Sex Specific Association of Physical Activity on Proximal Femur BMD in 9 to 10 Year-Old Children

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Abstract

The results of physical activity (PA) intervention studies suggest that adaptation to mechanical loading at the femoral neck (FN) is weaker in girls than in boys. Less is known about gender differences associated with non-targeted PA levels at the FN or other clinically relevant regions of the proximal femur. Understanding sex-specific relationships between proximal femur sensitivity and mechanical loading during non-targeted PA is critical to planning appropriate public health interventions. We examined sex-specific associations between non-target PA and bone mineral density (BMD) of three sub-regions of the proximal femur in pre- and early-pubertal boys and girls. BMD at the FN, trochanter (TR) and intertrochanter (IT) regions, and lean mass of the whole body were assessed using dual-energy x-ray absorptiometry in 161 girls (age: 9.7±0.3 yrs) and 164 boys (age: 9.7±0.3 yrs). PA was measured using accelerometry. Multiple linear regression analyses (adjusted for body height, total lean mass and pubertal status) revealed that vigorous PA explained 3–5% of the variability in BMD at all three sub-regions in girls. In boys, vigorous PA explained 4% of the variability in IT BMD and 6% in TR BMD. PA did not contribute to the variance in FN BMD in girls. An additional 10 minutes per day of vigorous PA would be expected to result in a ~1% higher FN, TR, and IT BMD in boys (p<0.05) and a ~2% higher IT and TR BMD in girls. In conclusion, vigorous PA can be expected to contribute positively to bone health outcomes for boys and girls. However, the association of vigorous PA to sub-regions of the proximal femur varies by sex, such that girls associations are heterogeneous and the lowest at the FN, but stronger at the TR and IT, when compared to boys.

Introduction

Mechanical loading by impact or muscle forces is a contributing factor to skeletal health throughout the life course; however, mechanical loading is particularly important during the transition period from childhood to adolescence. This may be due to the efficient response of bone to loading during middle childhood (elementary school years), since the magnitude of bone accrual associated with mechanical loading is reported to be greater when compared to early childhood and adulthood [1–6]. In addition, the evidence shows that the amount and intensity of PA levels are highest during middle childhood compared to other time points across childhood and adolescence [7]. The amount and intensity of PA during middle childhood is important since PA levels dramatically decrease during adolescence [7–9]. Consequently, a timely intervention in children’s activity habits when bone appears to be most responsive to activity’s effect and PA is more easily accepted could be an important public health strategy for optimal bone development.

Studies conducted with pre- and early-pubertal children have shown augmented bone mineral accrual in several skeletal sites in both girls [10–17] and boys [15–17] after high-impact mechanical loading (running and jumping) interventions. The effect of high-impact loading exercise on bone has been shown to be site specific, with accrual occurring at weight bearing sites such as lumbar spine [11] and proximal femur [18–22]. The loads imposed during targeted exercise are likely to represent the “best case scenario” and might not generalize to the spontaneous PA choices of children.

Population-based observational studies provide important information on the relationship of bone accrual to the type and amount of PA children voluntarily choose to do. Twenty-five to forty minutes of vigorous PA per day has been suggested as a minimum daily dose for optimal bone growth [23–26] but the relationship between bone mineral accrual and PA during the growing years has not been thoroughly examined. Of the studies that exist, there is a lack of consensus on whether sex moderates the association between mechanical loading and bone accrual at the proximal femur [18–22]. However, it has been suggested that the proximal femur of boys is more sensitive to mechanical loading than girls’ [27]. This idea was not derived from work specifically powered to examine sex differences [23,27–31]. Furthermore, most studies reporting a lower responsiveness of PA in girls (when compared to boys) analysed only the femoral neck and not other sub-regions of the proximal femur (i.e. trochanter and intertrochanter).

A potential explanation for sex-specific bone response at the proximal femur is the lower intensity mechanical loading on the
Methods

Sample

This cross-sectional cohort study included 325 pre and early pubertal subjects (Tanner stage 1 and 2), aged 9–10 years (164 boys and 161 girls) living in the island of Madeira (Portugal) and drawn from the European Youth Heart Study (EYHS). Selection procedures and methods are described in detail elsewhere [24]. None of the subjects were taking any medication affecting bone and none reported a history of bone fracture in lower limbs. The research protocol was in accordance with the Helsinki Declaration. Parents or legal guardians provided written informed consent and the study was approved by the Ethics Committee attached to the scientific board of the Faculty of Human Movement.

