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Associations between objectively-measured sedentary behaviour and physical activity with bone mineral density in adults and older adults, the NHANES study

S.F.M. Chastin [a,*], O. Mandrichenko [a], J.L. Helbostadt [b], D.A. Skelton [a]

[a] Glasgow Caledonian University, School of Health and Life Sciences, Glasgow, UK
[b] Norwegian Institute of Science and Technology, Department of Neuroscience, Trondheim, Norway

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ABSTRACT

Background: Lack of physical activity (PA) is an important modifiable risk factor for bone mineral density (BMD). Time spent in sedentary behaviour (SB), or time spent in non-exercising seated and reclining postures, has recently emerged as a new public health risk, independent of the amount of time someone spends being active. As national surveys report that adults spend on average 8 h per day being sedentary, rising to 10 h a day in older age, it has been hypothesised that a repeated exposure to sitting in modern daily life, whether it is for travelling, working or leisure, might have a deleterious effect on bone health in a way that mirrors the results of studies into the effect of lengthy periods of bed-rest. The aim of this study was to investigate for the first time a) how time spent in SB is associated with bone mineral density (BMD), b) whether this association changes depending on the amount of time spent engaging in different intensity levels of PA, and c) if the pattern of accumulation of SB and long uninterrupted periods of SB are associated with BMD.

Methods: The 2005/2006 National Health and Nutrition Examination Survey (NHANES), is a cross-sectional study of a representative sample of the US population that is conducted biannually by the National Centers for Disease Control. PA and SB were assessed objectively over 7 days using an Actigraph accelerometer and BMD was measured via dual-energy X-ray absorptiometry. In this study, data are presented on four regions of the femur (femoral neck, trochanter, inter trochanter and total femur) and total spine (L1–L4). The associations between BMD, SB and PA levels were examined using multiple linear regressions stratified by gender. In addition, the association between the pattern of accumulation of SB (quantified as frequency and duration of SB) and BMD was also investigated. All models were adjusted for known risk factors associated with BMD. In total, data for 2117 individuals, aged 23–90+ years (males N = 1158), were available to analyse SB and femur BMD and 1942 individuals (males N = 1053) for analysis of SB and spine BMD.

Results: There was no evidence of an association between SB time and hip or spinal BMD in men. For men, time spent doing moderate to vigorous activity (MVPA) and vigorous activity (VIG) was associated with higher total femur and the other hip sub-region BMD. The regression coefficient was $B_{MVPA} = 0.306$ (95% CI: 0.021–0.591) g/cm^2 for each 10 minute increment in daily MVPA. For VIG, the regression coefficient is $B_{VIG} = 0.320$ (95% CI: 0.058–0.583) but this cannot be interpreted linearly as time spent in vigorous activity was square root transformed. In women, SB was negatively associated with total femur BMD and all sub-regions but not MVPA nor VIG. The regression coefficient for total femur BMD was $B_{SB} = -0.159$ (95% CI: −0.241–0.076) g/cm^2 for each 10 minute increment spent being sedentary each day. In addition, the duration of SB bouts was deleteriously associated with BMD for the total femur and of other hip sub-regions, but the number of bouts of SB did not have a significant effect. These associations were found to be independent of the amount of MVPA and VIG that women engage in. No associations were found between SB or PA and spinal BMD for either men or women.

Conclusions: These results provide the first evidence that repeated exposure to sitting (SB), measured objectively in daily life, is deleteriously associated with BMD of the total femur and of all hip sub-regions in women, independent of the amount of time women engage in moderate and vigorous activity. This suggests that SB might be a risk factor for bone health in women independent of whether they engage in physical activity. In addition, the duration of SB bouts, rather than their frequency, appears to be deleteriously associated with BMD of the total femur and of all hip sub-regions. Future research should investigate the effect on bone health of interventions which set out to reduce SB and the duration of SB bouts in comparison, and as adjunct, to the promotion of PA.

* Corresponding author at: Glasgow Caledonian University, School of Health and Life Sciences, Cowcaddens Road, Glasgow G4 0BA, UK.
E-mail address: sebastien.chastin@gcu.ac.uk (S.F.M. Chastin).

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For men, SB is not significantly associated with BMD of the femur or spine and the results appear to confirm that moderate and vigorous activity has a protective effect.

Introduction

Osteoporotic fractures are a major public health issue and a growing concern with an ageing population. Loss of bone mass is part of the normal ageing process, [1,2] and is determined by factors such as gender [3], ethnicity [4,5] and genetics [6]. However a number of modifiable health behaviours, such as smoking [7], dietary intake [8] and exercise [9], also influence the rate of bone mineral density (BMD) loss and the development of osteoporosis in older age. To reduce the risk of fractures and their associated cost to society, as well as the impact on an individual's health and quality of life, it is important to identify modifiable risks associated with poor bone health. It has long been recognised that long periods of enforced inactivity, reduced weight bearing and muscle loading, such as bed rest [10,11] and time spent in reduced gravity [12,13], change the bone turnover and mineral homeostasis. After only a few weeks the bone loss is equivalent to that which would be expected in a decade of normal ageing [14]. These studies suggest that bed rest and weightlessness have two distinct effects on bone health. Firstly, there is a direct physiological increase in bone resorption [10] and secondly, there is an indirect effect through the decreased stimulation of bone formation [15,16], caused by the lack of physical activity (PA), which modulates the amount and quality of bone that is produced [17].

The effect of long periods of reduced weight-bearing such as enforced bed rest and weightlessness in space might seem irrelevant for the majority of the population. However modern societal and technological changes have dramatically increased the amount of time spent in low impact and reduced weight-bearing postures in everyday life [18]. Sedentary behaviours (SBs), defined as the time spent in sitting or reclining postures and involving low energy expenditure [19], are ubiquitous in modern lifestyles during leisure time, work and transportation [20]. National surveys show that adults spend on average 8 h of the waking day being sedentary [21], rising to 10 h in older age [22], with 67% of older adults spending more than 8.5 h a day sitting [23]. Temporal patterns of SB show that the majority of this sedentary time is accumulated in a small number of long uninterrupted bouts, some of which last several hours [24].

