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Mechanical Loading during Growth Is Associated with Plane-specific Differences in Vertebral Geometry: A Cross-sectional Analysis Comparing Artistic Gymnasts vs. Non-gymnasts

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Abstract

Lumbar spine geometry, density and indices of bone strength were assessed relative to menarche status, using artistic gymnastics exposure during growth as a model of mechanical loading. Paired posteroanterior (PA) and supine lateral (LAT) DXA scans of L3 for 114 females (60 ex/gymnasts and 54 non-gymnasts) yielded output for comparison of paired (PALAT) versus standard PA and LAT outcomes. BMC, areal BMD, vertebral body dimensions, bone mineral apparent density (BMAD), axial compressive strength (IBS) and a fracture risk index were evaluated, modeling vertebral body geometry as an ellipsoid cylinder. Two-factor ANCOVA tested statistical effects of gymnastic exposure, menarche status and their interaction, adjusting for age and height as appropriate. Compared to non-gymnasts, ex/gymnasts exhibited greater PABMD, PABMC, PAWIDTH, PA CROSS-SECTIONAL AREA (CSA), PAVOLUME, LATBMD, LATBMAD, PALATCSA and PALATIBS ($p < 0.05$). Non-gymnasts exhibited greater LATDEPTH/PAWIDTH, LATBMC/PABMC, LATVHEIGHT, LATAREA and Fracture Risk Index. Using ellipsoid vertebral geometric models, no significant differences were detected for PA or PALAT BMAD. In contrast, cuboid model results (Carter 1992) suggested erroneous ex/gymnast PABMAD advantages, resulting from invalid assumptions of proportional variation in linear skeletal dimensions. Gymnastic exposure was associated with shorter, wider vertebral bodies, yielding greater axial compressive strength and lower fracture risk, despite no BMAD advantage. Our results suggest the importance of plane-specific vertebral geometric adaptation to mechanical loading during growth. Paired scan output provides a more accurate assessment of this adaptation than PA or LAT plane scans alone.

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Keywords

DXA; mechanical loading; exercise; gymnastics; pediatric; bone strength

Introduction

A large body of literature documents greater lumbar spine areal bone mineral density (BMD) and bone mineral content (BMC) in artistic gymnasts and ex-gymnasts compared to non-gymnasts, as measured by postero-anterior dual energy x-ray absorptiometry (PA DXA) [1–11]. As such, artistic gymnastics provides a convenient extreme model for the evaluation of mechanical loading during human growth. Several gymnastics studies have evaluated lumbar spine bone mineral apparent density (BMAD) in an effort to account for bone size variability and improve evaluation of bone volumetric density [1,7,8,12–18]. However, few gymnastics studies have investigated skeletal geometric indices at the lumbar spine [1,12,13,18].

Numerous problems affect PA DXA assessment of the lumbar spine. First, PA scans integrate vertebral body BMC (primarily trabecular tissue) with BMC of the posterior elements (primarily cortical tissue) [19–21]. This merger contaminates efforts to evaluate trabecular tissue properties that are widely believed to be most relevant to osteoporotic fracture risk. Second, PA scans assess vertebral projected area (a function of width and height), but they fail to account for vertebral depth [19,22]. Third, PA DXA of the lumbar spine is influenced by fan beam magnification error (FBME), which varies systematically with tissue distance from the X-ray source [19,23,24]. FBME is particularly problematic in comparisons of individuals of changing or disparate body sizes, thereby posing a major concern in pediatric and longitudinal studies [19,20,23,24].