Physical Activity. PA was assessed with a uniaxial accelerometer (model WAM 6471, Manufacturing Technology Incorporated, Fort Walton Beach, FL), over two weekdays and two weekend days. The subjects were asked to wear the accelerometer all day except during water activities, in a representative week of their normal activity, and the procedure was repeated in all cases in which any abnormal event was reported. MAHULife software (www.mrc-epid.cam.ac.uk) was used to analyze and process activity data. Outcome variables were time (minute/day) spent in light, moderate and vigorous intensity of PA. The intensity of PA was defined according the counts per minute (cpm) as follows: light intensity from 100 to 1999 cpm; moderate intensity from 2000 to 2999 cpm; and vigorous intensity over 2999 cpm. All of the activity data were averaged over the 4-day period and subjects who failed to provide a minimum of 3 days of ≥600 minutes of accelerometer data were excluded. PA procedures are detailed in previous report [41].

Clinic Measures

Standing height (to the nearest millimetre) was measured on a stadiometer (Secca 770, Hamburg, Germany) without shoes. Body weight (kilograms), total fat (kilograms), and lean mass without bone (kilograms) were determined from a total body scan by dual x-ray absorptiometry (DXA) (QDR-1500, high-speed performance mode, software 5.7) (Hologic, Waltham, MA; pencil beam, software 5.73). Sexual maturity was assessed using self-report and Tanner’s 5-stage scale for breast development in girls and pubic hair in boys. Children were stratified as prepubertal (Tanner stage 1) or having started puberty (Tanner stage 2) [42].

BMD from three proximal femur sub-regions, i.e., femoral neck, trochanter and intertrochanter, were measured with DXA (QDR-1500, high-speed performance mode, software 4.76). Quality

| Table 1. Characteristics of participants as mean±standard deviation. |
|-------------------------|-------------------------|-------------------------|
|                        | Girls (n = 161)         | Boys (n = 164)          | p*         |
| Age, y                  | 9.7±0.3                | 9.7±0.3                | 0.780      |
| Tanner Stage (1/2), %   | 40/60                  | 4/96                   | <0.001     |
| Body Weight, kg         | 34.2±9.0               | 34.1±7.8               | 0.960a,b   |
| Body Height, cm         | 137.2±0.1              | 137.0±0.1              | 0.813      |
| Body Fat, kg            | 10.2±5.7               | 8.2±5.7                | 0.002a     |
| Body Lean Mass, kg      | 23.1±3.2               | 25.1±2.9               | <0.001     |
| Calcium Intake, mg/d    | 1020±424               | 1048±407               | 0.553      |
| Light PA, min/d         | 296±47                 | 278±49                 | 0.001      |
| Moderate PA, min/d      | 142±47                 | 169±55                 | <0.001     |
| Vigorous PA, min/d      | 18±14.3                | 30±21                  | <0.001a,b  |
| Moderate and Vigorous PA, min/d | 159±56           | 198±70                 | <0.001     |
| Total PA, min/d         | 456±77                 | 476±90                 | 0.030      |
| PA Average Intensity, count/min/d | 586±189  | 732±273               | <0.001     |
| Proximal Femur BMD, g/cm² | 0.690±0.07            | 0.753±0.08             | <0.001     |
| Femoral Neck BMD, g/cm² | 0.656±0.06            | 0.722±0.07             | <0.001a,b  |
| Trochanter BMD, g/cm²   | 0.544±0.06            | 0.591±0.07             | <0.001a,b  |
| Intertrochanter BMD, g/cm² | 0.762±0.08         | 0.820±0.09             | <0.001a,b  |
| Femoral Neck BMD/Trochanter BMD | 1.21±0.08    | 1.22±0.08              | 0.095      |
| Femoral Neck BMD/Intertrochanter BMD | 0.86±0.05    | 0.88±0.05              | 0.001a     |
| Trochanter BMD/Intertrochanter BMD | 0.72±0.05    | 0.72±0.05              | 0.678      |

*Student’s t-test comparing boys to girls was performed when both variables have normal distribution with the same variance. In cases of no normality or no homogeneity of variances, Mann-Whitney nonparametric test was used. aGirl’s variable without normal distribution; bBoy’s variable without normal distribution. PA - physical activity BMD – bone mineral density.