It is conceivable that repeated exposure to reduced weight-bearing activity during daily life, especially in long continuous bouts, might have a similar effect on bone metabolism that single bouts of prolonged bed-rest or weightlessness have. The hypothesis is that detrimental effects of SB on bone health could stem from two possible factors: a lack of musculature activation and unloading of bone structure as in reduced gravity [25]. Recent studies in adolescents suggest that there is, indeed, an association between bone mineral density and time spent in sedentary pursuits such as watching TV and spending time sitting in front of a computer [26–28]. Furthermore, this association was found to be only partially counteracted by the engagement in oesteogenic physical activity throughout the rest of the day. In a study of white older women aged 65 and over, the risk of fracture was found to double where those individuals spent 4 h or less standing per day, while the fracture risk was only 30% lower for those who walked regularly [29]. However, this study used self-reported measures of sitting and lying time which are imprecise and may affect estimations of the effect of SB on bone health [30]. To date, a potential association between objectively-measured SB and bone health in adulthood has not been investigated.

We used data from the National Health and Nutritional Examination Survey 2005–06 (NHANES) to explore the cross-sectional associations between SB and BMD.

The objectives of this study were to examine: a) whether total time spent in SB is associated with BMD of the hip and lumbar spine, b) if the effects of SB on BMD are compounded when accumulated in long uninterrupted bouts and c) whether this association changes depending on the amount of time spent at different PA intensities.

Method

Study

The National Health and Nutrition Examination Survey (NHANES) is a cross-sectional study conducted annually by the National Center for Health Statistics at the Centers for Disease Control and Prevention (CDC). It uses a complex, multi-stage probability design to obtain a representative sample of the USA civilian non-institutionalized population over a two-year cycle. Details of the surveys and NHANES methods are available from the CDC website. The measurement of BMD via dual-energy X-ray absorptiometry (DXA) has been part of the survey since NHANES III and serves as a reference value for the diagnosis of osteoporosis [31]. Concurrent objective measurement of PA and SB with accelerometry and DXA is available for the 2003–4 and 2005–6 cycles of NHANES. However the NHANES DXA data for the 2003–4 cycle contains a systematic and non-random pattern of missing data and the CDC released a set of imputed values for this cycle. For this study, only the data free of imputation is included and therefore uses data from the 2005–6 cycle. The NHANES study complies with the Declaration of Helsinki and the National Center for Health Statistics Ethics Review Board approved the protocols.

Study sample

In the NHANES 2005–6 cycle, there were 4775 adults over the age of 22 in a total population of 10348 individuals. Amongst these 4775 adults, 4206 wore an accelerometer for 7 days during waking hours but valid accelerometry data were available for only 2635 individuals (see Section 2.4 and Fig. 1). In addition, valid DXA measurements were available for only 3297 individuals for the femur region and 3096 individuals for the spine region. In this analysis, only subjects with valid DXA, accelerometry and covariate data were included. In total, 2117 individuals (males, N = 1158) were included to analyse femur BMD and 1942 individuals (males, N = 1053) to analyse spine BMD (Fig. 1).

Bone mineral density measurement

BMD of the proximal femur and the lumbar (L1–L4) spine was measured using dual-energy X-ray absorptiometry. The DXA scans were performed with Hologic QDR-4500A fan-beam densitometers (Hologic, Inc., Bedford, Massachusetts) by trained and certified radiology technologists. The scans were analysed using the Hologic software, APEX v3.0, which has been shown to have good precision [32]. A high level of quality was maintained throughout the data collection with a rigorous quality control protocol, which included regular anthropomorphic phantom scan checks. Further details of the DXA data-acquisition protocol are described in the Body Composition Procedures Manual on the NHANES website.

In this study, data are presented on four regions of the femur: femoral neck, trochanter, inter trochanter and total femur. NHANES also has data for the Ward's triangle, but this has not been included in this analysis as it has a larger measurement error in living subjects than other femoral sub-regions [33]. As recommended by

