineral and lean body mass by dual-photon absorptiometry. I. Theory and measurement procedure. Calcif Tissue Int 1981;33:353–9.

21. Riggs BL, Wahner HW, Seeman E, et al. Changes in bone mineral density of the proximal femur and spine with aging: difference between the postmenopausal and senile osteoporosis syndromes. J Clin Invest 1982;70:716–23.

22. Dawson-Hughes B, Deehr MS, Berger PS, Dallal GE, Sadowski LJ. Correction of the effects of source, source strength, and soft tissue thickness on spine dual-photon absorptiometry measurements. Calcif Tissue Int 1989;44:251–7.

23. Cann CE. Low-dose CT scanning for qualitative spinal mineral analysis. Radiology 1981;140:813–5.

24. Cameron JR, Sorenson J. Measurement of bone mineral in vivo: an improved method. Science 1961;142:230–2.

25. Wahner HW, Dunn WL, Riggs L. Assessment of bone mineral part 1. J Nucl Med 1984;25:1134–41.

26. Cohn SH, Dombrowski CS, Pate HR, Robertson JS. A whole-body counter with an invarient response to radionuclide distribution and body size. Phys Med Biol 1969;14:645–58.

27. Dilmanian FA, Weber DA, Yasumura S, et al. The performance of the BNL delayed- and prompt-gamma neutron activation systems. In: Yasumura S, Harrison JE, McNeill KG, Woodhead AD, Dilmanian FA, eds. In vivo body composition studies: recent advances. New York: Plenum Press, 1990:309–16.

28. Shukla KK, Ellis, KJ, Dombrowski CS, Cohn SH. Physiological variation of total-body potassium in man. Am J Physiol 1973;224:271–4.

29. Longcope C, Watson DJ, Williams KIH. The effects of synthetic estrogens on the metabolic clearance and production rates of estrone and estradiol. Steroids 1974;24:15–30.

30. Reinhold JG, Faradji B, Abadi P, Ismail-Bergi F. Decreased absorption of calcium, magnesium, zinc and phosphorus by humans due to increased fiber and phosphorus consumption as wheat bread. J Nutr 1976;106:493–503.

31. Larsen K. Creatinine assay by a reaction kinetic principle. Clin Chim Acta 1972;41:209–17.

32. Wassner SJ, Schlitzer JL, Li JB. A rapid, sensitive method for the determination of 3-methylhistidine levels in urine and plasma using HPLC. Anal Biochem 1980;104:284–9.

33. Haroon Y. Rapid assay for gamma-carboxyglutamic acid in urine and bone by precolumn derivatization and reversed-phase chromatography. Anal Biochem 1984;140:343–8.

34. Helwig JT. SAS introductory guide. Cary, NC: SAS Institute Inc, 1978.

35. Zamenhof RGA. Techniques for bone mineral measurement. Radiology 1989;1:1–11.

36. Riggs BL, Melton LJ. Involutional osteoporosis. N Engl J Med 1986;314:1676–86.

37. Cochet B, Jung A, Griessen M, Bartholdi P, Schaller P, Donath A. Effects of lactose on intestinal calcium absorption in normal and lactose-deficient subjects. Gastroenterology 1983;84:935–40.

38. Dawson-Hughes B, Dallal GE, Krall EA, Sadowski L, Sahyoun N, Tannenbaum S. A controlled trial of the effect of calcium supplementation on bone density in postmenopausal women. N Engl J Med 1990;323:878–83.

39. National Research Council. Recommended dietary allowances. 10th ed. Washington, DC: National Academy Press, 1989.

40. Riis B, Thomsen K, Christiansen C. Does calcium supplementation prevent postmenopausal bone loss? A double-blind, controlled clinical trial. N Engl J Med 1987;316:173–7.

41. Smith EL, Gilligan C, Smith PE, Sempos CT. Calcium supplementation and bone loss in middle-aged women. Am J Clin Nutr 1989;50:833–42.

42. Margen S, Chu JY, Kaufmann NA, Calloway DH. Studies in calcium metabolism I. The calciuretic effect of dietary protein. Am J Clin Nutr 1974;27:540–9.

43. Spencer H, Kramer L, Osis D. Do protein and phosphorus cause calcium loss? J Nutr 1988;118:657–60.

44. Nelson ME, Meredith CN, Dawson-Hughes B, Evans WJ. Hormone and bone mineral status in endurance-trained and sedentary postmenopausal women. J Clin Endocrinol Metab 1988;66:927–33.

45. Gallagher JC, Riggs BL, Eisman J, Hamstra A, Arnaud SB, DeLuca HF. Intestinal calcium absorption and serum vitamin D metabolites in normal subjects and osteoporotic patients: effect of age and dietary calcium. J Clin Invest 1979;64:729–36.

46. Bennet AE, Wahner HW, Riggs BL, Hintz RL. Insulin-like growth factors I and II: aging and bone density in women. J Clin Endocrinol Metab 1984;59:701–4.