### Effect of Rhythmic Gymnastics on Volumetric Bone Mineral Density and Bone Geometry in Premenarcheal Female Athletes and Controls

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**Context and Objective:** Weight-bearing exercise during growth exerts positive effects on the skeleton. Our objective was to test the hypothesis that long-term elite rhythmic gymnastics exerts positive effects on volumetric bone mineral density and geometry and to determine whether exercise-induced bone adaptation is associated with increased periosteal bone formation or medullary contraction using tibial peripheral quantitative computed tomography and bone turnover markers.

**Design and Setting:** We conducted a cross-sectional study at a tertiary center.

**Subjects:** We studied 26 elite premenarcheal female rhythmic gymnasts (RG) and 23 female controls, aged 9–13 yr.

Main Outcome Measures: We measured bone age, volumetric bone mineral density, bone mineral content (BMC), cortical thickness, cortical and trabecular area, and polar stress strength index (SSIp) by peripheral quantitative computed tomography of the left tibia proximal to the distal metaphysis (trabecular) at 14, 38 (cortical), and 66% (muscle mass) from the distal end and bone turnover markers.

**Results:** The two groups were comparable according to height and chronological and bone age. After weight adjustment, cortical BMC, area, and thickness at 38% were significantly higher in RG (P < 0.005-0.001). Periosteal circumference, SSIp, and muscle area were higher in RG (P < 0.01-0.001). Muscle area was significantly associated with cortical BMC, area, and SSIp, whereas years of training showed positive association with cortical BMC, area, and thickness independent of chronological age.

Conclusions: RG in premenarcheal girls may induce positive adaptations on the skeleton, especially in cortical bone. Increased duration of exercise is associated with a positive response of bone geometry. (J Clin Endocrinol Metab 95: 2755–2762, 2010)

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Abbreviations: aBMD, Areal BMD; AG, artistic gymnastics; BMC, bone mineral content; BMD, bone mineral density; BMI, body mass index; BTM, bone turnover marker; CRTHK, cortical thickness; CSA, cross-sectional area; CV, coefficient of variation; DXA, dual-energy x-ray absorptiometry; ECLIA, electrochemiluminescence immunoassay; iPTH, intact PTH; 25(OH)D, 25-hydroxyvitamin D; P1NP, procollagen type 1 N-propeptide; pQCT, peripheral quantitative computed tomography; RG, rhythmic gymnastics or gymnasts; sCTX, serum β-C-telopeptide cross-linked collagen type I; SDS, so score; SSIp, polar stress strength index; vBMD. volumetric BMD.

Regular physical exercise is a modifiable lifestyle factor that might exert favorable effects on the skeleton. The inconsistency between studies (1) lies in the fact that the skeletal response to exercise (1, 2) depends both on training-related factors, such as type of exercise, intensity, and duration of the training program, and on individual-related factors such as baseline training status, nutritional and hormonal factors, and most importantly age at which training intervention is started. Importantly, the most consistent favorable results are reported in children and adolescents after weight-bearing exercise in which bone adaptation to applied loads is associated with gains in areal bone mineral density (aBMD) (1, 2) and positive changes in size and geometry (3).

Contrary to low or moderate intensity exercise, participation in elite artistic gymnastics (AG) and rhythmic gymnastics (RG) might result in a more favorable skeletal response due to the high-volume, high-impact training and the involvement at an early age. In particular, cross-sectional (4-7) and longitudinal studies (4, 7, 8) in AG show positive results of both aBMD and bone geometry (9, 10). Although the same responses concerning aBMD can be observed in RG, at least in cross-sectional studies (11, 12), the two types of gymnastics are quite different in terms of degree, type of impact loading, and possibly individualrelated factors such as nutrition and growth potential (13). Generally, RG requires less arm and body strength, loading mostly the lower limbs, whereas AG imposes a greater mechanical load in the upper and lower limbs and the trunk (14). Accordingly, a recent study comparing RG with AG showed higher regional aBMD and muscle mass in AG, despite delayed pubertal development (15).

