Daily Physical Education in the School Curriculum in Prepubertal Girls During One Year is Followed by an Increase in Bone Mineral Accrual and Bone Width - Data from the Prospective Controlled Malmö Pediatric Osteoporosis Prevention (POP) Study

Ö. Valdimarsson MD., PhD., C. Linden MD., O. Johnell MD., PhD.,

P. Gardsell MD., PhD. and M. K. Karlsson MD., PhD.

Clinical and Molecular Osteoporosis Research Unit, Department of Clinical Sciences,

Lund University, Malmö University Hospital, SE-205 02 Malmö, Sweden

Correspondence: Correspondence: Örnólfur Valdimarsson, Department of Orthopaedics, Malmö University Hospital, SE - 205 02 Malmö, Sweden. Tel: +46 40 331000 ; Fax: +46 40 336200; E-mail: ornolfur.valdimarsson@med.lu.se

Running title: Exercise and accrual of bone mass in girls.

Abstract

The aim of this study was to evaluate a general school-based one-year exercise intervention program in a population-based cohort of girl at Tanner stage I. Fifty-three girls aged 7 to 9 years were included. The school curriculum based exercise intervention program included 40 minutes per school day. Fifty healthy age-matched girls assigned to the general school curriculum of 60 minutes physical activity per week served as controls. Bone mineral content (BMC; g) and areal bone mineral density (aBMD; g/cm^2) were measured with dual X-ray absorptiometry (DXA) of the total body (TB), lumbar spine (L2–L4 vertebra), the third lumbar vertebra (L3), the femoral neck (FN) and the leg. Volumetric bone mineral density (vBMD; g/cm^3) and bone width were calculated at L3 and FN. Total lean body mass and total fat mass were estimated from the total body scan. No differences were at baseline found in age, anthropometrics or bone parameters when the groups were compared. The annual gain in BMC was in the cases 4.7 percentage points higher in lumbar spine and 9.5 percentage points higher in L3 than in the controls (both $p<0.001$). The annual gain in aBMD was in the cases 2.8 percentage points higher in lumbar spine and 3.1 percentage points higher in L3 than in the controls (both $p<0.001$). The annual gain in bone width was in the cases 2.9 percentage points higher in L3 than in the controls ($p<0.001$).

A general school-based exercise program in 7 - 9-year-old girls enhances the accrual of BMC and aBMD and increases bone width.

Key Words: Bone mineral density – Bone width – Girls - Physical activity – Prepubertal.

Introduction

Exercise during growth is generally accepted as one factor that could influence peak bone mass and possibly prevent future osteoporosis (1,2). Especially the ages 11.5 to 13.5 in girls could be of interest, as this is the years that include peak bone velocity, a period were 26 % of the adult bone mineral content (BMC) is gained, an amount that approximates the amount of bone lost during the postmenopausal years (3,4). Furthermore, 47 % of the adult BMC is acquired during the 4 perimenarcheal years (5), a period were small differences in the accrual of areal bone mineral density (aBMD) over several years may result in a great impact in the future fracture risk, as a decrease in aBMD by one SD are calculated to double the fracture risk (6-9). Previous cross-sectional and prospective uncontrolled and controlled exercise trials in children have supported this hypothesis when reporting that weight-bearing physical activity increase the accrual of aBMD predominantly in the pre- and early peri-pubertal years (10-17). The so far published prospective controlled exercise intervention studies, most elegant support this view (14-16,18-20), whereas similar effects could not be found in the post-pubertal period (12). Furthermore, published data indicate that the physical activity ought to include high-impact activities (21-24) and that the training should probably be initiated before or in early puberty (25), if the purpose is to reach the most beneficial anabolic skeletal effects. There is also evidence that moderate intense school-based physical education classes do increase the level of physical activity and improve the physical fitness (26). Less is known if similar intervention program increase aBMD and at what age the program should be initiated (25,27). In addition, it must also be clarified if moderate intense exercise in children could influence bone width, another trait that contribute to the bone strength (28).

With this knowledge we designed this study in order to evaluate whether a general moderately intense exercise intervention program, possible for all children to perform, within the school curriculum in a population-based cohort of girls at Tanner stage I, also including children not specifically interested in exercise, could increase the accrual of BMC, aBMD and increase bone width. The purpose was not to verify already known benefits of exercise as prevention of diabetes and overweight and reduction of cardiovascular risk factors.

Material and Methods

The Malmö Pediatric Osteoporosis Prevention (POP) Study, is a prospective controlled exercise intervention study, annually following skeletal development in girls from school start. Baseline measurements in the intervention group were performed in August and September, just after school start and before the exercise intervention started. The follow-up evaluations were carried out during the same months one year later. The controls, selected from three neighboring schools, were evaluated in November and December with the follow-up measurements performed during the same months but two years later. We accepted a two year follow-up in the controls, as data in the literature consistently infer that both growth and accrual of BMC and aBMD over a year and during the ages this study span, occur in a linear fashion (4,29-38). The cases achieved during the study period one summer break for 9 weeks, were no intervention was given, and the controls two summer breaks. From these data we calculated the annual changes in the evaluated traits (changes per 365 days).