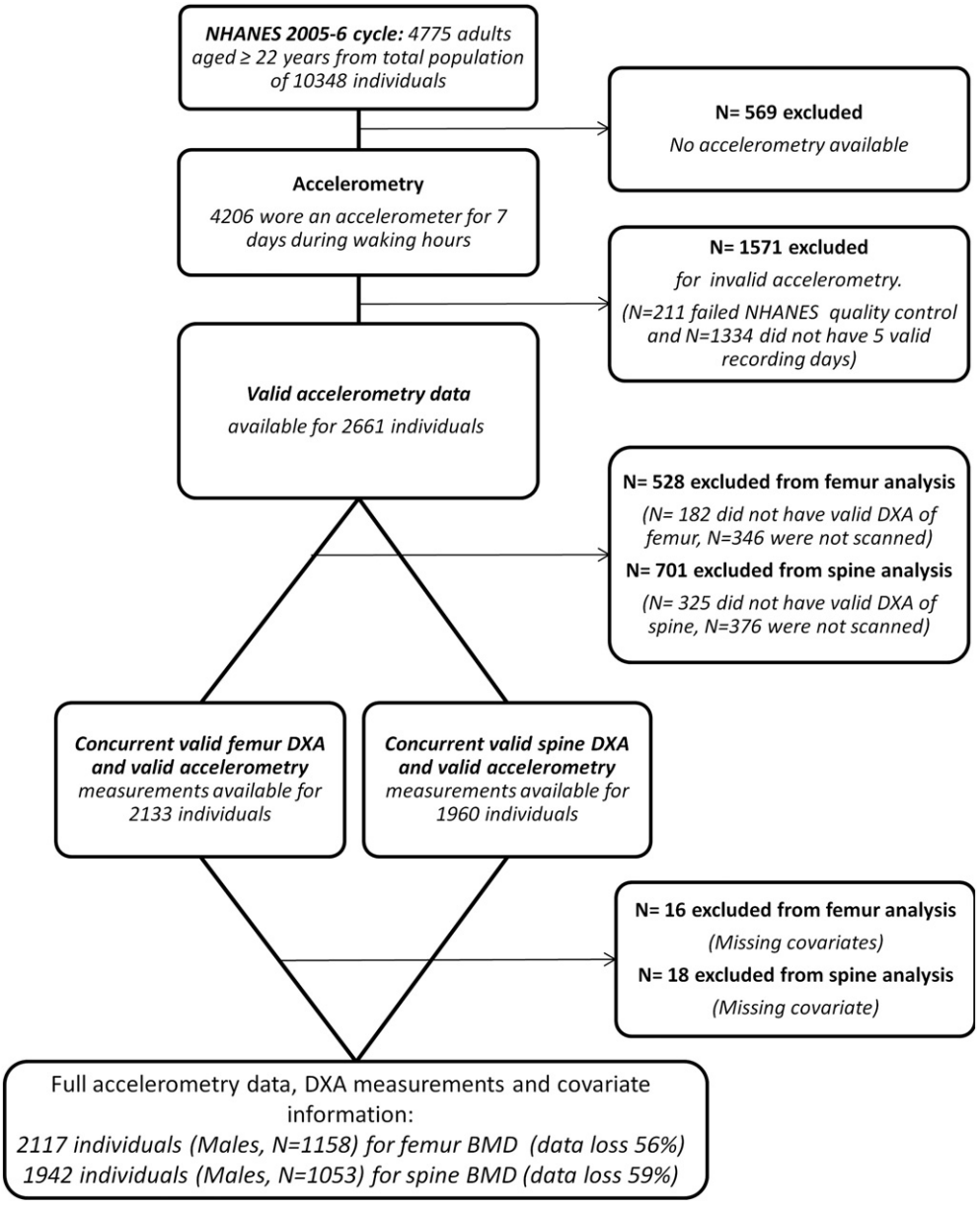

Fig. 1. Study inclusion flow chart.

the International Society for Clinical Densitometry [34], in this study only the total spine (L1–L4) BMD, rather than individual vertebrae levels, was examined.

Physical activity and sedentary behaviour monitoring

All ambulatory participants in the 2005–6 cycle of NHANES were eligible and asked to wear an Actigraph accelerometer (Actigraph 7164; Actigraph, LLC, Fort Walton Beach, FLA). The Actigraph accelerometer is a small (5.1 × 4.1 × 1.5 cm), lightweight (0.4 kg) device, worn on the hip that records acceleration information integrated as an activity count per 1 minute epoch. This monitor provides an objective estimate of the intensity of bodily movement (particularly ambulatory locomotion). Thresholds obtained from calibration studies enable the translation of accelerometer counts per minute (cpm) into physical activity intensity [21,35].

The accelerometer was worn for 7 days during waking hours (except for water-based activities). The devices were returned by mail to NHANES and data were downloaded and checked to ascertain if the device was still calibrated. Further details on the objective physical activity protocol can be found on the NHANES website.

Accelerometry data was first screened to exclude data retrieved from monitors that were not calibrated and data identified as not meeting the NHANES quality control (N = 211 were excluded because of this criteria). An automated programme [21] was adapted and used to implement these quality control procedures and isolate the time when the device was not worn. The standard definition of non-wear time from the CDC was adopted. This defines non-wear time as intervals of at least 60 consecutive minutes of 0 cpm, with allowances for up to 2 min of limited movement (<50 cpm) within these periods. Recorded days with at least 10 h of continuous wear time that did not contain spurious and excessive high counts (>20000 cpm) were considered valid. Individuals with at least 5 valid recording days of data, including at least one weekend day, were included in the analyses in line with current best practice in physical activity monitoring [36]. 1360 participants were excluded because of this criterion, and thus, a total of 2635 individuals with valid accelerometry data according to these criteria were included.

Each 1 minute epoch of accelerometry data was classified according to calibration equations [21,35] as sedentary (SB) if <100 cpm, light intensity activity (LIPA) if between 100–1951 cpm, moderate–vigorous intensity physical activity (MVPA) if between 1952–5724 cpm and vigorous physical activity (VIG) if >5724 cpm. For each valid day, adjacent epochs in the same class were aggregated into bouts. The length of the bout was equal to the number of epochs grouped together. All processing was done using MATLAB R11b (Mathworks Ltd).

Physical activity and sedentary behaviour time and pattern outcomes

From the accelerometry data the total volume of PA and SB and the pattern of accumulation of SB were extracted.

Total time

The total daily time spent in SB and the different PA levels (LIPA, MVAP, VIG) were obtained by totalling the duration of all the bouts at each level for each day. The values were normalised to total wear time and averaged over the number of valid days to derive an estimate of the mean time spent in SB and each PA level per day. Total time is presented as a percentage of the waking day.

Pattern of accumulation

The pattern of time spent in SB was characterised through the estimates of the average duration and frequency of daily bouts of SB. Estimation of frequency is obtained by computing the average number of daily bouts of SB. The estimation of the average duration of SB bout is computed according to [24]. SB bout durations are distributed as a power law and therefore standard statistical estimates, such as mean or median, are not reliable estimators. Instead, the average SB bout duration can be estimated by the non-linear fitting of the SB accumulation curve (accrued SB time as a function of bout length) to a sigmoid function of the form $t^n / (t^n + X^n)$ where t is the bout length, n is a free parameter and X is the average sedentary bout duration estimate.

Covariates

The NHANES 2005–6 cycle database was researched for information about known risk factors that are associated with lower BMD (age, smoking habits, BMI, ethnicity) and other potential confounders (intake of calcium, alcohol consumption, vitamin D levels, use of prednisone, family history of osteoporosis and levels of parathyroid hormones). Age (years) at time of screening, race and ethnicity, self-reported health and co-morbidities were obtained from the interviewer-administered demographic questionnaires. NHANES coded race and ethnicity in five categories; Mexican American, other Hispanic, non-Hispanic white, non-Hispanic black and other races including mixed multi-racial. BMI (kg/m^2) values were calculated by NHANES from measurement of height and weight taken during the physical examination of the participants. Smoking habits were inferred from serum-cotinine levels (ng/mL). Levels of serum parathyroid hormones (pg/mL) and serum vitamin D (ng/mL) were also obtained from laboratory analysis of serum samples collected during the NHANES 2005–6 cycle. All other covariates were self-reported via questionnaires. Alcohol intake habits were quantified as a continuous variable (the number of days in the last 12 months that alcohol was consumed). Calcium intake was deduced from dietary questionnaires and coded on a 5 point ordinal scale. Dichotomous variables were generated for the use of prednisone and the family history of osteoporosis, using questionnaires which asked about family history of osteoporosis and use of prednisone.