Fortunately, DXA skeletal outcomes are improved by pairing PA scans with supine lateral scans (LAT) [19]. LAT output isolates vertebral body BMC from posterior element BMC [20]. LAT scans also alleviate the confounding effects of FBME by centering the vertebral column to yield uniform distances between regions of interest and the X-ray source [19]. Critically, paired PA and LAT scans (PALAT) measure all three linear bone dimensions: vertebral width (PAWIDTH), depth (LATDEPTH) and height (LATVHEIGHT) (Figure 1). Thus, modeling vertebrae as ellipsoid cylinders, PALAT output allows for derivation of more accurate indices of bone geometry (vertebral volume and cross-sectional area) and total volumetric BMD (PALATBMAD) than those derived from PA or LAT scans alone [19,25](Figure 1). Finally, indices of bone strength can be derived from PALAT output, including an index of bone structural strength in axial compression (IBS) [26,27] and a modified version of a “fracture risk index” that has been associated with vertebral fracture risk in males and females (FRI) [28]. In outcomes derived from paired scans, lateral scan output is emphasized, reducing the influence of FBME for all but PAWIDTH.

To our knowledge, no studies have exploited the advantages of paired PA and LAT DXA to evaluate areal BMD, BMC, PALATBMAD, or any other geometric/volumetric outcomes in relation to models of human mechanical loading. It is clinically relevant to establish variability in plane-specific bone geometric parameters and volumetric BMD because: 1) bone cross-sectional area (CSA) and volumetric BMD determine axial compressive strength; 2) vertebral body height contributes to stresses generated during forward bending [28]. Therefore, it was the objective of the present study to evaluate paired PA and LAT lumbar spine data in relation to gymnastic loading exposure, comparing outcomes by scan plane. Using a mixed cohort of pre-menarcheal and post-menarcheal females, we evaluated indices of skeletal geometry and axial compressive strength in relation to mechanical loading,

accounting for potential interactions with physical maturity. We aimed to test the hypothesis that gymnastics exposure during growth yields plane-specific advantages in vertebral body volumetric density, geometry and theoretical axial compressive strength.

Material and Methods

The protocol was approved by the SUNY Upstate Medical University Institutional Review Board, and research was performed in accordance with the Declaration of Helsinki. All subjects provided written consent, or assent with parental consent, as appropriate for subject age. Paired PA and supine lateral DXA scans of the lumbar spine were obtained in mature (post-menarcheal, POST) and immature (pre-menarcheal, PRE) females as part of an ongoing longitudinal study of bone growth in relation to artistic gymnastic exposure. Pre-menarcheal gymnasts (PRE EX/GYM, n=35) included all girls who had participated in organized artistic gymnastics activity for an annual mean of greater than 6 hours per week (h/wk) during the year prior to the focal DXA scans. Post-menarcheal ex/gymnasts (POST EX/GYM, n=25) had participated in at least 6 h/wk of gymnastics for at least one year during growth. This exposure threshold has been associated with significant advantages in forearm bone parameters for ex/gymnasts versus non-gymnasts in our study population [29,30,31,32]. Non-gymnasts (PRE n=33, POST n=21) were not necessarily sedentary controls; most were multi-sport athletes who simply had not exceeded this gymnastic training threshold during growth (≥ 6 h/wk annual mean). Accordingly, these analyses provide conservative estimates of skeletal advantages attributable to gymnastic loading.

During growth, measurement and questionnaire sessions were performed semi-annually; beyond age 18, measurement and questionnaire sessions were held annually, coincident with scans. Height, weight and waist circumference were measured using standard protocols. Questionnaires were administered to record monthly means of organized physical activity (yielding annual mean h/wk), calcium intake (food frequency questionnaire) and physical maturity information (including date of menarche, menstrual function and self-assessed Tanner breast and pubic stages) [10]. Questionnaire administration was complemented by subject/parent interview, as appropriate. With reference to physical activity records, a similar methodology for recording gymnastic activity has been validated by our group through comparison with coaching staff logs (correlations ranged from $R=0.93$ for 1 month records to $R=0.97$ for annual means, $p<0.0001$, unpublished data).