All girls in grades 1 and 2 in one school in a middle-class area in Malmö, Sweden, chosen as the intervention school, were invited to attend. Out of 61 girls, 55 agreed to participate, an attendance rate of 90 %. One girl was excluded, as she was 11 months younger than the second-youngest girl. At follow-up, one girl declined participation, leaving 53 girls with a mean age of 7.7 ± 0.6 (range 6.5–8.7) at baseline to be included in this survey. The controls were volunteers from three neighboring schools in areas with a similar socioeconomic background to the cases. Sixty-four volunteers participated at baseline whereas at follow-up, 13 of the girls had moved out of the region or declined further participation, whereas one was excluded as she was being treated with growth hormone, leaving 50 controls with a mean age 7.9 ± 0.6 (range 6.8–8.9) at baseline to be included in this survey. All included were healthy Caucasian girls without any medication known to influence bone metabolism.

The intervention, which started at school start just after the baseline measurement was performed, consisted of the ordinary physical activity used within the Swedish school curriculum, now increased to 40 min per day (200 min per week). The physical education class was supervised by the ordinary teacher, so that no extra resources were needed. The intervention did not consist of any programs specifically designed as being osteogenic. Instead, the physical education included both indoor and outdoor general physical activities used in the Swedish school curriculum, mainly playing activities that included mostly ball games, running and jumping. In these ages the activities was predominantly mixed in a playing way so that virtually no specific sports training was conducted. The teachers also conducted a variety of different physical activities as not to bore the children with repeated standardized activities. This was done with the aim to minimizing the dropouts in the long term perspective, as reported to occur frequently in other exercise intervention studies (39). In the control schools, the same type of physical activities were used as in the intervention school, but at a level within the compulsory Swedish school curriculum of physical education, consisting of one to two sessions per week, in total 60 min per week.

The children were evaluated dressed in light clothes with no shoes. Bone mass was measured by dual-energy X-ray absorptiometry (DXA, DPX-L version 1.3z, Lunar®, Madison, WI). Pediatric software was used for children with a weight below 30 kg. Bone mineral content (BMC, g) and areal bone mineral density (aBMD, g/cm^2) were evaluated for the total body, the lumbar spine (L2-L4 vertebra), the third lumbar vertebra (L3), the femoral neck (FN) and the legs. The width of the L3 vertebra was estimated from the antero-posterior spine scan as the distance from one edge of the L3 vertebra to the other edge. The FN width was calculated from the hip scan as the FN area divided by the scan length of the measured FN area.

Volumetric bone mineral density (vBMD, g/cm^3) was calculated for L3 using the algorithm introduced by Carter (40), and for the FN using the formula vBMD = BMC/estimated FN volume (π x r^2 x FN length) where r = FN mid-diameter/2, assuming the FN to be cylindrical (41-43). During the measurement of the lumbar spine, the child was supine, and the physiological lumbar lordosis was flattened by elevation of the knees. The precision, evaluated by duplicate measurements in 14 healthy young adults, was for aBMD total body 0.4%, lumbar spine 0.5% and FN 1.6%. Daily calibration of the machine was executed with the Lunar® phantom. The technicians in our research group performed all the measurements and the software analyses. Body weight was measured with an electric scale to the nearest 0.1 kg and body height by a wall-tapered height meter to the nearest 0.5 cm.

A questionnaire, previously used in several studies (44-47), evaluated lifestyle factors such as socioeconomic and ethnic background, diseases, medications, fractures, dairy products, exclusion of anything in the food, coffee consumption, smoking, alcohol intake and physical activity. The total time spent in physical activity was calculated as weekly time spent in organized sport within the school curriculum and during leisure time. Tanner staging (48), assessed by self-grading using photographs, assessed the maturity of the children.

Informed consent was obtained from parents or guardians of participants prior to the study start. The study was approved by the Ethics Committee of Lund University and the Radiographic Committee at Malmö University Hospital, Malmö, Sweden. The Swedish Data Inspection Board approved the data collection and the setup of the database.

Statistical calculations were performed with Statistica®, version 5.3 (StatWin®). Data are presented as mean ± SD. Student's *t*-test between means and Fisher's exact test was used for group comparisons. ANCOVA/MANCOVA were used to adjust for chronological age at baseline and the annual increment in height and weight in the follow-up evaluations. Spearman's rank correlation was used to correlate the total mean physical activity, calculated as the mean of the total physical activity at baseline and at follow-up, with changes in the bone parameters during the study period. A p-value of < 0.05 was considered as a statistically significant difference.

Results

There were no differences at baseline in lifestyle factors when cases and controls were compared (Table 1). All girls remained premenarcheal and at Tanner I during the study. Before the intervention was initiated there was no difference in the duration of performed physical activity when the groups were compared. After the intervention was initiated, the cases spent more time on physical activity both in school and in total compared to the controls (Table 1). In addition, there was no difference at baseline in anthropometrics or bone parameters when cases and controls were compared (Table 2).