doi:10.1371/journal.pone.0050657.t001
Table 2. Standardized regression coefficients (β), level of significance (p) and coefficient of determination (R²) for proximal femur sub-region models, adjusted for sex, Tanner stage, body height and body lean mass, with data for boys and girls pooled together.

| Predictor Variables | β     | p         | R²     |
|---------------------|-------|-----------|--------|
| FN BMD              | BLM   | 0.308     | <0.001 | 0.204  |
| FN BMD              | Sex   | 0.239     | <0.001 | 0.100  |
| Body Height         | -0.064| 0.311     | -      |
| Tanner Stage        | 0.110 | 0.038     | 0.009  |
| Vigorous PA         | 0.191 | <0.001    | 0.033  |
| MVPA                | -0.004| 0.955     | -      |
| Moderate PA         | -0.003| 0.955     | -      |
| TR BMD              | BLM   | 0.372     | <0.001 | 0.209  |
| Sex                 | 0.170 | 0.001     | 0.024  |
| Body Height         | -0.096| 0.179     | -      |
| Tanner Stage        | 0.077 | 0.156     | -      |
| Vigorous PA         | 0.227 | 0.001     | 0.073  |
| MVPA                | 0.039 | 0.604     | -      |
| Moderate PA         | 0.031 | 0.604     | -      |
| IT BMD              | BLM   | 0.426     | <0.001 | 0.242  |
| Sex                 | 0.129 | 0.012     | 0.014  |
| Body Height         | 0.030 | 0.634     | -      |
| Tanner Stage        | 0.082 | 0.030     | -      |
| Vigorous PA         | 0.182 | <0.001    | 0.046  |
| MVPA                | 0.019 | 0.798     | -      |
| Moderate PA         | 0.015 | 0.798     | -      |

BMD – bone mineral density; FN BMD - femoral neck BMD; TR BMD - trochanter BMD; IT BMD - intertrochanter BMD; PA - physical activity; MVPA – moderate-through-vigorous PA; BLM – body lean mass.
doi:10.1371/journal.pone.0050657.t002

assurance tests were performed each morning. Precision errors were estimated from 2 measurements in 14 subjects [43]. The coefficients of variation of femoral neck, trochanter and intertrochanter BMD ranged from 1.2% to 1.5%. From the DXA coefficients of variation of femoral neck, trochanter and intertrochanter BMD variance; a 3% variation in the FNTR and variable explaining 6% of the trochanter BMD and 4% of the FNIT, with boys revealing a higher ratio than girls.

Results

The characteristics of the children are presented in Table 1. There were no differences in age, body weight and height between boys and girls. However, lean mass was higher and fat mass was lower in boys, who were also more active than girls. Boys spent more time in moderate and vigorous PA than girls, whereas girls spent more time in light activities. The proportion of participants in early puberty (Tanner 2) was higher in boys than in girls (40% of the girls and 4% of the boys were in the Tanner stage 1). The BMD of the proximal femur and of its three sub-regions was higher in boys than in girls, but statistically significant sex differences in BMDs ratios were not found, with the exception of the FNIT, with boys revealing a higher ratio than girls.

Associations between PA and BMD of the proximal femur sub-regions were analysed using multiple regression models, first with boys and girls pooled together (Table 2) and after separated (Table 3). Among PA variables (time spent at moderate, MVPA, and vigorous PA), vigorous PA was the one with the highest contribution to the R squared of each model (3–7%, p<0.001) (Table 2). None of the other two PA variables showed additional explanatory power once vigorous PA had entered the model. In the same table, body lean mass explained 20–24% of variance in all BMD models (p<0.001) while Tanner stage was responsible for ~1% variability of femoral neck BMD (p = 0.038). In all the three regression models ran with boys’ and girls’ data pooled together (Table 2), sex turned out to be a significant predictor variable, giving empirical ground for subsequent separated data treatment.

Three other similar models were run for the proximal femur BMD ratios with boys and girls together but none of them complied with the assumptions for regression analysis, having therefore been rejected. Conversely, the models of proximal femur BMD ratios were added to the initial three ones when data was considered separately for boys and girls to analyse associations between BMD and PA variables adjusted for Tanner stage, lean body mass and body height (Table 3). Among all the PA intensity variables examined, vigorous PA was the best predictor: it explained ~3–3% of the BMD variance (p<0.05) in boy’s femoral neck, trochanter and intertrochanter. However, none of the variation of BMD ratios in boys was predicted by PA intensity variables. In girls, vigorous PA was also the best PA predictor variable explaining 6% of the trochanter BMD and 4% of the intertrochanter BMD variance; a 3% variation in the FNTR and...
FNIT was also associated with vigorous and MVPA, respectively. In girls, with exception of femoral neck BMD, PA (vigorous and MVPA) explained 3–6% of all BMD variances. Unlike boys, in girls there was a negative association between PA variables and FNTR and FNIT ($p < 0.05$).

