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Walking Age Does Not Explain Term vs. Preterm Differences in Bone Geometry

Haifa Abou Samra and Bonny Specker

EA Martin Program in Human Nutrition, South Dakota State University, Brookings, SD 57007, United States

Abstract

Objective—To elucidate the relationship between bone geometry and onset of walking in former term and preterm children.

Study design—Cross-sectional study of 128 preschool children aged 3 to 5 years who had pQCT measures of bone size at the distal tibia. Linear models were developed, stratifying by sex, to determine whether bone differences between children born term and preterm were due to differences in walking age.

Results—Children with history of preterm birth walked later than term children (12.4 ± 0.5 vs. 10.9 ± 0.2 months, $p=0.004$); however, gestation-corrected walking age (11.4 ± 0.5 for preterm) did not differ. In multiple regression analysis, boys born preterm had larger periosteal and endosteal circumferences, and smaller cortical thickness and area, than boys born term (least square means 49.7 ± 1.3 mm, 43.0 ± 1.8 mm, 1.1 ± 0.11 mm, and 49.3 ± 3.2 mm² versus 47.0 ± 0.5 mm, 38.5 ± 0.7 mm, 1.4 ± 0.04 mm, and 56.9 ± 1.2 mm² respectively, all $p < 0.05$). Preterm birth remained statistically significant after adding age of walking to the models, but no longer significant when current activity levels were included.

Conclusion—Greater periosteal and endosteal circumferences, with smaller cortical bone thickness and area, were found in former preterm boys, but not girls, and were explained by differences in current activity levels, not age of walking.

Keywords

Walking; bone size; bone geometry; physical activity; preschool children; body weight

Although genetics is the major determinant of bone mass, modifiable environmental influences such as physical activity and diet, have been shown to optimize an individual's genetic potential (1). In addition, infant birth weight and growth are thought to influence adult bone geometry (2). An association between adult and childhood bone has been suggested and peak bone mass attained in early life is considered a major factor in predicting future osteoporosis risk (1,3). Beneficial bone effects of early childhood activity may persist beyond the period of the increased activity (4) and several longitudinal studies have shown high childhood activity to be associated with high adult bone density (5,6). The Department of Health and Human Services has recommended that physical activity levels should increase in early childhood to optimize bone health (7).

Address for correspondence: Bonny Specker, PhD, EA Martin Program in Human Nutrition, Box 2204 EAM Bldg, South Dakota State University, Brookings, SD 57007, Office 605 688 4645, Fax 605 688 4220. E-mail: Bonny.Specker@sdstate.edu

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Walking is thought to place significant strain on bone, and adult studies have shown walking to enhance bone density (8). Increased bone loading also is associated with increased bone size (9). Theoretically, walking during infancy should exert the same effect by placing strains on the skeleton leading to a beneficial bone response, and walking at an earlier age will lead to greater cumulative loading than walking at a later age. In a longitudinal study of 20 children, Ruff investigated the relationship between body size, muscle size, and bone structural development and strength. Data were obtained from serial radiographs that were taken at approximately six month intervals from near birth to late adolescence (10). He found that femoral bone strength velocity increased earlier during the second year with the beginning of walking, and humeral strength velocity declined as crawling stopped and assistance in standing and walking began. Because the peaks in bone strength velocities were not accompanied by changes in body size, Ruff suggested that bone strength is strongly dependent on mechanical loading (10). This study is the only one that we are aware of that has examined the association between bone strength and the beginning of walking.

The aim of this study was to investigate the relationship between bone size and the onset of independent walking in preschool children, and to determine whether walking age explains the bone differences between preschool children born preterm compared to children born at term. We previously reported similar periosteal circumferences, but larger endosteal circumference and smaller cortical area of the tibia in preschool children who were reported to be born preterm than those born at term (11). In the current study, we hypothesized that: 1) children who walk earlier will have larger periosteal circumference and cortical area and thickness, and smaller endosteal circumference, than children who walk later; and 2) children with history of preterm birth begin to walk at an older age compared to children born at term and this difference in walking age will explain the larger endosteal circumference and smaller cortical bone area that is observed in children with history of preterm birth.