During the study period, the annual gain in BMC was higher in the intervention group than in the controls, in the lumbar spine 16.3% versus (vs.) 11.6% ($p<0.001$) and in the L3 vertebra 19.8% vs. 10.3% ($p<0.001$) (Figure 1) (Table 2). The annual gain in aBMD was also higher in the intervention group, in the lumbar spine 6.6% vs. 3.8% ($p<0.001$) and in the L3 vertebra 6.7% vs. 3.6% ($p<0.001$) (Figure 1). Furthermore, the annual increase in bone width was also higher in the intervention group than in the controls, at the L3 vertebrae 6.0% vs. 3.1% ($p<0.001$) (Figure 1). There was no difference in the vBMD gain when cases and controls were compared (Table 2). The annual gain in total lean mass and total fat mass was also higher in the cases than in the controls, 11.1% vs. 9.2% ($p=0.002$) and 40.3% vs. 20.7% ($p<0.001$) (Table 2). All these group discrepancies also remained after adjusting for chronological age at baseline and annual changes in weight and height (Table 2). The total duration of physical activity during the study period, calculated as the duration of activity at baseline (after the intervention was started) and at follow-up divided by two, were when including all girls correlated with the annual changes in the third lumbar vertebra, in BMC ($r=0.26$, $p=0.008$), in aBMD ($r=0.31$, $p=0.002$) and in bone width ($r=0.20$, $p=0.04$) (Figure 2).

Discussion

This study suggests that merely by increasing the amount of general physical education in the school curriculum, the accrual of BMC and aBMD and the increase in bone width could be enhanced in a population-based cohort of prepubertal girls at Tanner stage I. In addition, the finding of a dose-response relationship between the total duration of physical activity and the gain in BMC, aBMD and bone width further strengthens the view that there exist a causal relationship between increased physical activity and increased accrual of the bone parameters. When discussing physical activity in prepubertal girls as a possible prevention strategy for future fractures, the results are of great importance as both BMC, aBMD and bone width contribute to the final bone strength (28).

The data in this study support previously published prospective controlled exercise intervention studies, trials that most elegant verify the exercise induced skeletal effects in young children (14-16,18-20). However, there are small differences in the current study design compared to previous reports. Some previous studies have included children that voluntarily could chose if they wanted to participate in the study or not, while we in the intervention group used a population based study design as to reduce the risk of self selection bias. Some previous studies have used specifically designed osteogenic intervention programs, such as jumping down a small height, programs that could be difficult to motivate the children to continue with for a long period, a hypothesis supported in previous reports (39). In contrast, we choose to continue with the ordinary physical classes including a variety of activities, hoping that this would make it easier to motivate the children to continue with a high level of exercise for a longer period.

This 12-month follow-up study is the second-longest published prospective controlled exercise intervention study in children. The longest follow-up so far spans 20 months, a study that supports the present data (19). However, it is still a matter of speculation whether the exercise-induced benefits are temporary or if they also remain with years of intervention, although cross-sectional and prospective observational studies do suggest that high physical activity is associated with a high peak bone mass (10-17).

A major strength of this study is that the intervention group could be regarded as a population-based cohort, as all girls in grades 1 and 2 in one school were invited and as the attendance rate was 90%. This also resulted in the inclusion of children with a minimal interest and participation in other physical activities, perhaps those who will achieve the most benefits from an intervention program. This is also why we chose a school as the intervention arena, as this is today the only place where we could reach all children. But, in spite of including less exercise-interested children, girls who probably put less effort into the intervention, the current report shows that exercise can be used to enhance the accrual of BMC and aBMD, and increase bone width in a population-based cohort of girls at Tanner stage I.

Furthermore, a variety of studies evaluating the effect of exercise in children have used specifically designed exercise programs with a high osteogenic effect. One study reported, for example, that exercise including jumping down a small step 10–30 minutes 3 times per week increased the accrual of aBMD in the greater trochanter by 1.4% over 8 months (14). Similar programs seem to be effective in the short term perspective (14-16,18-20), but also involve difficulties motivating the children to proceed with this type of exercise for a longer period

(14,49). This issue is extremely important, as continued physical activity seems to be essential if the purpose is to increase the peak bone mass (50) (51). However, this study implies that we could achieve similar benefits in the growing skeleton in girls by using a variety of activities generally used in the physical educational classes, a variety that possibly could make it easier to motivate the children to continue with the physical activity for a longer period.

We also deliberately let the ordinary schoolteachers lead the physical educational classes as previously done, without including any compulsory activities. By doing so, the ordinary lessons were kept intact, only increased in frequency compared to the control schools. Furthermore, we did not support the schools with extra resources, in order to show that a similar intervention could instantly be organized in all schools. This further increased the value of this report and enhances our inferences that physical activity could be a cost-effective strategy to increase aBMD and bone width, as this study confirms that exercise could be increased in the school curriculum with no extra costs.

Another strength of this study is the evaluation of bone width. Bone strength is not only dependent on aBMD but also on skeletal geometry, architecture and bone width (28). For example, women with spine fractures have smaller lumbar spine vertebrae but normal femoral neck size while women with hip fractures have a smaller femoral neck size but normal vertebral body size compared to controls (52). Also, straight mechanical calculations reveal the importance of bone width for resistance to fractures (28). There are even data suggesting that the age-related loss in aBMD is partly compensated by a periosteal apposition that increase the bone width, partly preserving bone strength (28). There are also previous cross-sectional studies indicating that high intense exercise increases bone width (53,54). However, this study further improves our knowledge by suggesting that also moderate physical activity in girls at Tanner stage I enhances the periosteal apposition.