Table 3 also shows that body lean mass was a significant predictor variable in all girls and boy’s models for the proximal femur’s regional BMDs, except in the girls TRIT BMD model.

Table 4 presents regression models using the three PA intensity variables mostly widely used in the literature, moderate PA, vigorous PA and MVPA. In our analysis, there was a higher absolute effect (estimated by unstandardized regression coefficients) of one minute per day of vigorous PA on the BMD than one minute per day of MVPA or moderate PA (only in girls). The effect of PA was not homogeneous for all proximal femur sub-regions and was dissimilar between boys and girls. For example, in girls the hypothetical BMD increase associated with an additional

| Predictor Variables | Girls | Boys |
|---------------------|-------|------|
| FN BMD              |       |      |
| BLM                 | 0.483 | 0.277|
| Body Height         | -0.075| -0.047|
| Tanner Stage        | 0.100 | 0.100|
| Vigorous PA         | 0.135 | 0.225|
| MVPA                | 0.125 | -0.092|
| Moderate PA         | 0.109 | -0.073|
| TR BMD              |       |      |
| BLM                 | 0.511 | 0.238|
| Body Height         | -0.139| -0.085|
| Tanner Stage        | 0.062 | -0.007|
| Vigorous PA         | 0.241 | 0.214|
| MVPA                | 0.076 | -0.016|
| Moderate PA         | 0.064 | -0.013|
| IT BMD              |       |      |
| BLM                 | 0.514 | 0.324|
| Body Height         | -0.017| 0.061|
| Tanner Stage        | 0.134 | -0.058|
| Vigorous PA         | 0.213 | 0.159|
| MVPA                | 0.102 | -0.063|
| Moderate PA         | 0.086 | -0.081|
| FNTR BMD            |       |      |
| BLM                 | -0.178| 0.027|
| Body Height         | 0.079 | 0.061|
| Tanner Stage        | 0.034 | 0.168|
| Vigorous PA         | -0.172| -0.033|
| MVPA                | -0.038| -0.072|
| Moderate PA         | -0.032| -0.078|
| FNIT BMD            |       |      |
| BLM                 | -0.250| -0.009|
| Body Height         | -0.119| -0.208|
| Tanner Stage        | 0.093 | 0.260|
| Vigorous PA         | -0.095| 0.057|
| MVPA                | 0.184 | 0.041|
| Moderate PA         | 0.313 | 0.029|
| TRIT BMD            |       |      |
| BLM                 | 0.029 | 0.029|
| Body Height         | 0.223 | 0.029|
| Tanner Stage        | 0.076 | 0.068|
| Vigorous PA         | 0.082 | 0.082|
| MVPA                | 0.078 | 0.324|

**Table 3.** Standardized regression coefficients ($\beta$), level of significance ($p$) and coefficient of determination ($R^2$) for proximal femur sub-region models, adjusted for Tanner stage, body height, and body lean mass, with data for boys and girls treated separately.