Statistical data analysis

All analyses were carried out with SPSS version 18 (IBM, Chicago, IL). The representativeness, in terms of age, gender, BMI, self-reported health status, co-morbidities and ethnic distribution, of the sample analysed, compared to the NHANES 2005–6 population, was checked using proportion and Chi square test for gender, self-reported health status, number of co-morbidities and ethnicity and non-parametric Wilcoxon rank test for age and BMI. In addition, potential selection bias resulting from adherence to accelerometry was assessed by comparing the age, gender, BMI, self-reported health status, co-morbidities and ethnic distribution of the included and excluded samples. This was conducted using proportion and Chi square test for gender, self-reported health status, number of co-morbidities and ethnicity and non-parametric Mann–Whitney *U* test for age and BMI. Associations between BMD, SB and PA levels were examined using multiple linear regressions, with BMD as the dependent variable. The analysis was stratified by gender and by sub-region. The amount of time spent in SB (Model 1) and in each PA intensity level (LIPA Model 2, MVPA Model 3, VIG Model 4) was entered individually as independent variables to ascertain associations with BMD in isolation. Model 1 was then adjusted for time spent in different levels of PA intensity (Model 5–6) to examine if the association between SB and BMD is changed by time spent in the different levels of PA intensity. In Model 5, time spent in SB and time spent in MVPA were both entered as independent variables and in Model 6 the time spent in VIG replaced MVPA. Models 7 and 8 examined whether there is an association between the pattern of accumulation of sedentary time and BMD. Frequency and duration of sedentary bouts were entered as independent variables in Models 7 and 8 respectively.

All the models were adjusted for known risk factors associated with lower BMD (age, smoking, body mass index and ethnicity) and additional potential confounders (vitamin D in blood, calcium intake, parental history of osteoporosis, use of prednisone, levels of parathyroid hormones and alcohol intake). Models for each gender and sub-region were adjusted differently to account only for factors that were significant predictors. Covariates were included in models if they were associated with BMD at a $p < 0.05$ significance level.

All continuous variables were checked for normality before being entered in the models, root square transformation was used to normalise time spent in vigorous activity. For each regression model, the linearity of the association between predictors and outcomes as well as all other required data conditions were examined. The multi-collinearity between independent variables was checked by performing variance inflation tests (VIF). It is considered that a VIF score greater than 10 indicates the presence of collinearity [37]. The VIF score exceeded this threshold when total SB and LIPA were entered in the same model, due to the strong correlation between SB and LIPA time ($r > 0.98$). These models were therefore discarded.

Sensitivity analysis was conducted to test for potential effects of selection bias due to adherence to accelerometry. The analysis was repeated by weighting the regression using inverse probability weighting based on the probability of selection in terms of BMI, self-reported health and co-morbidities.

Results

A sample average for the total amount of time and the pattern of SB, together with the average time spent at different physical activity intensities, is presented per gender in Table 1.

Results of modelling the relationships between of SB, PA and femur BMD are shown in Table 2 for men and Table 3 for women.

Sample

In the included sample the proportion of males over the age of 22 years was 54.7% compared to 48.1% in the NHANES 2005–6 cycle ($\chi^2 = 66.27$, $p < 0.001$). The median age of the sample analysed in this study, 52 years, was significantly higher ($p < 0.001$) than the median age of 47 (for adults over the age of 22) in the NHANES 2005–6 cycle. A significant difference in ethnic distribution was also

Table 1
Sample sedentary behaviour and physical activity profile, femur and spinal BMD and BMI per decade.

	Age (years)	SB (% of day)		LIPA (% of day)		MVPA (% of day)		VIG (% of day)		SB frequency (bouts)		SB bout duration (min)	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Men	22–29	52.4	(13.8)	42.4	(11.9)	4.9	(4.1)	0.3	(0.1)	92.5	(18.8)	4.89	(1.76)
	30–39	54.0	(13.4)	41.7	(12.4)	4.1	(2.9)	0.2	(0.1)	94.3	(18.5)	5.15	(2.02)
	40–49	53.3	(12.3)	42.3	(11.0)	4.3	(3.4)	0.2	(0.1)	96.6	(17.4)	4.89	(1.47)
	50–59	58.2	(11.9)	38.7	(10.9)	2.9	(2.3)	0.1	(0.1)	94.3	(19.3)	5.69	(2.02)
	60–69	60.6	(11.1)	37.2	(10.3)	2.1	(2.0)	0.1	(0.1)	88.3	(19.2)	6.15	(2.04)
	70–79	68.3	(10.5)	30.5	(10.0)	1.2	(1.2)	0.03	(0.03)	80.9	(17.4)	7.24	(2.63)
	80+	72.5	(11.0)	27.1	(11.0)	0.7	(0.7)	0.004	(0.004)	77.5	(19.5)	9.07	(4.37)
Women	22–29	56.6	(9.3)	40.9	(8.9)	2.3	(1.8)	0.1	(0.1)	98.4	(15.6)	4.80	(1.14)
	30–39	55.6	(10.5)	42.1	(10.0)	2.3	(1.9)	0.1	(0.1)	99.9	(16.3)	4.82	(1.48)
	40–49	55.0	(10.5)	42.3	(9.9)	2.5	(2.0)	0.2	(0.1)	99.5	(16.2)	4.83	(1.30)
	50–59	57.9	(9.9)	40.4	(9.4)	1.7	(1.4)	0.03	(0.03)	97.9	(16.7)	5.23	(1.50)
	60–69	60.4	(11.7)	38.3	(10.9)	1.3	(1.3)	0.02	(0.02)	91.9	(17.3)	5.82	(2.14)
	70–79	65.0	(11.9)	34.1	(11.5)	0.9	(0.9)	0.002	(0.002)	89.5	(18.2)	6.79	(3.50)
	80+	71.1	(10.4)	28.5	(9.9)	0.3	(0.3)	0.001	(0.001)	84.4	(19.3)	7.58	(2.71)