There are also limitations in the study. The trial is not a true randomized study. However, randomization was not practical possible, as the principals and the teachers made it clear that the school schedule could not accept that some children were sent to physical activity during compulsory school hours while others were not. Secondly, neither the parents, nor the pupils accepted this. Third, randomization within the classes would through time have led to enormous problems with children crossing over between the groups. Therefore we accepted that one specific school was the intervention school, to increase the potential to separate the activity levels in the two cohorts for a longer period. Fourth, in the control schools we did not achieve such an attendance rate that the controls could be regarded as a population-based cohort. This increased the risk of introducing a self-selection bias at baseline. However, as the children in the control cohort come from a similar socioeconomic area compared to the intervention group and as at baseline there were no differences in lifestyle, time spent in organized activities, ethnicity, anthropometrics or bone parameters, it seems probable that also the controls could be regarded as representing normative Malmö data. Fifth, it could be debated whether the duration of organized physical activity is the best method to quantify the level of physical activity. Even with the same duration of activity, different types of activities may have a different osteogenic response, as could different intensities of the same type of exercise performed during the same period. In this survey we used a self-evaluating questionnaire to assess the duration of physical activity within and outside the school curriculum, used in several previous studies (44-47). The questionnaire was answered together with the parents so as to minimize errors, with the knowledge that there are difficulties estimating an accurate level of physical activity in young children. However, the main purpose of this study was not to define exactly the duration of physical activity in each child. The main aim was to evaluate whether an exercise intervention program could enhance the accrual of aBMD in a population-based cohort of prepubertal girls. It could also be supposed that the group differences would have been even greater if the controls at follow-up not had had a higher duration of activity out of school (table 1). Sixth, the estimation of bone width and vBMD in this study was done by the DXA technique. Even if this method is regarded as the golden standard when measuring aBMD, we know that the estimation of vBMD and bone width from a two-dimensional imaging technique is associated with errors, especially in growing children (55). Seventh, during the summer break, we do not know the activity level in the children. Eight, there was a dramatically greater annual gain in fat mass in the intervention group (40% vs. 20%) that we could not explain and could suggest some differences between the groups in food intake or other habits. But, these is also a possibility that exercise influence the skeleton through changes in soft tissue composition, the reason why we only adjusted for chronological age at baseline and changes in weight and height with the purpose to adjust only for differences in growth. Ninth, the problem with seasonal differences in growth and aBMD accrual was solved by measuring the children at the same month both at baseline and at follow-up and in both cases and controls. Finally, due to lack of resources in our research laboratory, the controls were re-measured first after 2 years. But, as all the girls stayed prepubertal and at Tanner stage I during the study period, we could compare the annual changes in the groups, as the literature consistently infer that both growth and accrual of BMC and aBMD is linear estimated over a year in these ages. This view is supported by Swedish data describing that growth rate is linear from age 6 to peak height velocity and that only 5.6 % of the girls had reached peak height velocity at age 10 years (30). Peak height velocity are usually reached later, in girls at mean age 11.7 while peak bone mineral accrual occur even later, at mean age 12.5 years in girls (31). That is, there is an approximate one-year lag between peak height velocity and peak bone mineral velocity (4,32). One Australian study further support this view when reporting that peak bone mass accrual and peak height velocity occur from Tanner stage II until menarche whereas the growth and bone mineral accrual are linear in Tanner stage I and the ages spanned in the current study (29). The notion that increase in bone mineral accrual in girls occur first after age 10 years and in Tanner stage II are supported in several independent cohorts of children (33-38). Based on the consistency in this extensive literature and the fact that we followed girls at younger ages and all in Tanner stage, the study design by comparing the annual changes must be regarded as acceptable, even if there was a 1-year follow-up in the cases and a 2-year follow-up in the controls.

In summary, a school-based exercise intervention program during the first school years in pre-pubertal girls at Tanner stage I seem to increase the accrual of BMC and aBMD and increase the gain in bone width. We conclude that if the exercise-induced skeletal benefits remain in a long-term perspective this supports the view that physical activity could be recommended as a strategy to increase peak bone mass and increase the bone resistance to fracture.

Acknowledgments

Financial support for this study was received from the Swedish Research Council K2004-73X-14080-04A, Centre for Athletic Research 121/04, the Herman Jarnhardt and the Region Skane Foundations.

Table 1. Background factors at baseline and organized physical activity at baseline and at follow-up in the girls within the exercise intervention group (n=53) and in the control group (n=50). Data is presented as numbers of girls with the proportion (%) within brackets or as mean ± SD.

	Cases	Controls
Background factors		
Excluding diary products	0 (0%)	0 (0%)
Coffee cups per week	0 (0%)	0 (0%)
Tried to lose weight	1 (2%)	0 (0%)
Smoking/Alcohol	0 (0%)	0 (0%)
Chronic disease	3 (7%)	1 (2%)
Ongoing medication	5 (11%)	2 (4%)
Fractures	7 (15%)	7 (15%)
Menarche	0 (0%)	0 (0%)
Organized physical activity (hrs/w)		
Baseline		
Outside the school	0.7 ± 1.2	1.3 ± 1.6
School curriculum (after intervention in cases)	3.3	1.0 ‡
Total physical activity	4.0 ± 1.2	2.3 ± 1.6 ‡
Follow-up		
School curriculum	3.3	1.0 ‡
Outside the school	1.1 ± 1.3	1.9 ± 1.9 *
Total physical activity	4.4 ± 1.3	2.9 ± 1.9 ‡

* $p < 0.05$, ‡ $p < 0.001$.