| Predictor Variables | Girls | Boys |
|---------------------|-------|------|
| FN BMD              |       |      |
| BLM                 | 0.483 | 0.277|
| Body Height         | -0.075| -0.047|
| Tanner Stage        | 0.100 | 0.100|
| Vigorous PA         | 0.135 | 0.225|
| MVPA                | 0.125 | -0.092|
| Moderate PA         | 0.109 | -0.073|
| TR BMD              |       |      |
| BLM                 | 0.511 | 0.238|
| Body Height         | -0.139| -0.085|
| Tanner Stage        | 0.062 | -0.007|
| Vigorous PA         | 0.241 | 0.214|
| MVPA                | 0.076 | -0.016|
| Moderate PA         | 0.064 | -0.013|
| IT BMD              |       |      |
| BLM                 | 0.514 | 0.324|
| Body Height         | -0.017| 0.061|
| Tanner Stage        | 0.134 | -0.058|
| Vigorous PA         | 0.213 | 0.159|
| MVPA                | 0.102 | -0.063|
| Moderate PA         | 0.086 | -0.081|
| FNTR BMD            |       |      |
| BLM                 | -0.178| 0.027|
| Body Height         | 0.079 | 0.061|
| Tanner Stage        | 0.034 | 0.168|
| Vigorous PA         | -0.172| -0.033|
| MVPA                | -0.038| -0.072|
| Moderate PA         | -0.032| -0.078|
| FNIT BMD            |       |      |
| BLM                 | -0.250| -0.009|
| Body Height         | -0.119| -0.208|
| Tanner Stage        | 0.093 | 0.260|
| Vigorous PA         | -0.095| 0.057|
| MVPA                | 0.184 | 0.041|
| Moderate PA         | 0.313 | 0.029|
| TRIT BMD            |       |      |
| BLM                 | 0.029 | 0.029|
| Body Height         | 0.223 | 0.029|
| Tanner Stage        | 0.076 | 0.068|
| Vigorous PA         | 0.082 | 0.082|
| MVPA                | 0.078 | 0.324|

PA - physical activity; BMD – bone mineral density; FN BMD - femoral neck BMD; TR BMD - trochanter BMD; IT BMD - intertrochanter BMD; FNTR – BMD ratio of femoral neck for trochanter; FNIT – BMD ratio of femoral neck for intertrochanter; TRIT- BMD ratio of trochanter for intertrochanter; BLM – body lean mass.

doi:10.1371/journal.pone.0050657.t003
10 min/day of PA was comparable (~2%) for trochanter and intertrochanter regions (with no effect on femoral neck). The effect for boys was lower (~1%) but the response was similar among all three sub-regions of proximal femur.

Discussion

PA showed a positive contribution to the BMD variation of the three sub-regions of the proximal femur in boys but in girls PA did not help to explain femoral neck BMD variance. For the same duration of PA, the regression coefficients of more intense PA (vigorous PA) were always higher than those corresponding to a less intense PA (MVPA) in boys and girls. The extrapolation of our results suggest a ~2% higher BMD in the trochanter and intertrochanter regions in girls at the studied age range if an additional 10 minutes per day of vigorous PA is achieved. In boys, the corresponding gain is a ~1% higher femoral neck, trochanter and intertrochanteric BMDs.

The higher regression coefficients for PA of highest intensity – vigorous PA – compared to lower levels of intensity – MVPA or moderate PA – when regional BMDs of femoral neck, trochanter and intertrochanter are in stake underline the relevance of the PA intensity to bone mineral accrual during the studied pediatric years. The PA threshold under which the effects on bone mass could be modest has been proposed [24–26]. Given that boys are usually more active than girls [7–9], this could partially explain BMD differences between sexes at proximal femur sub-regions. However this difference seems not be homogeneous among sub-regions. Our results are consistent with studies that reported a positive response of girls' proximal femur BMD (or bone mineral content - BMC) to PA but also with studies that revealed a response of femoral neck BMD or BMC to PA only in boys. Particularly, our site specific response of girls' proximal femur in the trochanteric region is in line with the Iowa Bone Development Study which reported 5% and 14% more BMC at the total body and trochanteric region in the most active pre- and early pubertal boys and girls, when compared to inactive peers [29]. Similar effects regarding skeletal regions were also found by Stear et al., who reported greater BMC accrual at the trochanter (4.8%) than in the whole body (0.8%) or lumbar spine (1.9%) in 144 adolescent girls enrolled in a 45-min exercise-to-music classes programme, three times per week, after 15.5-month [44]. Witzke et al. reached analogous findings at the trochanter BMC in adolescent girls using a plyometric jump training programme with no significant differences for the femoral neck, spine or whole body BMC [45]. McKay et al. who examined the effect of an 8-month school-based jumping programme in pre and early pubescent girls, found that the intervention group showed a significantly greater change in trochanter BMD than the control group [16,18]. Additionally, increments (4.3%) for femoral neck BMC of 8 to 12 years old boys (compared to controls) were reported after 2 years of a high-impact circuit intervention [19]. These observations contradict the idea that girls proximal femur is not responsive to PA, although, notably none of these studies reported a positive effect of PA on girl's femoral neck.