Methods

Baseline data from 239 children aged 3-5 years who participated in a one year, randomized, placebo-controlled physical activity and calcium supplementation trial were used for this analysis (11). Out of the 239, 110 had tibia bone measurements with movement and were excluded from the study. A comparison between those who were excluded and those who were not, showed that excluded children were younger, lighter and shorter (all, $p < 0.001$). There were no differences in sex, history of preterm birth, physical activity levels, and walking age between excluded children and those not excluded.

Parents completed questionnaires that provided information on sex, race, feeding history during the first month of life, and parents' education. Parents were asked to recall the age the child first walked. Mothers also recalled whether the child was born early and if so, how many weeks based on their due date as determined by their obstetrician. Children were considered preterm if they were born at or less than 37 weeks gestation. The average length of prematurity was 4.1 ± 2 weeks. Fifty-six of the study participants were classified as Caucasian and 8 as non-Caucasian. Information on ethnicity and birth weight was not obtained. Although we did not collect specific information on whether the child was a multiple gestation, we did not have sibs with the same birth date in the study sample. Bone measurements using peripheral quantitative computerized tomography (pQCT) of the 20% distal tibia were obtained. PQCT measures included periosteal and endosteal circumferences, cortical thickness, and cortical bone area (cortBA). Settings for pQCT are described elsewhere (15). Our coefficients of variation in children aged 3-5 years are 3.6% for periosteal circumference and 5.4% for cortBA. Percent body fat (%BF) measurements were obtained using DXA whole body scans (6,15). The total effective dose of radiation exposure from both the pQCT and DXA was less than one mrem,

which is significantly lower than the allowable effective dose of 500 mrem per year (NCRP Report # 116).

Study participants wore accelerometers for a 48-hr period and data were expressed as percent of time spent in moderate plus vigorous activity (counts > 500/minute) or percent of time spent in vigorous activity (counts > 1000/minute). Methods and validity of Actiwatch motion sensors were previously described (11,16). Activity levels at baseline were available for 94% of the females and 83% of the males. Parents and child-care providers completed 3-day food records (2 weekdays and 1 weekend day). Records were reviewed for completeness by study personnel and nutrient intake analysis was performed using Nutritionist V database (First Data Bank, San Bruno, CA, USA). Quartiles of calcium intake (mg /day) were determined from lowest to highest as less than 709 mg/day, 709 to 875 mg/day, 876 to 1036 mg/day, and more than 1036 mg/d. The study was approved by the South Dakota State University Human Subjects Committee and parental written informed consent was obtained.

Data were entered onto an Access database and analyses were performed using JMP statistical software (SAS Institute, Cary, NC, USA). Data were tested for normality and relationships between bone measures and anthropometric measurements (height, weight, %BF), demographic characteristics and potential cofounders were examined. Data were stratified by sex due to differences in bone measures, percent body fat, and activity levels. Significant predictors ($p \leq 0.05$) of periosteal and endosteal circumferences, cortBA, and cortical thickness were determined by stepwise regression analysis. Once multiple regression models for each of the outcome variables were obtained, walking age and gestation-corrected walking age were individually added to the model to determine whether either one explained a significant amount of the remaining error. The significance of all two-way interactions were tested for significance. Walking age was adjusted by subtracting the months born early (number of weeks/4.3) from walking age. Data are mean $\pm$ SEM unless otherwise stated.

Results

There were 129 children with pQCT scans at baseline who had no movement. However, one of these children had a reported walking age of 7 months and an adjusted walking age of 5.6 months (< -3 SD from the mean). This was considered a recall error and this child was excluded from the analysis. There were 64 females and a total of 14 children with history of preterm birth. Overall, children with history of preterm birth walked later than children with term birth (12.4 ± 0.5 vs. 10.9 ± 0.2 months, $p=0.004$), but this difference was not significant for gestation-corrected age at walking (11.4 ± 0.5 vs. 10.9 ± 0.2 months, $p = 0.34$) (Table I). There were no sex differences in any of the bone measures, although boys had lower percent body fat and greater activity levels than girls (Table I). In both males and females bone measures were not associated with race, type of feeding during the first month, calcium intake, or season of enrollment.