Dual-energy x-ray absorptiometry (DXA) is the most widely used technique for measuring bone mineral content (BMC) and bone mineral density (BMD) in children due to its low cost, low radiation exposure, and availability. However, evaluation of bone strength in the growing skeleton, especially under the influence of exercise, requires information about both bone size and geometry, data that cannot be reliably derived from planar DXA images. Thus, although during growth aBMD (16) increases, volumetric BMD (vBMD) remains constant or slightly increases due to the proportional rise of both BMC and bone size (17). Moreover, DXA is unable to separate cortical from trabecular bone, components that respond differently to loading (18). In contrast, peripheral quantitative computed tomography (pQCT) has the advantage of simultaneous separate assessment of trabecular and cortical components as well as geometric characteristics of the peripheral skeleton, thus being suitable for the evaluation of training-induced skeletal adaptation (19, 20) at least in research settings due to the relative radiation exposure.

Given that the processes underlying bone geometry adaptation, especially at cortical sites, may include not only increased periosteal apposition but also increased endosteal apposition or decreased endosteal resorption (1, 3), the concomitant estimation of pQCT-derived variables with bone turnover markers (BTMs) might give some insight into the underlying mechanism, although data relating pQCT indices and fracture risk in children are scarce.

The present study was designed to test the hypothesis that long-term elite RG in premenarcheal females exerts positive effects on vBMD and geometry and to examine whether exercise-induced bone adaptation is associated with increased periosteal bone formation or medullary contraction using tibial pQCT and BTMs.

### **Subjects and Methods**

Participants were recruited from eight gymnastics centers and schools within the district of Attika over a 12-wk period from January to March 2008. Inclusion criterion was premenarcheal females aged 9-13 yr. Elite rhythmic gymnasts were included if they had been training for at least 2 yr. Gymnastics instructors identified children who exercised at least 24 h/wk and were the most promising athletes of their center. The parents of 53 gymnasts were approached, and 30 consented to participate. Controls were female premenarcheal school children, having only physical education-related activity, and matched with the athletes by age and height. Controls participating in other athletic activities, assessed by questionnaire, were excluded from the study (physical activity score <40 metabolic equivalents/day). All participants were Caucasian, were clinically healthy, were not receiving medication known to affect bone metabolism, and had not had any immobilizing surgery or fracture in the previous 12 months. The study was approved by the Ethics Committee of KAT Hospital and was conducted in accordance with the Declaration of Helsinki. Informed written consent was obtained from the parents or legal guardians of each child, and each child gave verbal assent to participate in the study.

### Anthropometric measurements and maturity assessment

Standing height was measured without shoes using a stadiometer to the nearest 0.1 cm (SECA 220; Seca Corp., Hanover, MD), and weight was measured using an electronic scale to the nearest 0.1 kg (Soehnle 7840; Soehnle Inc., Nassau, Germany), both in duplicate 1 wk apart [coefficients of variation (CVs), 0.97 and 0.98, respectively]. Body mass index (BMI) was calculated as weight (kilograms) per standing height (meters) squared. Skinfold sum is the sum of skinfold thickness (in millimeters) measured sequentially, in duplicate, using a skinfold caliper at biceps, triceps, subscapula, abdomen, suprailiac, calf, and thigh.

Skeletal age was calculated from left hand and wrist radiograph using the Greulich Pyle (21) method by two independent experienced pediatric radiologists, blinded to the status of the participant. In case of a difference of more than 6 months, x-rays were reevaluated blindly by two doctors (S.T. and I.P.) and an agreement was reached. Pubertal stage was self- assessed

using Tanner diagrams of five pubertal stages of breast development (22).