The positive associations that we found between PA and the BMD of the three proximal femur sub-regions in boys and only at the trochanter and at the intertrochanter region in girls is similar with the results of those studies that suggested no effect of PA in girls' proximal femur, whose analyses were focused in the femoral neck region [6,10,11]. The exception, seems to be the study conducted by Petit et al. [17] that showed significant gains in the BMD at the intertrochanter (1.7%) and at the femoral neck (2.6%) region in early pubertal girls (Tanner stages 2 and 3) when compared to controls after a 10-minute jumping programme, 3 times per week during 7 months.

Table 4. Effects of 10 minutes per day of additional physical activity on femoral neck, trochanter, and intertrochanter BMD, adjusted for Tanner stage, body height, and body lean mass.

|                | Girls               | Boys             |                |                |
|----------------|---------------------|------------------|----------------|----------------|
| β              | FN BMD              | TR BMD           | IT BMD         | FN BMD         | TR BMD           | IT BMD         |
| Moderate PA    | ns                  | 0.00022          | 0.00031        | ns             | ns              | ns             |
|                | (p = 0.008)         | (p = 0.008)      |                |                |                  |                |
| ModVig PA      | 0.00014             | 0.00002          | 0.00030        | 0.00015        | 0.00016          | 0.00013        |
|                | (p = 0.072)         | (p = 0.002)      | (p = 0.002)    | (p = 0.048)    | (p = 0.025)      | (p = 0.186)    |
| Vigorous PA    | 0.00059             | 0.00101          | 0.00127        | 0.00075        | 0.00056          | 0.00066        |
|                | (p = 0.056)         | (p = 0.001)      | (p = 0.001)    | (p = 0.003)    | (p = 0.016)      | (p = 0.033)    |

Δ BMD (%) associated to Δ 10 min/day of physical activity

|                | Girls               | Boys             |                |                |
|----------------|---------------------|------------------|----------------|----------------|
|                | FN BMD              | TR BMD           | IT BMD         | FN BMD         | TR BMD           | IT BMD         |
| Moderate PA    | ns                  | 0.4              | 0.4            | ns             | ns              | ns             |
| ModVig PA      | ns                  | 0.4              | 0.4            | 0.2            | 0.3             | ns             |
| Vigorous PA    | ns                  | 1.9              | 1.7            | 1.0            | 1.1             | 0.8            |

PA – physical activity; BMD – bone mineral density; FN - femoral neck; TR - trochanter; IT – intertrochanter; ns – non-significant regression coefficient.
The analysis of all sub-regions of proximal femur in both sexes was a distinctive aspect of our study that provided a more comprehensive examination of bone’s response to PA. Compared to boys, girls showed inferior BMD in the different sub-regions of the proximal femur, which is not new. However, we observed a lower or a tendency to a lower BMD in the femoral neck relative to other sub-regions (FNIT, girls: 0.86 vs. boys: 0.88, p = 0.001; FNTR, girls: 1.21 vs. boys: 1.22, p = 0.095), i.e. the proximal femur sub-region where we did not find any positive association with MVPA or vigorous PA in girls. Our study showed that the pattern of proximal femur responsiveness to PA was more homogeneous in boys, when compared to girls. Conversely, in girls, there were negative relationships between PA and FNTR and FNIT, suggesting a heterogeneous responsiveness favouring the trochanteric and intertrochanteric sub-regions of the proximal femur. If our response pattern findings were generalizable, it is not surprising that researchers using the neck region to represent the entire proximal femur suggest that boys' femur is more responsive to mechanical loading than girls’ at this age.

In addition to the well-known limitations of DXA technology in the assessment of bone, our study may have an additional limitation due to the self-report of children’s maturity status. The assessment of bone, our study may have an additional limitation due to the self-report of children’s maturity status. In addition, sample sizes should be large enough to allow investigators to test interactions among PA and hip biomechanical factors (as opposed to systemic factors as nutritional, hormonal or sun exposure factors) that can affect differently the BMD of specific regions of proximal femur. Our study shows a region-specific bone response to vigorous PA in pre and early pubertal girls and boys. More active girls have greater BMD in the trochanter and intertrochanter while more active boys have greater BMD in all sub-regions of the proximal femur.

Author Contributions

Conceived and designed the experiments: FB LBS GC. Performed the experiments: RO GC FB. Analyzed the data: GC RO LBS FB KFJ. Contributed reagents/materials/analysis tools: RO LBS FB GC. Wrote the paper: GC FB LBS KFJ.