Table 2. Baseline data and annual changes evaluating the effect of one year of exercise intervention in anthropometry and bone mineral parameters in the intervention group and the control group. Data presented as mean ± SD. Comparison of annual changes in cases and controls presented with p-value unadjusted and p-value adjusted for growth parameters.

	Baseline		Annual changes		P unadjusted	P adjusted for Chronological age, annual changes in weight and height
	Cases n=53	Controls n=50	Cases n=53	Controls n=50		
Age (yrs)	7.7±0.6	7.9±0.6				
Birthweight (g)	3328±485	3182±766				
Weight (kg)	27.6±5.5	27.3±5.5	3.5±2.2	3.2±1.3	0.42	
Height (cm)	128.0±5.2	129.1±7.9	6.0±1.2	5.7±1.0	0.20	
Lean mass (kg)	19.8±2.3	20.2±2.8	2.2±0.7	1.9±0.6	**0.01**	
Fat mass (kg)	5.3±3.9	5.2±3.3	1.9±1.5	1.0±0.9	**<0.001**	
BMC (g)						
Total body	935±148	933±181	140.9±53.0	137.3±41.2	0.70	0.43
L2-L4	15±3	15±3	2.4±1.1	1.8±0.7	**0.001**	**0.01**
L3	5±1.0	5.3±1.2	0.94±0.63	0.53±0.30	**<0.001**	**<0.001**
Femoral neck	2.6±0.6	2.7±0.7	0.37±0.77	0.27±0.32	0.39	0.64
Leg	282±59	283±72	66.2±25.2	64±21.1	0.64	0.68
aBMD (g/cm^2)						
Total body	0.843±0.04	0.839±0.05	0.026±0.01	0.024±0.01	0.38	0.67
L2-L4	0.693±0.08	0.696±0.08	0.044±0.03	0.026±0.01	**<0.001**	**<0.001**
L3	0.708±0.07	0.709±0.08	0.047±0.03	0.025±0.02	**<0.001**	**<0.001**
Femoral neck	0.726±0.09	0.713±0.10	0.047±0.10	0.040±0.04	0.65	0.94
Leg	0.762±0.06	0.759±0.07	0.050±0.02	0.048±0.02	0.56	0.99
vBMD (g/cm^3)						
L3	0.268±0.03	0.262±0.03	0.004±0.01	0.001±0.01	0.36	0.23
Femoral neck	0.381±0.04	0.368±0.05	0.003±0.04	0.006±0.02	0.53	0.57
Width (cm)						
L3	2.9±0.2	2.9±0.3	0.17±0.12	0.09±0.06	**<0.001**	**<0.001**
Femoral neck	2.4±0.2	2.5±0.3	0.15±0.33	0.09±0.15	0.28	0.17