Males

Preterm boys were older and had larger periosteal and endosteal circumferences than term boys (Table I). None of the bone measurements were associated univariately with walking age (Table II; available at www.jpeds.com). Periosteal circumference was univariately associated with weight ($r=0.34$, $p=0.005$) and height ($r=0.25$, $p=0.05$). Using multiple regression analysis, periosteal circumference was associated with age, weight, %BF, and history of preterm birth (Table III). Endosteal circumference was only associated with history of preterm birth in the univariate analyses. However, using multiple regression analysis endosteal circumference was associated with age, weight, %BF, and history of preterm birth (Table III). Cortical thickness was univariately associated with age ($r=0.46$, $p<0.001$), height ($r=0.40$, $p=0.001$), and weight ($r=0.31$, $p=0.01$). In multiple regression analysis, cortical thickness was associated with age

and history of preterm birth (Table III). CortBA was univariately associated with age ($r=0.55$, $p=0<0.001$), height ($r=0.54$, $p=.001$), and weight ($r=0.52$, $p=<0.001$). In multiple regression analysis, cortBA was associated with age, weight, and history of preterm birth (Table III). Least square means from the multiple regression models are shown in Figure 1[L1].

When walking age was added to the regression models, significant differences between term and preterm birth persisted (Table III). Similar findings were observed when walking age corrected for gestational age was added. However, when percent time in moderate plus vigorous activity was added to each of the multiple regression models shown in Table III, history of preterm birth no longer was a significant predictor of periosteal and endosteal circumferences, cortical thickness or cortBA ($p=0.46$, $p=0.26$, $p=0.06$, and $p=0.11$, respectively).

Females

There were no significant differences in any of the bone or anthropometric measures between term and preterm girls. Term and preterm girls had similar height, weight, percent body fat, and physical activity levels. Periosteal circumference was univariately associated with age ($r=0.33$, $p=0.007$), height ($r=0.43$, $p=<0.001$), and weight ($r=0.53$, $p=<0.001$). Age and height were not significant if weight was included in the analysis. Endosteal circumference was univariately associated with weight ($r=0.24$, $p=0.05$). Cortical thickness was univariately associated with age ($r=0.38$, $p=0.002$), height ($r=0.52$, $p<0.001$), and weight ($r=0.48$, $p<0.001$). In multiple regression analysis, height was the only variable associated with cortical thickness (Table IV; available at www.jpeds.com). CortBA was univariately associated with age ($r=0.48$, $p<0.001$), height ($r=0.66$, $p<0.001$), and weight ($r=0.65$, $p<0.001$). In multiple regression analysis, cortical BA was associated with both height and weight (Table IV).

Walking age was not associated with any of the bone measures (Table II) and similar findings were observed when walking age corrected of gestational age was added to the final regression models. Inclusion of percent time in moderate plus vigorous activity did not alter these findings.

Discussion

Due to previous reports of persistent effects of mechanical loading on bone geometry, we examined the effect of age of walking on tibial bone size. We hypothesized that children who walked at a younger age would have differences in bone geometry than children who walked at an older age, and because age of walking may differ between term and preterm children, we proposed that differences in age at walking would explain bone differences between term and preterm children. We did not, however, find any association between measures of bone size.

We examined the relationship between bone size and age at walking in preschool children. Onset of walking is influenced by several developmental factors, which are of environmental, neural and biomechanical origins, as well as the interactions among these factors (17). Biomechanical changes such as redistribution of leg fat and muscle mass, strengthening of back and abdominal muscles, and lowering of the infant's center of mass are necessary for walking to occur (17). Mechanical loading to bones of the lower extremities that occurs at the initiation of walking is associated with increased femoral bone strength and decreased humeral bone strength velocities (10). Whether age at attainment of independent walking has a persistent effect on the size of weight-bearing bones later in life remains unclear. We did not, however, observe such a relationship in the current study.