References

1. Bass SL (2000) The prepubertal years: a uniquely opportune stage of growth when the skeleton is most responsive to exercise? Sports Med 30: 73–84.
2. Khan K, McKay H, Haapasalo H, Bennett K, Forwood M, et al. (2000) Does childhood and adolescence provide a unique opportunity for exercise to strengthen the skeleton? J Sci Med Sports 3: 130–64.
3. Hughes J, Novotny S, Wetzsteon R, Petit M (2007) Lessons learned from school based skeletal loading intervention trials: putting research into practice. Med Sport Sci 51: 137–56.
4. MacKelvie K, Khan K, McKay H (2002) Is there a critical period for bone response to weightbearing exercise in children and adolescents? A systematic review. Br J Sports Med 36: 250–7.
5. Bass S, Saxon L, Daly R, Turner C, Rohling A, et al. (2002) The effect of mechanical loading on the size and shape of bone in pre-, peri-, and postpubertal girls: a study in tennis players. J Bone Miner Res 17: 2274–80.
6. Romann M, Zahner L, Schindler C, Puder J, Kraenzlin M, et al. (2011) Effect of a general school-based physical activity intervention on bone mineral content and density: A cluster-randomized controlled trial. Bone 48: 792–7.
7. Troiano R, Birogari D, Dodd K, Masse L, Tilton T, et al. (2008) Physical activity in the United States measured by accelerometry. Med Sci Sports Exerc 40: 181–8.
8. Baptista F, Santos DA, Silva AM, Mota J, Santos R, et al. (2012) Prevalence of the Portuguese population attaining sufficient physical activity. Med Sci in Sports Exerc 44: 667–73.
9. Nader PR, Bradley RH, Houts RM, McRitchie SI, O’Brien M (2008) Moderate-to-vigorous physical activity from ages 9 to 15 years. JAMA 300: 295–305.
10. Morris F, Naughton G, Gibbs J, Carlson J, Wark J (1997) Prospective ten month exercise intervention in prepuberal girls: positive effects on bone and lead. J Bone Miner Res 12: 1453–62.
11. Heinonen A, Sievanen H, Kannus P, Oja P, Pasanen M, et al. (2000) High-intensity exercise and bones of growing girls: A 9-month controlled trial. Osteoporos Int 11: 1010–7.
12. MacKelvie K, McKay H, Khan K, Crocker P (2001) A school-based loading intervention augments bone mineral accrual in early pubertal girls. J Pediatr 139: 561–8.
13. MacKelvie K, Khan K, Petit M, Jansson P, Crocker P, et al. (2003) A school-based exercise intervention elicits substantial bone health benefits: a 2-year randomised controlled trial in girls. Pediatrics 112: 447–52.
14. Linden C, Aiborg H, Beijak J, Gardeell P, Karlsson M (2006) A school curriculum-based exercise program increases bone mineral accrual and bone size in prepubertal girls: two-year data from the Pediatric Osteoporosis Prevention (POP) study. J Bone Miner Res 21: 829–35.
15. Fuchs R, Bauer J, Snow C (2001) Jumping improves hip and lumbar spine bone mass in prepubescent children: A randomized controlled trial. J Bone Miner Res 16: 146–58.
16. McKay H, Petit M, Schutz R, Prior J, Barr S, et al. (2000) Augmented trochanteric BMD after modified physical education classes: a randomized school based exercise intervention study in prepubescent and early pubescent children. J Pediatr 136: 156–62.
17. Petit M, McKay H, MacKelvie K, Heinonen A, Khan K, et al. (2002) A randomised school-based jumping intervention confers site and maturity specific benefits on one structural properties in girls: a hip structural analysis study. J Bone Miner Res 17: 963–72.
18. McKay H, MacLean I, Petit M, MacKelvie-O’Brien K, Janss P, et al. (2005) Bounce at the Bell: a novel program of short bouts of exercise improves proximal femur bone mass in early pubertal children. Br J Sports Med 39: 521–6.
19. MacKelvie K, Petit M, Khan K, Beck T, McKay H (2004) Bone mass and structure are enhanced following a 2-year randomised controlled trial of exercise in prepubertal boys. Bone 34: 735–44.
20. Courtes D, Jaffe C, Lespessailles E, Benhamou L (2005) Cumulative effects of calcium supplementation and physical activity on bone accretion in premenarcheal children: a double-blind randomised placebo-controlled trial. Int J Sports Med 26: 332–8.