### Physical activity and dietary assessment

Based on their training logs, athletes performed two training sessions per day, 6 d/wk, with a total training volume of more than 20,000 foot-contacts per year. Controls participated only in physical education classes and had minimal physical activity weekly as reported in a 4-d physical activity questionnaire (metabolic equivalents/day) (23). To determine nutrient intakes, 5-d diet (three weekdays and a weekend) recalls were completed. Subjects' parents were taught how to complete diet recall questionnaires and determine food serving and sizes. Diet records were analyzed using the computerized nutritional analysis system Science Fit Diet 200A (Science Technologies, Athens, Greece) (24).

# vBMD and geometric properties estimation by pQCT

pQCT bone mineral measurements and analyses were performed at the left tibia using an XCT-3000 device (Stratec Gmbh, Pforzheim, Germany) as previously described (25). Participants were asked about dominance of hand, and the limb to be measured was the contralateral leg. All subjects were right-handed, so the left tibia was measured in all. A single-energy x-ray source was used, and all computed tomography scans had a slice thickness of 2.4 mm and a voxel size of 0.5 mm<sup>3</sup>. The distal end of the tibia was used as an anatomical marker; the bone cross-sectional area (CSA) was imaged at 10 mm proximal to the distal surface of the distal metaphysis and 14, 38, and 66% of the total tibia length. Analyzing each slice vBMD (milligrams/centimeter<sup>3</sup>), corresponding BMC (milligrams) and CSA (millimeters<sup>2</sup>) of tibia bone section were estimated, as well as cortical thickness (CRTHK; millimeters), endosteal and periosteal circumference (both in millimeters), and polar stress strength index in torsion (SSIp; millimeters<sup>3</sup>). Image analysis was performed using integrated software provided by Stratec Gmbh, version 5.4. Total (from the periosteum included area of the bone and bone marrow), trabecular, and cortical bone density in milligrams/centimeter<sup>3</sup> and the CSAs of the corresponding bone portions in millimeters<sup>2</sup> were calculated by the following procedure: 1) voxels outside the bone (soft tissue) with lower attenuation coefficients than the selected threshold (181 mg/cm<sup>3</sup>) are removed within the region of interest; and 2) the cortical and trabecular structures are separated by the areal distribution of both bone structures. By default, 55% of the outer bone area is concentrically separated and defined as cortical-subcortical region. The remaining 45% of the inner core is defined as trabecular bone. To calculate pure cortical density and area without including subcortical area, all voxels within the region of interest that have an attenuation coefficient below threshold 710 mg/cm<sup>3</sup> density are removed. CRTHK was defined as the mean distance between the inner and outer edges of the cortical shell. SSIp lies within the theory of stability of mechanical structures against bending or torsion. From computed tomography cross-sectional images, the determination of bone strength is based on the calculation of the cross-sectional moment of inertia. Division of the cross-sectional moment of inertia by the maximum distance of any voxel from the center of gravity (r<sub>max</sub>) yields the section modulus, which is directly proportional to maximum stress in bone. To also take the material properties into consideration, the section modulus

is multiplied with the quotient of calculated cortical density and normal cortical density of 1200 mg/cm<sup>3</sup>, yielding the calculation of SSIp. At the 66% slice, the muscle CSA was also calculated. The long-term *in vitro* (phantom) precision of the pQCT in 12-month daily measurements was 0.12% for total vBMD and 0.3% for trabecular vBMD. The *in vivo* precision derived from 25 postmenopausal women subjected to duplicate measurements within 1 month was: total vBMD, 0.2%; trabecular vBMD, 0.46%; cortical area, 0.3%; SSIp, 1%; and CRTHK, 0.8%.