Previous studies that have investigated age of walking attainment in infants with history of preterm birth find that, even after adjusting for gestational age differences, preterm infants have later onset of walking attainment and different patterns of locomotion compared to term

infants (18,19). Investigators speculated that children with history of preterm birth, in the absence of cerebral palsy and pathological conditions, might have subtle problems with muscle power coordination and timing (18). Muscle deficiencies were believed to be due to delayed motor function and inability of preterm infants to coordinate flexion-extension activities (18). If such limitations in muscle function exist among preterm infants the activity levels may be affected and consequently mechanical loading of bone may be diminished. Although we found significant differences between term and preterm infants in the age of independent walking attainment, this difference was no longer significant when corrected for gestational age differences.

In our previous analysis of these data, we reported a 2.1% lower total body bone mineral content in children born preterm compared with children born at term. We also found a larger endosteal circumference, with a similar periosteal circumference, and a lower cortical bone area in children born preterm compared to children born at term (11). Former preterm children had significantly lower activity levels than former term children, and activity levels explained differences in bone measures between groups (11). However, in our previous analysis we did not stratify by sex. After stratifying by sex for the current analysis, history of preterm birth was no longer a significant predictor of bone measures in girls and there were no significant differences in any of the outcome and predictive variables between term and preterm girls. Preterm boys did, however, have larger periosteal and endosteal circumferences, with smaller cortical thicknesses and cortBA than former term boys, consistent with our previous analysis of boys and girls combined. The current findings are consistent with Backstrom et al. who found that prematurity-associated bone effects were more pronounced in males than females (20). Lower bone stress index (BSI) in the weight bearing tibia, due to smaller cortical bone area and a smaller cross-section at the mid-shaft, was apparent in males 18-27 years old who had history of preterm birth, but not in females. Backstrom et al. speculated that weaker muscle performance in younger men born preterm could have contributed to their findings (20).

Previous studies showed that male infants born preterm have lower survival rates than females, and show more signs of neurological dysfunction than females (21,22). Males born preterm, even by only 3-4 weeks were shown to have lower motor and social developmental scores than female counterparts (23). Our study is the first to find differential sex effects of gestational age on bone measures in preschool children. These differences between term and preterm boys remained significant when walking age was included in the analysis. When physical activity measures were included in the statistical analysis, however, history of preterm birth was no longer a significant predictor of bone size. Differences in physical activity between term and preterm children need be investigated further.

One of the limitations of the current study is that our measure of activity levels was only available for 94% of the females and 83% of the males. It is possible that the small number of males with activity measures makes it more difficult to detect term-preterm differences in a multiple regression analysis that includes activity levels. However, if these findings are confirmed in larger populations it would have significant implications for developmental assessment and interventions in boys with history of preterm birth. This study has other limitations. First, parents' recall of their child's age of independent walking and gestational age may be inaccurate because they were asked about events that occurred up to four years before data were collected. In addition, parents may have been biased towards reporting an earlier walking age than a later one, especially in children with history of preterm birth because delayed onset of walking might imply that their child had developmental limitations. In addition, defining walking age might vary from one parent to another depending on the number of steps a child would take before he or she is considered by the parents to be walking independently.

The average weeks of lost gestation in our study participants with history of preterm birth was approximately four weeks. Therefore, these infants were not likely to be very low birth weight, or to have had complicated postnatal medical history. Detecting a difference in outcome variables between groups of term and preterm children is more difficult than detecting a difference between term and very preterm children, and would require a larger sample size. If delayed onset of independent walking is associated with preterm birth and also a predictor of bone size, a larger sample size may be needed to detect such a relationship. Our post hoc power for detecting a difference in bone measures between children in lowest and highest walking quartiles was 48% for boys and 21% for girls with a sample size of 64. Our results also could have been confounded by other important factors such as birth weight, appropriateness of weight for gestational age, presence of complications during delivery, intensive care nursery admission, length of stay, medication use, and maternal drug use and smoking. Unfortunately, such information was not available to us and could not be included in the analysis.