References

1. Hui SL, Slemenda CW, Johnston CC, Jr. 1990 The contribution of bone loss to postmenopausal osteoporosis. Osteoporos Int **1**(1)**:**30-4.
2. Kelly PJ, Morrison NA, Sambrook PN, Nguyen TV, Eisman JA 1995 Genetic influences on bone turnover, bone density and fracture. Eur J Endocrinol **133**(3)**:**265-71.
3. Bailey DA, Faulkner RA, McKay HA 1996 Growth, physical activity, and bone mineral acquisition. Exerc Sport Sci Rev **24:**233-66.
4. Bailey DA, McKay HA, Mirwald RL, Crocker PR, Faulkner RA 1999 A six-year longitudinal study of the relationship of physical activity to bone mineral accrual in growing children: the university of Saskatchewan bone mineral accrual study. J Bone Miner Res **14**(10)**:**1672-9.
5. Sabatier JP, Guaydier-Souquieres G, Benmalek A, Marcelli C 1999 Evolution of lumbar bone mineral content during adolescence and adulthood: a longitudinal study in 395 healthy females 10-24 years of age and 206 premenopausal women. Osteoporos Int **9**(6)**:**476-82.
6. Duppe H, Gardsell P, Nilsson B, Johnell O 1997 A single bone density measurement can predict fractures over 25 years. Calcif Tissue Int **60**(2)**:**171-4.
7. Hui SL, Slemenda CW, Johnston CC, Jr. 1989 Baseline measurement of bone mass predicts fracture in white women. Ann Intern Med **111**(5)**:**355-61.
8. Johnell O, Gullberg B, Kanis JA, Allander E, Elffors L, Dequeker J, Dilsen G, Gennari C, Lopes Vaz A, Lyritis G, et al. 1995 Risk factors for hip fracture in European women: the MEDOS Study. Mediterranean Osteoporosis Study. J Bone Miner Res **10**(11)**:**1802-15.
9. Cummings SR, Nevitt MC, Browner WS, Stone K, Fox KM, Ensrud KE, Cauley J, Black D, Vogt TM 1995 Risk factors for hip fracture in white women. Study of Osteoporotic Fractures Research Group [see comments]. N Engl J Med **332**(12)**:**767-73.
10. Kannus P, Haapasalo H, Sievanen H, Oja P, Vuori I 1994 The site-specific effects of long-term unilateral activity on bone mineral density and content. Bone **15**(3)**:**279-84.
11. Haapasalo H, Sievanen H, Kannus P, Heinonen A, Oja P, Vuori I 1996 Dimensions and estimated mechanical characteristics of the humerus after long-term tennis loading. J Bone Miner Res **11**(6)**:**864-72.
12. Blimkie CJ, Rice S, Webber CE, Martin J, Levy D, Gordon CL 1996 Effects of resistance training on bone mineral content and density in adolescent females. Can J Physiol Pharmacol **74**(9)**:**1025-33.
13. Bradney M, Pearce G, Naughton G, Sullivan C, Bass S, Beck T, Carlson J, Seeman E 1998 Moderate exercise during growth in prepubertal boys: changes in bone mass, size, volumetric density, and bone strength: a controlled prospective study [see comments]. J Bone Miner Res **13**(12)**:**1814-21.
14. McKay HA, Petit MA, Schutz RW, Prior JC, Barr SI, Khan KM 2000 Augmented trochanteric bone mineral density after modified physical education classes: a randomized school-based exercise intervention study in prepubescent and early pubescent children [see comments]. J Pediatr **136**(2)**:**156-62.
15. Morris FL, Naughton GA, Gibbs JL, Carlson JS, Wark JD 1997 Prospective ten-month exercise intervention in premenarcheal girls: positive effects on bone and lean mass. J Bone Miner Res **12**(9)**:**1453-62.
16. Fuchs RK, Bauer JJ, Snow CM 2001 Jumping improves hip and lumbar spine bone mass in prepubescent children: a randomized controlled trial. J Bone Miner Res **16**(1)**:**148-56.
17. Bass S, Pearce G, Bradney M, Hendrich E, Delmas PD, Harding A, Seeman E 1998 Exercise before puberty may confer residual benefits in bone density in adulthood: studies in active prepubertal and retired female gymnasts. J Bone Miner Res **13**(3)**:**500-7.
18. Heinonen A, Sievanen H, Kannus P, Oja P, Pasanen M, Vuori I 2000 High-impact exercise and bones of growing girls: a 9-month controlled trial. Osteoporos Int **11**(12)**:**1010-7.
19. MacKelvie KJ, Khan KM, Petit MA, Janssen PA, McKay HA 2003 A school-based exercise intervention elicits substantial bone health benefits: a 2-year randomized controlled trial in girls. Pediatrics **112**(6 Pt 1)**:**e447.
20. Mackelvie KJ, McKay HA, Khan KM, Crocker PR 2001 A school-based exercise intervention augments bone mineral accrual in early pubertal girls. J Pediatr **139**(4)**:**501-8.
21. Courteix D, Lespessailles E, Peres SL, Obert P, Germain P, Benhamou CL 1998 Effect of physical training on bone mineral density in prepubertal girls: a comparative study between impact-loading and non-impact-loading sports. Osteoporos Int **8**(2)**:**152-8.