### **Biochemical analysis**

Each subject reported to the laboratory for blood sampling after an overnight fast and 72-h abstinence from exercise. Serum total calcium (corrected to a serum albumin concentration of 4 g/liter) (26) and serum inorganic phosphate were measured by colorimetry using a Roche Hitachi 902 analyzer (Roche, Indianapolis, IN). The intra- and interassay CVs for calcium and phosphate determinations were 0.9 and 1.5%, and 0.9 and 1.4%, respectively. Plasma intact PTH (iPTH) was measured by an electrochemiluminescence immunoassay (ECLIA) (Roche). The sensitivity was 1.2 pg/ml, and the intra- and interassay CVs were 4 and 4.3%, respectively. Serum 25-hydroxyvitamin D [25(OH)D] was determined by EIA (OCTEIA; Immunodiagnostic Systems Ltd., Boldon, UK). The sensitivity was 5 nmol/liter, and the intra- and interassay CVs were 5.3 and 4.6%, respectively. Serum procollagen type 1 N-propeptide (P1NP) was measured by ECLIA (Roche). The sensitivity was 5  $\mu$ g/liter, and the intra- and interassay CVs were 2.2 and 2.9%, respectively. Serum β-C-telopeptide cross-linked collagen type I (sCTX) was determined by ECLIA (Roche). The sensitivity was 0.01 ng/ml, and the intra- and interassay CVs were 1.6 and 4.3%, respectively.

### Statistical analysis

Normality was tested using the Kolmogorov-Smirnov test. Accordingly, data are presented as mean  $\pm$  SE or median (range) in case of not normally distributed variables. Between-group differences were analyzed by unpaired t test or Mann-Whitney U test concerning continuous variables and  $\chi^2$  test for categorical variables, as appropriate. Mean differences of all vBMD and geometric parameters (adjusted for weight) between gymnasts and controls were assessed by analysis of covariance (4, 5, 27). Partial correlations were performed to estimate the association of vBMD and bone geometry indices with selected variables, independent of differences in weight.

All tests were two-tailed, and P < 0.05 was considered significant. All data analysis was performed using the Statistical Package for Social Sciences (version 10.0) software (SPSS Inc., Chicago, IL).

### **Results**

### **Baseline characteristics**

Baseline characteristics are presented in Table 1. Of the 60 children initially evaluated (30 athletes and 30 controls), four athletes and seven controls were excluded due to either unsatisfactory pQCT scan (movement artifacts, two athletes and four controls) or recent trauma that could affect BTM levels (two athletes and three controls). The

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**TABLE 1.** Physical and dietary characteristics of the two groups

	RG (n = 26)	Controls ( $n = 23$ )	P
Age (yr)	11.26 ± 0.17	10.87 ± 0.13	0.096
Weight (kg)	$31.07 \pm 0.69$	$40.34 \pm 1.71$	< 0.001
Height (cm)	$142.75 \pm 1.60$	145.76 ± 1.10	0.13
Height <sub>sps</sub>	-0.14(2.2)	0.30 (2.49)	0.085
Sitting height (cm)	$75.80 \pm 0.60$	$76.81 \pm 0.80$	0.33
BMI $(kg/m^2)$	$15.20 \pm 0.18$	$18.85 \pm 0.58$	< 0.001
BMI <sub>SDS</sub>	-1.10(2.44)	0.23 (3.02)	< 0.001
Skinfold sum (mm)	42.63 (32.55)	81.00 (183.00)	< 0.001
Bone age (yr)	$10.61 \pm 0.25$	$11.24 \pm 0.23$	0.079
Tanner stage <sup>a</sup>	I:10, II:13, III:3	I:7, II:12, III:4	0.767
Tibial length (cm)	$32.84 \pm 0.34$	$33.65 \pm 0.56$	0.22
Training age (yr)	$4.34 \pm 0.25$	NA	
Dietary calcium intake (mg/d)	$1004.16 \pm 62.97$	$916.83 \pm 60.72$	0.32
Dietary vitamin D intake (IU/d)	$147.48 \pm 7.92$	146.17 ± 12.14	0.92
Protein intake (mg/d)	$60.26 \pm 3.16$	$64.77 \pm 3.52$	0.34
Energy intake (KJ/d)	$5985.40 \pm 377.99$	$6943.75 \pm 439.15$	0.103
Physical activity (METs/d)	$46.8 \pm 1.36$	$34.4 \pm 1.20$	0.002

Data are presented as means  $\pm$  se or median (range) as appropriate. Training age, Number of years each participant participated in RG training and competition; NA, not applicable.

two groups were comparable according to chronological age, height, height SD score (height<sub>SDS</sub>), calcium, vitamin D daily intake, and recreational daily physical activity. There was a nonsignificant trend for lower bone age in RG. Athletes had significantly lower weight, BMI, BMI<sub>SDS</sub> and skinfold sum (P < 0.001). Thus, differences in bone density and geometry parameters were adjusted for weight.