21. Van Langendonck L, Claessens A, Vlistnick R, Derom C, Beunen G (2003) Influences of weight-bearing exercises on bone acquisition in prepubertal monoszygotic female twins: a randomized controlled prospective study. Calcif Tissue Int 72: 666–74.
22. Gunter K, Baxter-Jones A, Mirwald R, Almsteed H, Fuchs R, et al. (2006) Impact exercise increases BMC during growth: An 8-year longitudinal study. J Bone Miner Res 23: 986–93.
23. Baptista F, Barrigas C, Vieira F, Santa-Kla-Ha R, Mil-Homeno P, et al. (2011) The role of lean body mass and physical activity in bone health in children. J Bone Miner Metab 30: 100–8.
24. Sardinha LB, Baptista F, Eldlund U (2008) Objectively measured physical activity and bone strength in 9 year old boys and girls. Pediatrics 122: 728–36.
25. Janke Z, Burns T, Torner J, Levy S, Paulo R, et al. (2001) Physical activity and bone measures in young children: the Iowa Bone Development Study. Pediatrics 107: 1367–93.
26. Gracia-Marco L, Morreno I, Ortega F, León F, Sioon I, et al. (2011) Levels of physical activity that predict optimal bone mass in adolescents: the HELENA study Am J Prev Med 40: 599–607.
27. Kriemler S, Zahner L, Puder J, Braun-Fahrlander C, Schnurrer C, et al. (2006) Weight-bearing exercises are more sensitive to physical exercise in boys than in girls during pre- and early puberty: a cross-sectional study. Osteoporos Int 19: 1749–58.
28. Schneider B, Gardell P, Johnell O, Karlsson M, Ornslien E, et al. (2001) Peripubertal moderate exercise increases bone mass in boys but not in girls: a population-based intervention study. Osteoporos Int 12: 230–8.
29. Janke Z, Gilmour J, Burns T, Levy S, Torner J, et al. (2006) Physical activity augments bone mineral accrual in young children: the Iowa Bone Development Study. J Pediatr 148: 793–9.
30. Jones G, Dwyer T (1998) Bone mass in prepubertal children: gender differences and the role of physical activity and sunlight exposure. J Clin Endocrinol Metab 83: 4274–9.
31. Cardadeiro G, Baptista F, Zymbal V, Rodrigues L, Sardinha L (2010) Ward’s area location, Physical Activity and body composition in 8 and 9 years old boys and girls. J Bone Miner Res 25: 1–10.
32. Kelly T, Wilson K, Heymsfield S (2009) Dual Energy X-Ray Absorptiometry Body Composition. Reference Values from NHANES 4: 1–8.
33. Garnett S, Hodg W, Blades B, Baur L, Peat J, et al. (2004) Relation between hormones and body composition, including bone, in prepubertal children. Am J Clin Nutr 80: 966–72.
34. Chumanov ES, Wall-Scheffler C, Heiderscheit BC (2008) Gender differences in walking and running on level and inclined surfaces. Clin Biomech 23: 1260–8.
35. Guerra-Garcia M (2011) Incidence of hip fractures due to osteoporosis in relation to the prescription of drugs for their prevention and treatment in Galicia, Spain Atencion Primaria 43: 82–8.
36. El Maghraoui A, Koumba B, Jroundi I, Achemlal L, Ahmed B, et al. (2005) Epidemiology of hip fractures in 2002 in Rabat, Morocco. Osteoporos Int 16: 597–602.
37. Shao C (2009) A nationwide seven-year trend of hip fractures in the elderly population of Taiwan. Bone 44: 123–9.
38. Lin WP, Wen CJ, Jiang GC, Hou SM, Chen CY, et al. (2011) Risk factors for hip fracture sites and mortality in older adults. J Trauma 71: 191–7.
39. Sardinha LB, Ornelas R, Andersen LB, Froberg K, Andersen S et al. (2008) Objectively measured time spent sedentary is associated with insulin resistance independent of overall and central body fat in 9-to 10-Year-Old Portuguese children. Diabetes Care 31: 569–75.
40. Tanner JM (1962) Growth at adolescence. Oxford, Blackwell Scientific Publications.
41. Bonnick SL, Lewis LA (2002) Bone Densitometry for technologists. Totowa, New Jersey, Humana Press. 169–181.
42. Witzke K, Snow C (2000) Effects of plyometric jump training on bone mass in adolescent girls. Med Sci Sports Exerc 32: 1051–7.