22. Heinonen A, Oja P, Kannus P, Sievanen H, Haapasalo H, Manttari A, Vuori I 1995 Bone mineral density in female athletes representing sports with different loading characteristics of the skeleton. Bone **17**(3)**:**197-203.
23. Cassell C, Benedict M, Specker B 1996 Bone mineral density in elite 7- to 9-yr-old female gymnasts and swimmers. Med Sci Sports Exerc **28**(10)**:**1243-6.
24. Heinonen A, Kannus P, Sievanen H, Oja P, Pasanen M, Rinne M, Uusi-Rasi K, Vuori I 1996 Randomised controlled trial of effect of high-impact exercise on selected risk factors for osteoporotic fractures. Lancet **348**(9038)**:**1343-7.
25. Kannus P, Haapasalo H, Sankelo M, Sievanen H, Pasanen M, Heinonen A, Oja P, Vuori I 1995 Effect of starting age of physical activity on bone mass in the dominant arm of tennis and squash players. Ann Intern Med **123**(1)**:**27-31.
26. Briss PA, Zaza S, Pappaioanou M, Fielding J, Wright-De Aguero L, Truman BI, Hopkins DP, Mullen PD, Thompson RS, Woolf SH, Carande-Kulis VG, Anderson L, Hinman AR, McQueen DV, Teutsch SM, Harris JR 2000 Developing an evidence-based Guide to Community Preventive Services--methods. The Task Force on Community Preventive Services. Am J Prev Med **18**(1 Suppl)**:**35-43.
27. Haapasalo H, Kannus P, Sievanen H, Pasanen M, Uusi-Rasi K, Heinonen A, Oja P, Vuori I 1998 Effect of long-term unilateral activity on bone mineral density of female junior tennis players. J Bone Miner Res **13**(2)**:**310-9.
28. Ahlborg HG, Johnell O, Turner CH, Rannevik G, Karlsson MK 2003 Bone loss and bone size after menopause. N Engl J Med **349**(4)**:**327-34.
29. Bass S, Delmas PD, Pearce G, Hendrich E, Tabensky A, Seeman E 1999 The differing tempo of growth in bone size, mass, and density in girls is region-specific [see comments]. J Clin Invest **104**(6)**:**795-804.
30. Liu YX, Wikland KA, Karlberg J 2000 New reference for the age at childhood onset of growth and secular trend in the timing of puberty in Swedish. Acta Paediatr **89**(6)**:**637-43.
31. MacKelvie KJ, Khan KM, McKay HA 2002 Is there a critical period for bone response to weight-bearing exercise in children and adolescents? a systematic review. Br J Sports Med **36**(4)**:**250-7; discussion 257.
32. Fournier PE, Rizzoli R, Slosman DO, Theintz G, Bonjour JP 1997 Asynchrony between the rates of standing height gain and bone mass accumulation during puberty. Osteoporos Int **7**(6)**:**525-32.
33. Faulkner RA, McCulloch RG, Fyke SL, De Coteau WE, McKay HA, Bailey DA, Houston CS, Wilkinson AA 1995 Comparison of areal and estimated volumetric bone mineral density values between older men and women. Osteoporos Int **5**(4)**:**271-5.
34. Bonjour JP TGea 1991 Critical years and stages of puberty for spinal and femural bone mass accumulation during adolscence. J Clin Endocrin Soc **73:**555-63.
35. Kroger H, Kotaniemi A, Vainio P, Alhava E 1992 Bone densitometry of the spine and femur in children by dual-energy x-ray absorptiometry. Bone Miner **17**(1)**:**75-85.
36. Kroger H, Kotaniemi A, Kroger L, Alhava E 1993 Development of bone mass and bone density of the spine and femoral neck--a prospective study of 65 children and adolescents. Bone Miner **23**(3)**:**171-82.
37. Theintz G, Buchs B, Rizzoli R, Slosman D, Clavien H, Sizonenko PC, Bonjour JP 1992 Longitudinal monitoring of bone mass accumulation in healthy adolescents: evidence for a marked reduction after 16 years of age at the levels of lumbar spine and femoral neck in female subjects. J Clin Endocrinol Metab **75**(4)**:**1060-5.
38. Glastre C, Braillon P, David L, Cochat P, Meunier PJ, Delmas PD 1990 Measurement of bone mineral content of the lumbar spine by dual energy x-ray absorptiometry in normal children: correlations with growth parameters. J Clin Endocrinol Metab **70**(5)**:**1330-3.
39. Bass LS I-BS, Naughton G, Daly R, Nowson C, Briganti E, Austen S. 2003 Limitations of long term exercise interventions aimed at improving bone health in normally active boys. Journal of Bone and Mineral Research (18)**:**294.
40. Carter DR, Bouxsein ML, Marcus R 1992 New approaches for interpreting projected bone densitometry data. J Bone Miner Res **7**(2)**:**137-45.
41. Lu PW, Cowell CT, SA LL-J, Briody JN, Howman-Giles R 1996 Volumetric bone mineral density in normal subjects, aged 5-27 years. J Clin Endocrinol Metab **81**(4)**:**1586-90.
42. Ruff CB, Hayes WC 1988 Sex differences in age-related remodeling of the femur and tibia. J Orthop Res **6**(6)**:**886-96.
43. Cowell CT, Lu PW, Lloyd-Jones SA, Briody JN, Allen JR, Humphries IR, Reed E, Knight J, Howman-Giles R, Gaskin K 1995 Volumetric bone mineral density--a potential role in paediatrics. Acta Paediatr Suppl **411:**12-6, discussion 17.
44. Duppe H, Gardsell P, Johnell O, Nilsson BE, Ringsberg K 1997 Bone mineral density, muscle strength and physical activity. A population-based study of 332 subjects aged 15-42 years. Acta Orthop Scand **68**(2)**:**97-103.

45. Sundberg M, Duppe H, Gardsell P, Johnell O, Ornstein E, Sernbo I 1997 Bone mineral density in adolescents. Higher values in a rural area--a population-based study of 246 subjects in southern Sweden. Acta Orthop Scand **68**(5)**:**456-60.
46. Sundberg M, Duppe H, Gardsell P, Johnell O, Ornstein E, Sernbo I 1997 Bone mineral density in adolescents. Higher values in a rural area--a population-based study of 246 subjects in southern Sweden. Acta Orthop Scand **68**(5)**:**456-60.
47. Sundberg M, Gardsell P, Johnell O, Karlsson MK, Ornstein E, Sandstedt B, Sernbo I 2001 Peripubertal moderate exercise increases bone mass in boys but not in girls: a population-based intervention study. Osteoporos Int **12**(3)**:**230-8.
48. Duke PM, Litt IF, Gross RT 1980 Adolescents' self-assessment of sexual maturation. Pediatrics **66**(6)**:**918-20.
49. Bass L 2003 Limitations of long term exercise interventions aimed at improving bone health in normally active boys. J Bone Miner Res **18**((2 Suppl))**:**M 151.
50. Magnusson H, Karlsson M 2001 Bone mass, bone size and bone metabolism in male and female athletes. The Annual Meeting of the Swedish Orthopedic Society, Vaxjo, Sweden**:**95.
51. Valdimarsson O 2005 Reduced training is associated with increased loss of bone mineral density. J Bone Miner Res **www.jbmr-online.com**.
52. Duan Y, Parfitt A, Seeman E 1999 Vertebral bone mass, size, and volumetric density in women with spinal fractures. J Bone Miner Res **14**(10)**:**1796-802.
53. Haapasalo H, Kontulainen S, Sievanen H, Kannus P, Jarvinen M, Vuori I 2000 Exercise-induced bone gain is due to enlargement in bone size without a change in volumetric bone density: a peripheral quantitative computed tomography study of the upper arms of male tennis players. Bone **27**(3)**:**351-7.
54. Kontulainen S, Sievanen H, Kannus P, Pasanen M, Vuori I 2003 Effect of long-term impact-loading on mass, size, and estimated strength of humerus and radius of female racquet-sports players: a peripheral quantitative computed tomography study between young and old starters and controls. J Bone Miner Res **18**(2)**:**352-9.
55. Zebaze RM WF, Juliano-Burns S, Evans A, Seeman E. 2004 The femoral neck is ellipsoid: the assumption of circularity or parallelepipedal shape introduces errors in volume and volumetric bone mineral density. J Bone Miner Res **19**(Suppl 1)**:**366.