	Age (years)	BMI (kg/m^2)		Total femur BMD (g/cm2)		Femoral neck BMD (g/cm2)		Trochanter BMD (g/cm2)		Inter-trochanter BMD (g/cm2)		Spine (L1–L4) BMD (g/cm2)	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Men	22–29	26.3	(4.6)	1.10	(0.14)	0.98	(0.14)	0.83	(0.12)	1.29	(0.17)	1.09	(0.12)
	30–39	28.2	(4.7)	1.06	(0.13)	0.92	(0.13)	0.79	(0.11)	1.25	(0.16)	1.04	(0.12)
	40–49	28.0	(4.3)	1.05	(0.14)	0.89	(0.13)	0.80	(0.11)	1.24	(0.16)	1.04	(0.14)
	50–59	28.3	(4.9)	1.02	(0.15)	0.85	(0.14)	0.78	(0.14)	1.20	(0.18)	1.05	(0.15)
	60–69	28.3	(4.9)	1.02	(0.15)	0.82	(0.14)	0.78	(0.13)	1.20	(0.18)	1.08	(0.16)
	70–79	27.6	(4.0)	0.96	(0.14)	0.78	(0.13)	0.74	(0.13)	1.13	(0.16)	1.07	(0.17)
	80+	25.8	(4.2)	0.92	(0.14)	0.73	(0.14)	0.73	(0.15)	1.08	(0.19)	1.07	(0.20)
Women	22–29	27.4	(6.0)	0.96	(0.13)	0.86	(0.12)	0.71	(0.11)	1.12	(0.14)	1.03	(0.13)
	30–39	28.9	(6.2)	0.97	(0.13)	0.86	(0.12)	0.70	(0.11)	1.14	(0.15)	1.07	(0.11)
	40–49	29.0	(6.7)	0.97	(0.14)	0.85	(0.14)	0.73	(0.12)	1.14	(0.16)	1.05	(0.15)
	50–59	29.9	(7.1)	0.90	(0.14)	0.78	(0.13)	0.67	(0.12)	1.07	(0.17)	0.99	(0.15)
	60–69	29.6	(5.8)	0.87	(0.14)	0.75	(0.13)	0.65	(0.12)	1.02	(0.16)	0.95	(0.15)
	70–79	27.3	(5.9)	0.79	(0.14)	0.67	(0.12)	0.59	(0.11)	0.94	(0.17)	0.89	(0.15)
	80+	27.7	(4.9)	0.73	(0.14)	0.61	(0.13)	0.55	(0.11)	0.86	(0.17)	0.89	(0.17)

SB = Sedentary behaviour time; LIPA = light intensity physical activity; MVPA = moderate to vigorous physical activity; VIG = vigorous physical activity.

found between the sample analysed and the NHANES population ($\chi^2 = 19.15$, $p = 0.001$). The sample analysed has a higher proportion of non-Hispanic whites by 2.8% and Mexican Americans by 0.4% compared to the NHANES population. The non-Hispanic black, other Hispanic and other race groups were under represented by 0.3%, 2.6% and 0.3% respectively in the analysis sample compared to the NHANES population. Despite the fact that the median BMI (27.6 kg/m^2) of the included sample is similar to that of the total NHANES 2005–6 cycle population (27.6 kg/m^2) or that of the excluded sample (27.8 kg/m^2), a significant difference ($p < 0.001$) was found in terms of BMI distribution. The included sample had 13% fewer obese and 10% fewer underweight participants compared to the NHANES population and excluded sample. Similarly, the sample analysed had less participants (7%) with poor self-reported health than expected from the NHANES population ($\chi^2 = 124.96$, $p < 0.001$). The analysed sample also included a significantly disproportionate number of participants with one or more self-reported co-morbidities (2%) compared to expected value in the NHANES population ($\chi^2 = 6.38$, $p = 0.046$).

No significant change in results was observed during the sensitivity analysis that weighted the regression to account for these sample differences.

Men

For men, time spent being sedentary and time spent in LIPA were not significantly associated with BMD at the femur and sub-regions (Table 2 Models 1 and 2 for total femur, femoral neck, trochanter and inter trochanter BMD). Time spent in MVPA and time spent in VIG were associated positively with total femur BMD. The regression coefficients were $B_{MVPA} = 0.306$ (95% CI: 0.021–0.591) and $B_{VIG} = 0.320$ (95% CI: 0.058–0.583). For MVPA this corresponds to 0.306 g/cm^2 higher total femur BMD for 1% more time spent per day (this is equivalent on average to 10 min per day). As VIG was transformed, a similar interpretation of findings is not feasible. These associations were not consistently found to be significant for all sub-regions (Table 2, Models 3 and 4 for the femoral neck, trochanter and inter-trochanter BMD). Time spent in MVPA and time spent in VIG were associated with higher BMD in the inter-trochanteric area. These associations have coefficients $B_{MVPA} = 0.358$ (95% CI: 0.023–0.693) and $B_{VIG} = 0.335$ (95% CI: 0.030–0.640). Only time spent in VIG was significantly associated with a higher BMD in the femoral neck area. The regression coefficient for this association was $B_{VIG} = 0.263$ (95% CI: 0.007–0.519). When the models were controlled for the amount of time spent in SB (Models 5 and 6), the associations between total femur BMD (and all sub-regions) and both MVPA and VIG, remained unchanged. The regression coefficients were not attenuated or changed significantly, so the association between BMD and both MVPA and VIG seems to be independent of the amount of time spent being sedentary.

In Models 5 and 6, for the total femur, based on standardised coefficients, BMI had the strongest effect ($\beta = 0.375$), followed by age ($\beta = -0.283$), serum parathyroid hormone levels ($\beta = -0.073$) and smoking ($\beta = -0.055$). In these models, the effect size of MVPA was $\beta = 0.064$ (Model 5) and VIG $\beta = 0.067$ (Model 6). The effect size observed for covariates, MVPA and VIG and their relative magnitude, did not differ noticeably in Models 5 and 6 for the femoral sub-regions.

No significant associations were detected between the spine BMD and SB and PA levels. In Model 5 for spinal BMD (Table 2), which was based on standardised coefficients, BMI had the strongest effect, followed by smoking, alcohol and age. In Model 5, the standardised coefficients were $\beta_{BMI} = 0.190$, $\beta_{smoking} = -0.096$, $\beta_{alcohol} = 0.040$

Table 2
Multivariate association between femur and spine BMD, total sedentary time and total time spent at different physical activity intensities (light intensity LIPA, moderate to vigorous MVPA, vigorous VIG) in men.