#### **Biochemical characteristics**

Biochemical analysis revealed that RG had higher total calcium (9.85  $\pm$  0.08 vs. 9.56  $\pm$  0.09 mg/dl; P=0.025), whereas phosphate was comparable (Table 2). iPTH was lower in RG compared with controls (P=0.012). RG had higher 25(OH)D (102.22  $\pm$  4.20 vs. 87.46  $\pm$  3.74 nmol/liter; P=0.013). However, all subjects had levels higher than 50 nmol/liter, with five controls and two RG having levels between 50.16 and 74.64 nmol/liter. P1NP and sCTX were comparable.

**TABLE 2.** Biochemical characteristics of the study group

RG	Control	P
$9.85 \pm 0.08$	9.56 ± 0.09	0.025
$5.41 \pm 0.11$	$5.19 \pm 0.10$	0.17
$36.01 \pm 1.85$	$45.34 \pm 3.14$	0.012
$102.22 \pm 4.2$	$87.46 \pm 3.74$	0.013
$684.62 \pm 49.49$	7 00.00 = .5	0.133
$1.65 \pm 0.07$	$1.77 \pm 0.08$	0.320
	$9.85 \pm 0.08$ $5.41 \pm 0.11$ $36.01 \pm 1.85$ $102.22 \pm 4.2$	9.85 $\pm$ 0.089.56 $\pm$ 0.095.41 $\pm$ 0.115.19 $\pm$ 0.1036.01 $\pm$ 1.8545.34 $\pm$ 3.14102.22 $\pm$ 4.287.46 $\pm$ 3.74684.62 $\pm$ 49.49785.88 $\pm$ 43.1

Data are presented as means  $\pm$  se. NR, Normal range.

# Comparison of vBMD and bone geometry between groups by pQCT

# Trabecular site (10 mm proximal to the distal surface of the distal metaphysis of the tibia)

At this site, trabecular BMC, area, and vBMD were comparable between groups (Table 3). Total BMC was higher in RG (P = 0.022), whereas total area and vBMD were comparable.

#### Cortical site (38% of the tibia length)

At 38% site, total and cortical BMC and area were significantly higher in RG compared with controls (all  $P \le 0.001$ ). Indeed cortical BMC was 30.26% higher, whereas cortical area was increased by 30.07%. Cortical vBMD was comparable. Concerning geometric indices, RG demonstrated higher CRTHK (25.83%; P < 0.001), which was due to an increase in periosteal circumference (7.70%; P = 0.001), whereas endosteal circumference was comparable, indicating that intensive RG is associated with increased periosteal apposition. Moreover, SSIp, an index of bone strength, was increased (31.53%; P < 0.001), indicating a positive response to applied loads.

### Transition zone (14% of the tibia length)

At the 14% site, total, trabecular, and cortical BMC and area were increased in RG compared with controls (P < 0.05-0.001), whereas the corresponding vBMDs were comparable. SSIp was increased in RG (18.98%, P = 0.002). Muscle area was 11.13% higher in RG compared with controls (P = 0.002).

<sup>&</sup>lt;sup>a</sup> Tanner stage of breast development.