Figure legends

Figure 1. The relative annual changes (%) in girls aged 7 to 9 years at the third lumbar vertebra in bone mineral content (BMC), bone mineral density (BMD) and bone width in the intervention group (n=53) who received 40 minutes of daily physical education within the school curriculum (200 minutes per week) and in controls (n=50) who received 1 to 2 sessions per week (60 minutes per week).

Figure 2. The correlation between total duration of physical activity, calculated as duration of activity at baseline, after the intervention was started, and at follow-up divided by two, and the annual changes in the third lumbar vertebra bone mineral content (BMC), bone mineral density (BMD) and bone width in girls aged 7 to 9 years (including both the intervention and the control group).

Annual Changes in the Third Lumbar Vertebra

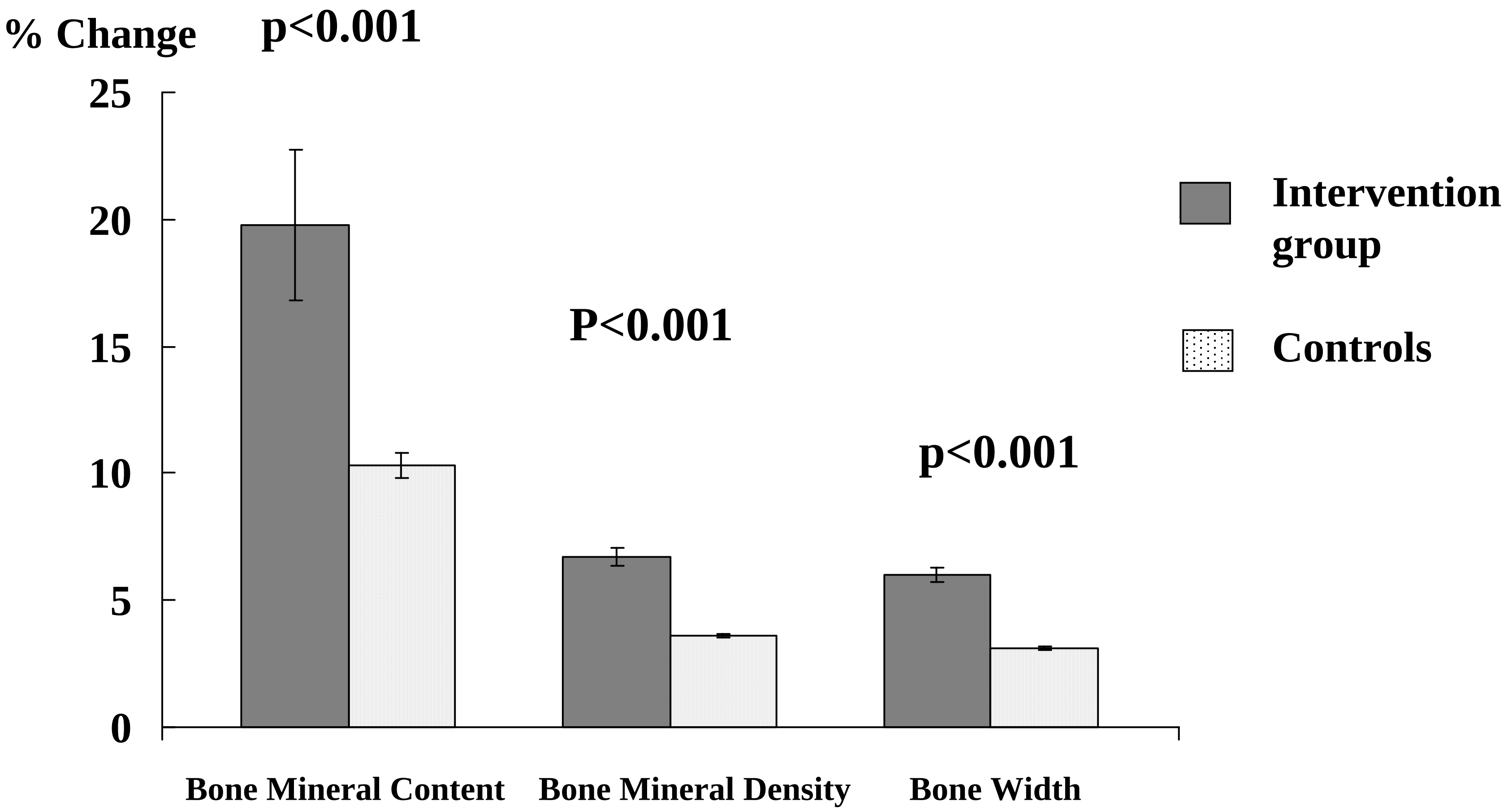

Figure 2: Annual Changes in the Third Lumbar Vertebra