In summary, bone size in preschool children was not associated with age of walking in either males or females. However, bone size was associated with history of preterm birth, but only among males. Greater periosteal and endosteal circumferences, but thinner bones with smaller cortical bone area were found in former preterm boys. Our findings need to be confirmed in larger populations and factors contributing to these differences need to be further explored.

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List of Abbreviations

BSI	bone stress index
cortBA	cortical bone area
%BF	percent body fat
pQCT	peripheral quantitative computerized tomography

Figure 1. Mean ($\pm$ SEM) periosteal (Peri-C) and endosteal circumferences (Endo-C), cortical thickness (CortThk) and cortical area (Cort A), and in term and preterm males. Black is preterm males and grey is term males

Figure 2. Mean ($\pm$ SEM) periosteal (Peri-C) and endosteal circumferences (Endo-C), cortical thickness (CortThk), and cortical area (Cort A) in term and preterm females. Black is preterm females and grey is term females. (online only)

Table 1

Population Characteristics by History of Preterm Birth and Gender.

Variables	Term * Females (58)	Preterm Females (6)	Term Males (56)	Preterm Males (8)	Female vs. Male ^a	Term vs. Preterm ^a
Outcome variables:						
Periosteal Circumference (mm)	46.5 ± 0.4	46.1 ± 1.3	46.9 ± 0.5	50.4 ± 1.4 ^b	0.18	0.08
Endosteal Circumference (mm)	38.5 ± 0.5	39.0 ± 1.5	38.5 ± 0.7	42.9 ± 1.9 ^b	0.49	0.04
Cortical thickness (mm)	1.3 ± 0.03	1.1 ± 0.1	1.3 ± 0.04	1.1 ± 0.1	0.35	0.09
Cortical bone area (mm ²)	54.1 ± 1.4	47.9 ± 4.4	56.2 ± 1.5	54.2 ± 4.0	0.23	0.24
Predictive Variables:						
Walking Age (months)	10.9 ± 0.2	11.8 ± 0.7	11.0 ± 0.2	12.8 ± 0.6	0.50	0.004
Gestation-Corrected Walking Age (months)	10.9 ± 0.2	11.1 ± 0.7	11.0 ± 0.2	11.6 ± 0.6	0.34	0.64
Age (year)	4.1 ± 0.1	3.9 ± 0.2	4.1 ± 0.1	4.6 ± 0.2	0.39	0.31
Height (cm)	103.8 ± 0.8	100.1 ± 2.4	103.7 ± 0.7	107.3 ± 1.8	0.46	0.76
Weight (kg)	17.2 ± 0.3	15.9 ± 1.1	17.3 ± 0.3	18.6 ± 0.9	0.47	0.83
Body fat (%)	28 ± 0.6	29 ± 1.8	24 ± 0.6	22 ± 1.6	<0.001	0.61
Calcium Intake (mg/d)	875 ± 38	781 ± 122	908 ± 40	876 ± 103	0.48	0.51
% Time in Moderate + Vigorous Activity	11.7 ± 0.5	8.9 ± 1.6	13.8 ± 0.4	11.9 ± 1.3	0.003	0.05

Data are mean ± SEM

* Term is defined as >37 weeks gestation.

^a Significance value based on 2-way ANOVA

^b Significantly different from term infants within the same gender, p<0.05

Table 2

Correlation Coefficients between Bone Measurements, Walking Age, and Gestation-Corrected Walking Age by Gender.