**TABLE 3.** vBMD, BMC, and bone geometric characteristics assessed by pQCT

	RG	Control	P
10 mm proximal to the distal surface of the distal metaphysis			
Total BMC (mg)	$251.10 \pm 10.43$	$210.96 \pm 11.24$	0.022
Total vBMD (mg/cm <sup>3</sup> )	$301.45 \pm 6.58$	$283.75 \pm 7.09$	0.105
Total CSA (mm <sup>2</sup> )	$836.53 \pm 31.18$	$741.97 \pm 33.61$	0.069
Trabecular BMC (mg)	$92.92 \pm 6.21$	$73.24 \pm 6.70$	0.058
Trabecular vBMD (mg/cm³)	$243.61 \pm 9.7$	$216.05 \pm 10.46$	0.088
Trabecular CSA (mm <sup>2</sup> )	$376.31 \pm 14.03$	$333.75 \pm 15.13$	0.069
38% site			
Total BMC (mg)	$272.73 \pm 5.47$	$218.19 \pm 5.90$	< 0.001
Total vBMD (mg/cm <sup>3</sup> )	$789.76 \pm 12.47$	$732.55 \pm 13.45$	0.007
Total CSA (mm <sup>2</sup> )	$346.87 \pm 8.04$	$300.34 \pm 8.67$	0.001
Cortical BMC (mg)	$243.82 \pm 5.45$	$187.17 \pm 5.88$	< 0.001
Cortical vBMD (mg/cm³)	$1043.70 \pm 8.28$	$1042.31 \pm 8.93$	0.919
Cortical CSA (mm <sup>2</sup> )	$233.85 \pm 5.28$	$179.78 \pm 5.70$	< 0.001
CRTHK (mm)	$4.53 \pm 0.09$	$3.60 \pm 0.10$	< 0.001
PERI (mm)	$65.96 \pm 0.80$	$61.22 \pm 0.86$	0.001
ENDO (mm)	$37.48 \pm 0.91$	$38.59 \pm 0.98$	0.461
SSIp (mm <sup>3</sup> )	$1129.74 \pm 38.01$	$858.92 \pm 40.98$	< 0.001
14% site			
Total BMC (mg)	$187.77 \pm 3.58$	$162.27 \pm 3.86$	< 0.001
Total vBMD (mg/cm <sup>3</sup> )	$471.99 \pm 10.71$	$449.81 \pm 11.55$	0.210
Total CSA (mm²)	$402.70 \pm 11.01$	$362.94 \pm 11.87$	0.032
Trabecular BMC (mg)	$29.30 \pm 2.13$	$20.96 \pm 2.30$	0.021
Trabecular vBMD (mg/cm³)	$164.88 \pm 12.75$	129.12 ± 13.75	0.092
Trabecular CSA (mm²)	$181.07 \pm 4.95$	$163.17 \pm 5.34$	0.032
Cortical BMC (mg)	$123.64 \pm 2.98$	$104.79 \pm 3.21$	< 0.001
Cortical vBMD (mg/cm <sup>3</sup> )	$984.79 \pm 7.69$	$971.57 \pm 8.29$	0.296
Cortical CSA (mm <sup>2</sup> )	$125.63 \pm 2.77$	$107.71 \pm 2.99$	< 0.001
SSIp (mm³)	$964.57 \pm 31.70$	$810.68 \pm 34.18$	0.005
66% site			
Muscle area (mm²)	4551.06 ± 85.76	4094.89 ± 92.46	0.002

Data are presented as means  $\pm$  se. All data are adjusted for differences in weight. PERI, Periosteal circumference; ENDO, endocortical circumference.

#### Correlation analyses

Partial correlation analysis, adjusted for weight, revealed modest positive association of age, height, and muscle area with cortical area ( $r_{age} = 0.51$ ,  $r_{height} = 0.63$ ,  $r_{muscle area} =$ 0.46; P < 0.001 - 0.01), trabecular area ( $r_{age} = 0.33, r_{height} = 0.001 - 0.01$ ) 0.41,  $r_{\text{muscle area}} = 0.36$ ; P < 0.01-0.05), periosteal circumference ( $r_{age} = 0.51$ ,  $r_{height} = 0.59$ ,  $r_{muscle\ area} = 0.45$ ; P < 0.001-0.01), and SSIp ( $r_{age} = 0.47$ ,  $r_{height} = 0.57$ ,  $r_{\text{muscle area}} = 0.48$ ; P < 0.001-0.01), whereas skinfold sum, an index of adiposity, showed modest negative association. Furthermore, at cortical sites training age, assessed only in RG, had positive association with area (r = 0.58; P <0.01), BMC (r = 0.53; P < 0.01), and CRTHK (r = 0.48; P <0.05), which remained significant even after adjustment for chronological age or bone age ( $r_{area} = 0.51$ ,  $r_{BMC} = 0.44$ ,  $r_{CRTHK} = 0.47$ ; P < 0.05-0.01; and  $r_{area} = 0.59$ ,  $r_{BMC} =$ 0.56,  $r_{CRTHK} = 0.52$ ; P < 0.05 - 0.01, respectively).