Model		Model R^2_{adj} R^2 change	B (95% CI)
	Total femur BMD[a]	0.250	
1	Total sedentary time	0.000	−0.028 (−0.092–0.036)
2	Total LIPA time	0.000	0.012 (−0.057–0.082)
3	Total MVPA time	0.003	0.306 (0.021–0.591)*
4	Total VIG time[b]	0.002	0.320 (0.058–0.582)*
5	Total sedentary time + Total MVPA time	0.003	0.005 (−0.067–0.077) 0.316 (0.005–0.638)*
6	Total sedentary time + Total VIG time[b]	0.003	0.028 (−0.092–0.036) 0.320 (0.057–0.582)*
	Femoral neck BMD[a]	0.285	
1	Total sedentary time	0.000	−0.013 (−0.088–0.037)
2	Total LIPA time	0.000	0.019 (−0.049–0.087)
3	Total MVPA time	0.001	0.154 (−0.128–0.435)
4	Total VIG time[b]	0.002	0.263 (0.007–0.519)*
5	Total sedentary time + Total MVPA time	0.001	−0.013 (−0.088–0.058) 0.128 (−0.189–0.444)
6	Total sedentary time + Total VIG time[b]	0.003	0.025 (−0.088–0.037) 0.263 (0.007–0.519)*
	Trochanter BMD[a]	0.150	
1	Total sedentary time	0.000	−0.026 (−0.084–0.033)
2	Total LIPA time	0.000	0.012 (−0.046–0.079)
3	Total MVPA time	0.002	0.212 (−0.047–0.472)
4	Total VIG time[b]	0.002	0.202 (−0.035–0.439)
5	Total sedentary time + Total MVPA time	0.002	−0.005 (−0.071–0.060) 0.201 (−0.092–0.494)
6	Total sedentary time + Total VIG time[b]	0.003	−0.026 (−0.084–0.032) 0.202 (0.035–0.439)*
	Inter trochanter BMD[a]	0.256	
1	Total sedentary time	0.000	−0.046 (−0.120–0.029)
2	Total LIPA time	0.000	0.030 (−0.050–0.111)
3	Total MVPA time	0.002	0.358 (0.023–0.693)*
4	Total VIG time	0.002	0.335 (0.030–0.640)*
5	Total sedentary time + Total MVPA time	0.001	−0.012 (−0.096–0.072) 0.334 (−0.044–0.711)*
6	Total sedentary time + Total VIG time[b]	0.002	−0.046 (−0.120–0.029) 0.358 (0.023–0.693)*
	Total spine BMD[c]	0.092	
1	Total sedentary time	0.000	0.034 (−0.045–0.112)
2	Total LIPA time	0.000	−0.030 (−0.115–0.056)
3	Total MVPA time	0.000	−0.180 (−0.519–0.158)
4	Total VIG time[b]	0.000	0.110 (−0.205–0.425)
5	Total sedentary time + Total MVPA time	0.000	0.018 (−0.071–0.107) −0.144 (0.528–0.240)
6	Total sedentary time + Total VIG time[b]	0.000	0.033 (−0.045–0.112) 0.108 (−0.207–0.423)

[a] Model adjusted for age, body mass index, ethnicity, parathyroid hormones and smoking.
[b] Square root transformed.
[c] Model adjusted for age, body mass index, ethnicity, alcohol and smoking.
* Statistically significant association with BMD ($p < 0.05$).

Table 3
Multivariate association between femur and spine BMD, total sedentary time and total time spent at different physical activity intensities (light intensity LIPA, moderate to vigorous MVPA, vigorous VIG) in women.

Model		Model R^2_{adj} R^2 change	B (95% CI)
	Total femur BMD[a]	0.376	
1	Total sedentary time	0.012	−0.159 (−0.241–0.076)***
2	Total LIPA time	0.012	0.168 (0.080–0.255)***
3	Total MVPA time	0.001	0.318 (−0.158–0.793)
4	Total VIG time[b]	0.000	0.120 (−0.205–0.445)
5	Total sedentary time + Total MVPA time	0.012	−0.165 (−0.256–0.074)*** −0.085 (−0.607–0.436)
6	Total sedentary time + Total VIG time[b]	0.012	−0.158 (−0.240–0.075)*** 0.097 (−0.229–0.867)
	Femoral neck BMD[a]	0.402	
1	Total sedentary time	0.004	−0.086 (−0.160–0.005)*
2	Total LIPA time	0.004	0.095 (0.013–0.178)*
3	Total MVPA time	0.000	−0.074 (−0.519–0.370)
4	Total VIG time[b]	0.000	0.014 (−0.290–0.318)
5	Total sedentary time + Total MVPA time	0.005	−0.108 (−0.193–0.022)* −0.337 (−0.827–0.153)
6	Total sedentary time + Total VIG time[b]	0.004	−0.082 (−0.160–0.005) * 0.000 (0.303–0.303)
	Trocanter BMD[a]	0.279	
1	Total sedentary time	0.014	−0.137 (−0.209–0.064)***
2	Total LIPA time	0.014	0.141 (0.064–0.218)***
3	Total MVPA time	0.000	0.373 (−0.044–0.790)
4	Total VIG time[b]	0.000	0.115 (−0.171–0.869)
5	Total sedentary time + Total MVPA time	0.015	−0.133 (−0.213–0.053)** −0.048 (−0.411–0.506)
6	Total sedentary time + Total VIG time[b]	0.015	−0.136 (−0.208–0.063) *** 0.091 (−0.192–0.375)
	Inter trocanter BMD[a]	0.357	
1	Total sedentary time	0.015	−0.205 (−0.304–0.106)***
2	Total LIPA time	0.014	0.216 (0.111–0.321)***
3	Total MVPA time	0.000	0.436 (−0.138–1.00)
4	Total VIG time[b]	0.000	0.143 (−0.248–0.533)
5	Total sedentary time + Total MVPA time	0.014	−0.211 (−0.321–0.102)*** −0.084 (−0.710–0.542)
6	Total sedentary time + Total VIG time[b]	0.014	−0.204 (−0.303–0.105)*** 0.107 (−0.280–0.494)
	Total spine BMD[c]	0.092	
1	Total sedentary time	0.001	−0.021 (−0.111–0.068)
2	Total LIPA time	0.001	0.022 (−0.073–0.117)
3	Total MVPA time	0.001	0.117 (−0.421–0.656)
4	Total VIG time[b]	0.001	−0.188 (−0.564–0.187)
5	Total sedentary time + Total MVPA time	0.001	−0.016 (−0.115–0.083) −0.076 (−0.521–0.673)
6	Total sedentary time + Total VIG time[b]	0.002	−0.024 (−0.114–0.066) −0.194 (−0.570–0.183)

[a] Model adjusted for age, body mass index, ethnicity, parathyroid hormones and smoking.
[b] Square root transformed.
[c] Model adjusted for age, body mass index, ethnicity, prednisone use and smoking.
* Statistically significant association with BMD ($p < 0.05$).
** Statistically significant association with BMD ($p < 0.01$).
*** Statistically significant association with BMD ($p < 0.001$).

and $\beta_{age} = -0.031$. These did not differ noticeably and their relative importance remained the same in the other models for spinal BMD.