	Males	Females
Walking Age		
Periosteal Circumference (mm)	-0.10	-0.02
Endosteal Circumference (mm)	-0.04	-0.03
Cortical thickness (mm)	0.08	0.04
Cortical bone area (mm ²)	-0.13	0.03
Gestation-Corrected Walking Age		
Periosteal Circumference (mm)	-0.15	-0.01
Endosteal Circumference (mm)	-0.09	-0.03
Cortical thickness (mm)	-0.06	0.06
Cortical bone area (mm ²)	-0.12	0.06
% Time in Moderate+Vigorous Activity		
Periosteal Circumference (mm)	-0.09	-0.13
Endosteal Circumference (mm)	-0.04	-0.13
Cortical thickness (mm)	-0.05	0.04
Cortical bone area (mm ²)	-0.11	-0.03

Table 3

Multiple Regression Models Predicting Bone Measures in Males.

	Estimate	SE	P Value	Estimate	SE	P Value
Periosteal Circumference (mm) - full model						
				R² =0.31		
Intercept	48.98	4.32	<0.001	52.29	6.63	<0.001
Weight (kg)	1.11	0.26	<0.001	1.06	0.28	<0.001
% Body Fat	-39.67	12.57	0.003	-36.09	13.41	0.006
Age (y)	-2.57	0.96	0.01	-2.64	1.01	0.01
Preterm Birth (no)	-1.33	0.70	0.06	-1.58	0.81	0.06
Walking Age (months)	---	---	---	-0.21	0.31	0.51
Endosteal Circumference (mm)- full model						
				R² =0.24		
Intercept	49.87	6.09	<0.001	57.66	9.27	<0.001
Weight (kg)	1.02	0.36	0.007	0.9	0.39	0.03
% Body Fat	-40.04	17.73	0.03	-37.5	18.74	0.05
Age (y)	-4.21	1.36	0.003	-4.46	1.42	0.003
History of Preterm (no)	-2.23	0.98	0.03	-2.83	1.33	0.02
Walking age (months)	---	---	---	-0.44	0.43	0.31
Cortical Thickness (mm) - full model						
				R² =0.33		
Intercept	0.016	0.264	0.95	-0.514	0.453	0.26
Age (y)	0.288	0.060	<0.001	0.332	0.065	<0.001
History of Preterm (no)	0.141	0.057	0.02	0.184	0.063	0.005
Walking age (months)	---	---	---	0.029	0.023	0.22
Cortical Area (cm²) - full model						
				R² =0.47		
Intercept	-4.58	9.2	0.62	-25.68	15.29	0.10
Weight (kg)	1.47	0.49	0.004	1.76	0.52	0.001
Age (y)	7.72	2.04	<0.001	8.74	2.1	<0.001
History of Preterm (no)	3.77	1.71	0.03	5.29	1.88	0.007
Walking age (months)	---	---	---	0.99	0.71	0.17

Table 4

Multiple Regression Models Predicting Bone Measures in Females.

	Estimate	SE	P Value	Estimate	SE	P Value
<u>Periosteal Circumference (mm) - full model</u>						
				R² =0.28	R² =0.30	
Intercept	35.22	2.31	<0.001	37.39	3.0	<0.001
Weight (kg)	0.66	0.13	<0.001	0.69	0.14	<0.001
Walking Age (months)	---	---	---	-0.25	0.21	0.25
<u>Endosteal Circumference (mm) - full model</u>						
				R² =0.06	R² =0.07	
Intercept	32.55	3.07	<0.001	34.12	4.0	<0.001
Weight (kg)	0.35	0.18	0.05	0.38	0.19	0.05
Walking age (months)	---	---	---	-0.19	0.28	0.51
<u>Cortical Thickness (mm) - full model</u>						
				R² =0.28	R² =0.27	
Intercept	-1.16	0.5	0.02	-1.14	0.53	0.04
Weight (kg)	0.02	0.00	<0.001	0.03	0.00	<0.001
Walking age (months)	---	---	---	-0.01	0.02	0.42
<u>Cortical Area (cm²) - full model</u>						
				R² =0.46	R² =0.46	
Intercept	-38.92	24.37	0.11	-37.3	25.94	0.16
Weight (kg)	1.36	0.78	0.086	1.32	0.8	0.11
Height (cm)	0.67	0.34	0.05	0.75	0.36	0.04
Walking age (months)	---	---	---	-0.82	0.62	0.19