Correlation of BTMs with bone strength indices gives an insight of the underlying mechanism of bone adaptation to applied loads. Thus, sCTX, an index of bone resorption, had modest negative association with CRTHK (r = -0.48; P < 0.001) and positive association with endocortical circumference (r = 0.43; P < 0.01). On the con-

trary, P1NP levels did not show any particular association with bone strength indices, apart from a marginal positive association with trabecular CSA (r = 0.33; P < 0.05).

#### **Discussion**

In this study, we evaluated the effect of elite RG on vBMD and bone geometry assessed by pQCT at the tibia in premenarcheal athletes. The major findings were: 1) RG was associated with a positive bone adaptation especially at cortical sites, characterized by an increase in the degree of mineralization and area, whereas vBMD remained unchanged; 2) geometric parameters such as CRTHK, periosteal circumference, and ultimately bone strength were increased, providing evidence that intensive long-term RG in the growing years might result in bone health benefits; and 3) muscle mass was associated with a positive response of both quantitative and qualitative bone parameters, whereas training age showed similar association, possibly independent of chronological age.

The effect of weight-bearing exercise on vBMD and bone geometry has been evaluated in a relatively small number of studies, which differ in terms of methodology (e.g. DXA with or without hip structural analysis, quantitative computed tomography, magnetic resonance imaging, or pQCT), region of evaluation (humerus, radius, femur, tibia, or spine), type (cross-sectional using inactive subjects as controls, cross-sectional examining side-toside differences and prospective), and more importantly study population differing in terms of age, sex, race, pubertal maturation, and type and intensity of exercise (28). Limited data to date suggest that in prepubertal years the shafts of the long bones respond to increased loading by increasing periosteal apposition, whereas distal sites increase tissue density rather than bone size (28). However, to our knowledge, no study has examined the effect of long-term elite RG on bone geometry using pQCT at the tibia, along with detailed evaluation of maturational stages, dietary habits, and bone turnover.

In the present study, we observed the most prominent effects of RG at cortical sites, subject to bending and torsional forces. At the respective 38% of the tibia, both area and BMC were higher by about 30% compared with controls, whereas cortical vBMD was comparable. This difference was due to the deposition of bone on the periosteal surface, leading to significant increase in bone strength assessed by SSIp. *Post hoc* power analysis indicated that our study (n = 49) had at least 90% power to detect a 30% between-group difference in SSIp, with a significance level of 0.05.

In contrast, the data concerning the effect of RG at trabecular sites subject to axial and compressive forces indicate that bone appears to increase BMC rather than its size, leading to a relatively higher tissue density. Indeed, at the distal tibia and at 14%, trabecular BMC was higher by 26.80 and 39.80%, respectively, whereas the corresponding area was higher by 12.70% and 10.90%, leading to a trend for higher trabecular vBMD. These findings are in agreement with Ward et al. (9), who reported a trend for higher trabecular vBMD at this site. Taking into account the inherent limitation of pQCT for estimating trabecular architecture due to low resolution, it is possible that more sensitive methods such as magnetic resonance would provide more information concerning the effect of weightbearing exercise on trabecular bone. Nevertheless, it appears that at distal skeletal sites, weight-bearing exercise increases tissue density to more efficiently transmit loads through the joint surface.