Women

In women, time spent in SB was negatively associated with total femur BMD and all its sub-regions (Models 1 Table 3). For the total femur the regression coefficient was $B_{SB} = -0.159$ (95% CI: −0.241–0.076), which can be interpreted as a lower BMD by 0.159 g/cm^2 for 1% more of the day spent in SB (this is equivalent to around 10 min more of daily SB time). The regression coefficients for the sub-regions were; $B_{SB} = -0.086$ (95% CI: −0.160–0.005) for the neck of femur BMD, $B_{SB} = -0.137$ (95% CI: −0.209–0.064) for the trochanter BMD and $B_{SB} = -0.205$ (95% CI: −0.306–0.106) for the inter-trochanter BMD. These can be interpreted in the same manner. These associations were not attenuated when the models were controlled for the amount of time spent in MVPA (Model 5 Table 3) and VIG time (Model 6 Table 3). This suggests that the association between SB and total femur BMD (and all sub-regions) is independent of the amount of MVPA and VIG activity for women. In addition, results of the Models 7–8 in Table 4 show that the longer bouts of SB are deleteriously associated with total femur BMD (and all sub-regions) but the number of bouts of SB does not seem to have a significant effect. The association between sedentary bout length and BMD is, however, not independent of the total time spent being sedentary. In Models 5 and 6, which have been corrected for MVPA and VIG, the computed standardised coefficients show that the strongest effect on total femur BMD (and all sub-regions) is from BMI ($\beta = 0.404$), followed by age ($\beta = 0.396$), serum parathyroid hormones ($\beta = -0.133$) and smoking ($\beta = -0.087$). In these models, the effect size for SB was $\beta = -0.124$. For Models 5 and 6 of the sub-regions,

Table 4
Multivariate association between femur bone mineral density (BMD) and pattern of sedentary time in women.

Model[a]	Total femur BMD		Femoral neck		Trochanter		Inter-trochanter	
	R^2_{adj}	B (95% CI)	R^2_{adj}	B (95% CI)	R^2_{adj}	B (95% CI)	R^2_{adj}	B (95% CI)
Bouts[b]	0.398	0.000 (0.000–0.001)	0.405	0.000 (0.000–0.001)	0.308	0.000 (0.000–0.001)	0.379	0.000 (−0.001–0.001)
Bout duration (min)	0.395	−0.009*** (−0.015–0.004)	0.404	−0.006*** (−0.011–0.001)	0.312	−0.008*** (−0.012–0.004)	0.375	−0.012*** (−0.018–0.006)

[a] Model adjusted for total age, body mass index, ethnicity, parathyroid hormones, and smoking.
[b] Model also adjusted for wear-time.
*** Statistically significant association with BMD ($p < 0.001$).

the strength of effect observed for the covariates and SB did not change notably.

No significant associations were observed between time in MVPA and VIG and total femur BMD and all sub-regions (Models 3 and 4, Table 3). However the time spent in LIPA was found to be positively associated with total femur BMD and BMD of all sub-regions (Model 2, Table 3), with coefficients $B_{LIPA} = 0.168$ (95% CI: 0.080–0.255) for total femur BMD, $B_{LIPA} = 0.095$ (95% CI: 0.013–0.178) for the femoral neck BMD, $B_{LIPA} = 0.141$ (95% CI: 0.064–0.218) for the trochanter and $B_{LIPA} = 0.216$ (95% CI: 0.111–0.321) for the inter-trochanter BMD. These coefficients can be interpreted as higher BMD in g/cm^2 for 1% more of the day in LIPA (this corresponds roughly to 10 min). Due to the collinearity between LIPA and SB time we could not test whether both the effects of LIPA and SB are independent, but their effects are of comparable size and in opposite direction.

For the spinal BMD, no associations were found for any of the PA intensity levels or for SB. In Model 5 for spinal BMD (Table 2), based on standardised coefficients, age ($\beta_{age} = -0.350$) had the strongest effect, followed by BMI ($\beta_{BMI} = 0.190$), use of prednisone ($\beta_{prednisone} = -0.079$) and smoking ($\beta_{smoking} = -0.040$). These did not differ noticeably and their relative importance remained the same in the other models for spinal BMD.

Discussion

In this cross-sectional study the amount of time spent engaging in SB is negatively associated with BMD of the femur in women, independently of the amount of time they spent doing moderate and vigorous activity. This effect was not seen in men.

This appears to corroborate reports of an increased risk of osteoporotic fractures that are associated with higher self-reported sitting times in older women [29] and a negative association between SB and femur BMD found in female adolescents [27,28]. Together, these associations suggest that SB might be a modifiable risk factor for bone health in women, which warrants further research. In addition, even if the strength of the association between SB on total femur BMD in women appears small ($\beta = -0.124$) compared to that of age ($\beta = -0.396$) and BMI ($\beta = -0.404$) which are known to have strong influence on BMD, it is not negligible and in fact appears comparable to that of smoking ($\beta = -0.087$), a known risk factor for bone health. The results of this study also suggest that the association between BMD of the total femur and all sub-regions and SB in women is independent of their level of moderate and vigorous activity as found in female adolescents [27]. This means that even at equal levels of engagement in MVPA, women who are sedentary for a longer time during the rest of the day might still have a lower BMD of the femur. Considering that it is possible for an individual to meet recommended guidelines for PA and still spend a large proportion of the day in SB [21], there might be benefits in promoting a reduction in SB as a lifestyle intervention for improving bone health in women. Future research should therefore consider investigating the effectiveness of combined SB reduction and PA engagement interventions. Interestingly, time spent in LIPA seem to have the opposite effect to SB suggesting that PA interventions could include light daily activities.

In women, there was no significant association between the frequency of SB and total femur BMD (and all sub-regions) but bout duration was found to be negatively associated with total femur BMD (and all sub-regions). This suggests that the manner in which SB time is accumulated could affect BMD and if this is the case, reducing the length of SB periods could have a beneficial effect on BMD at the hip. Thus future research, rather than focussing on a reduction in number of bouts, should instead promote a reduction in long bouts.

The fact that no association between sedentary time and spinal BMD was found, also suggests that the effects of SB on femur BMD is due to the seated posture rather than a lack of physical activity. Indeed when seated the quadriceps and hamstring muscles are rarely activated but the load on the lumbar bone and muscle structures is increased [38]. Therefore it is possible that the relationship between SB and hip BMD is due to the lack of oesteogenic tension and loading on the femoral bone when sitting [39]. The measurement of spine BMD is often compromised in older age due to degenerative changes in the spine. Hence it is also possible that measurement errors in spine BMD are responsible for the lack of association found between SB and spinal BMD.

However, the effect of SB on BMD seen in women was not found in men. Instead, men's BMD was strongly associated with levels of MVPA and VIG. One possible explanation for this is the difference in activity profiles between men and women. High levels of SB in older men appear to be associated with both higher muscle mass and higher levels of vigorous activity [40]. It is also possible that SB has specific effects on the hormonal system, in similar ways to those identified in weightlessness and bed rest studies [41], which could affect women more strongly. However, the notable differences in results for the femoral and spinal regions strongly suggest that the effect of PA and SB on BMD is localised, mechanical and not systemic, just as the effect of exercise on BMD is localised and not systemic.

The association between physical activity and BMD are comparable and consistent with previous cross-sectional, prospective and longitudinal studies [42–45]. The effect sizes for physical activity reported here seem high. The regression coefficient describing the relationship between total femur BMD and MVPA in men after correction for covariates indicates that a change of around 10 min of daily MVPA would correspond to a 30% change in BMD (0.30 g/cm^2). This seems unrealistic, however it is a more conservative figure than has previously been derived using self-reported measure of PA. Langsetmo et al. [42] reported a change of 0.022 g/cm^2 for the difference of 1 MET·min/day. This is approximately one order of magnitude lower than the results presented here, but in Langsetmo et al. the unit of physical activity is MET·min/day which is a very small change in physical activity. If the results of Langsetmo et al. are interpreted for the same 10 minute difference in MVPA/day, then the corresponding change in femoral BMD would be roughly 0.88 g/cm^2. Therefore the results of this study, using objective monitoring of PA, seem to correct some overestimation obtained

from self-reported data. There is a dearth of other studies using objectively measured PA, especially in men, that would allow further comparisons.

The effect sizes reported here should be interpreted in light of the unit of measurement of PA and the non-linear nature of the dose response curve between health and PA, especially at low levels of PA, such as in this sample. First, the unit of change of PA is roughly 10 min of daily MVPA. This is a large amount of MVPA, rarely achieved sustainably in physical activity interventions. It corresponds roughly to 70 min per week of MVPA or almost half of the recommended guidelines for PA [46]. Second, although the shape of the dose–response between BMD and PA is not precisely known, it is likely to be non-linear as for other physical performance impacts of PA [46]. It is likely that a greater magnitude of change can occur from a moderate increase in activity starting from a low baseline than starting from a higher level of baseline physical activity. Considering that in the NHANES sample the levels of activity are low with most subjects not meeting the recommended guidelines [47] it is likely that the associations reported here correspond to the steepest part of the dose–response curve leading to seemingly large effect size. These effect sizes are likely to decrease for people who are routinely more active. In addition it is unlikely that each additional change of 10 min of MVPA will lead to the same corresponding change in BMD.

This study, which used objective measures of PA and sedentary time, confirms previous findings, based on self report, that both moderate and vigorous activities have a protective effect on men's BMD. However, this does not seem to be the case for women. These results are intriguing and counterintuitive because activities such as walking, which should be recorded accurately by accelerometry, have known oesteogenic effects in women [9]. However, there are consistent reports using both objective and subjectively measured physical activity in women that concur with the results of this study [42,43,45]. While the total volume of physical activity appears to be positively associated with BMD, the time spent specifically doing moderate and vigorous activity does not. In this study, LIPA constitutes the largest proportion of the time women spent being active. A recent study showed that there is no association between activity intensity and BMD in women [44]. This is confirmed by experimental studies which showed that very infrequent high impact activity (such as 30 jumps per week) has more effect on BMD than continuous moderate or vigorous activity [48]. Another, but related, explanation is related to the fact that bouts of moderate and vigorous activity are very short. Most accelerometry results are reported on a 1 minute epoch basis, therefore it is possible that some epochs containing short bouts of activity are misclassified by the calibration equations resulting in an underestimate of the total time spent in moderate and vigorous activity. Future research using accelerometers should therefore use shorter epochs and try to identify high impact events more effectively.

The strengths of this study are the use of objective measures of SB and PA, a robust sample size and the quality of the dataset, in particular the quality control and calibration of all BMD and physical activity measures.

The main limitations of this study are those of all cross-sectional studies. There may be cohort effects not considered and other changes to lifestyle and health may have impacted on accrual of bone mass other than the factors considered in this analysis. In particular, care should be taken in generalising the results as we found statistically significant differences in age, gender and ethnic distribution and self-reported health for the sample analysed compared to the population of the NHANES 2005–6 cycle. Selection bias occurred due to adherence to accelerometry. Fewer participants (7%) with poor self reported health returned valid accelerometry data. Although the sensitivity analysis indicates that this did not seem to affect the results, there is the potential that the results might be different in populations with worse self reported health. In addition, sedentary time may have been over estimated, as it was not inferred from a sensor that measures the sitting posture [24] but from an activity count threshold that would have included some periods of quiet standing.

Conclusion

This study reports first evidence that objectively measured sedentary time is negatively associated with bone mineral density of the femur region in women. This association appears independent of the level of PA and is not attenuated by the engagement in moderate and vigorous activity during the rest of the day. There is also an association between LIPA and BMD. Collinearity between LIPA and SB time does not allow for checks for independence between the two, however, the strength of this positive effect is comparable to the negative effect seen for total SB time.

This suggests that SB might be a modifiable risk factor for women's bone health, independent from the lack of engagement in PA and warrants further exploration. Research should consider investigating the effect on bone health of lifestyle interventions specifically aimed at reducing SB (particularly long duration bouts of sitting) and increasing LIPA. In particular, the effect of reducing the duration of long sedentary bouts should be researched, as the results of this study suggest that repeated exposure to long continuous periods of SB might be deleterious to bone health.

This association between SB and BMD was not found in men. Instead this study replicates data from self reported activity, showing that MVPA and VIG activity have the most positive effects on BMD in men. Interventions should therefore concentrate on increasing MVPA and VIG in men for improved bone health.

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