The effect of duration and intensity of weight-bearing exercise on the osteogenic response in prepubertal children has been assessed in a number of prospective controlled trials (3). Given the limitations of the cross-sectional design, the positive association of training age with bone geometry and BMC at cortical sites, independent of

chronological or bone age, indicates that elite RG is associated with a continuous positive response at least during premenarcheal years. Furthermore, Frost's "mechanostat" theory (29) suggests that bone can become accustomed to constant loading of similar magnitude, and no osteogenic response will be elicited until a higher magnitude load is applied. Given that older gymnasts had higher weight, foot contacts per year based on training logs increased with years of training, whereas type of exercise and thus generated ground reaction forces expressed in times body weight remained the same, the association of training years with the positive bone adaptation is in accordance with Frost's theory.

Several studies have assessed the association of muscle mass, a surrogate of muscle force, with bone strength indices at the tibia and radius (20, 30, 31). Our data are almost in complete accordance with Macdonald et al. (20), who found strong association of muscle area with bone area and strength at the distal tibia and the midshaft in a large group of early pubertal and prepubertal children. Furthermore, Daly et al. (31), who examined side-to-side differences in the playing and nonplaying arm in female tennis players using magnetic resonance imaging found that percentage differences in muscle area were positively associated with differences in all bone traits, except in the medullary area. However, as reported by Macdonald et al. (20), the variance in bone traits explained by differences in muscle area was less than 16%, indicating that other factors associated with loading contribute to the skeletal adaptive response.

To our knowledge, this is the first study examining bone strength indices in athletes along with a detailed evaluation of calcium homeostasis and BTMs. Our data are consistent with findings of previous studies in children that obesity is associated with lower 25(OH)D levels (32). Although all subjects had vitamin D levels higher than 50 nmol/liter, this difference was associated with higher iPTH and lower calcium levels in controls and was completely abolished after adjustment for weight. Considering the fact that between-group vitamin D intake and daytime physical activity were comparable, the only possible explanation might be related to less sunlight exposure due to reduced mobility or clothing habits, reduced production of vitamin D after sun exposure, and reduced mobilization of vitamin D from fat depots. Concerning bone markers, our data indicate that long-term elite training in premenarcheal females does not significantly modify BTMs. This is in line with studies (7, 33) in this age group that did not find differences in BTMs between female gymnasts and controls. However, given the relatively small number of subjects studied, additional work is needed to settle this issue.

The long-term significance of intensive exercise-induced skeletal adaptations for fracture reduction remains uncertain because several studies indicate a higher rate of aBMD loss after detraining (1, 34). Nevertheless, structural changes, such as increased CRTHK, might be retained even after detraining. Indeed, a recent study (35) found greater BMC, CSA, CRTHK, and SSIp at the tibia in female gymnasts 6 yr after retirement. However, given that bone response to exercise is site-specific, the results cannot be applied to other skeletal sites, including typical fracture sites (hips, vertebrae, forearms). Furthermore, because waning of the positive effects of exercise on bone might be expected over time, the long-term effect on fracture risk is difficult to assess, even if the observed bone response could be generalized to typical fracture sites.

There are limitations to this study. First, the cross-sectional design is limiting, and long-term longitudinal studies are needed to assess whether the observed positive effects are maintained or further improved with or without sustained activity. Furthermore, we did not perform power analysis; however, *post hoc* power analysis indicated that our study had at least 80% power to detect adjusted differences between groups concerning important pQCT variables. Moreover, although athletes and controls were comparable according to chronological age, height, and bone age, we failed to match our subjects by weight and BMI; thus, we adjusted bone strength indices according to weight. Finally, our data concern only elite RG and should not be extrapolated to other sports or to normally active children.

In conclusion, our findings indicate that intensive RG in premenarcheal girls is associated with positive effects on the skeleton, especially in cortical bone, characterized by increased bone mass and improved geometric properties. Increased duration of exercise is associated with a positive response of bone geometry.

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