At annual scan sessions, a 60 second sit-up test was used as an index of axial muscular function and endurance. Total body DXA scans (Hologic Discovery A, software v. 12.7.3.2:3) provided body composition output (total body non-bone lean mass, nbFFM; percent body fat). Paired PA and supine lateral DXA scans were performed using a standard protocol, yielding areal bone mineral density (BMD), projected area (AREA) and bone mineral content (BMC) for both PA and LAT scans, as well as LATHEIGHT (mean vertebral height). All scans were analyzed by a single investigator, and non-standard variables were calculated from standard computer output. Two subjects were excluded from analyses due to excessive PA VS. LAT geometric deviation, attributed to vertebral torsion. Only data from the third lumbar vertebra (L3) were included in analyses, as lateral scans may be limited by inclusion of rib and iliac crest overlap within the region of interest for L2 and L4 [19,20,25,33]. This allows direct comparison with most in vivo and ex vivo studies, which include or specifically evaluate L3 [20,21,28,34,35].

To assess variability of plane-specific bone geometry, volumetric density and strength indices, we evaluated paired scan output, including mean vertebral width (PAWIDTH= PAAREA/LATVHEIGHT), mean vertebral height (LATVHEIGHT) and mean vertebral depth (LATDEPTH= LATAREA/LATVHEIGHT). The ratio of LATBMC/PABMC was

calculated to assess the relative proportion of bone mineral content in the vertebral bodies versus the posterior elements [19,21]. Similarly, the ratio of LATDEPTH/PAWIDTH was calculated to evaluate the relationship between vertebral depth and width. Additional indices of vertebral geometry, volumetric density and axial compressive strength were derived using formulae based upon simplified geometric models (Figure 1, Table 1)[19,25–28]. Outcomes included PA, LAT and PALAT indices of vertebral body cross-sectional area (CSA), volume, BMAD and IBS. For PAVOLUME and PABMAD, Carter's equations based on cuboid geometric assumptions [27] were also applied for comparison of our results to those of previous PA DXA studies (PAVOLUME_C and PABMAD_C, Table 1).

In exploratory analyses, it was evident that for software version 12.7.3.2:3, Hologic's standard width-adjusted BMD (WABMD) and width-adjusted volume (WAVOLUME) did not correspond with outcomes derived using paired modalities and the formulae reported by Leonard et al. (PALATBMAD, PALATVOLUME) [19]. Rather, Hologic WABMD output corresponded exactly with LATBMAD, derived solely from lateral scan outcomes (LATDEPTH, LATBMC, LATVHEIGHT). Thus, WABMD (Hologic Discovery A, software version 12.7.3.2:3) does not incorporate PAWIDTH in its assessment of volumetric density. Similarly, WAVOLUME did not correspond to our calculated PALATVOLUME (based on PAWIDTH, LATDEPTH, LATVHEIGHT). Because it was unclear how WAVOLUME is derived (values were intermediate between PALATVOLUME and LATVOLUME), these data were not analyzed. Accordingly, calculated PALATVOLUME and PALATBMAD were substituted for Hologic WAVOLUME and WABMD in our analyses.

To evaluate associations between indices of bone geometry, axial compressive strength and fracture risk, we generated a fracture risk index (FRI) based upon equations reported by Duan et al. [28]. To improve evaluation of three dimensional, total L3 vertebral body characteristics: 1) we substituted "total" L3 PALATCSA and PALATBMAD for lateral measures of the "middle 1/3"; 2) we substituted LATDEPTH calculated from Hologic output for hand-measured "extensor moment arm". The first modification evaluates bone geometry in all three dimensions, as opposed to 2-dimensional lateral depth and height. The second modification assumes that computer-measured vertebral depth varies proportionally with actual 3-dimensional extensor moment arm (including out of plane depth). Because hand-measured extensor moment arm introduces human error and does not account for out of plane depth, we believe that this modification improves upon the original method of Duan et al. [28].

Our group has previously shown that waist circumference is linearly related to the distance from the center of the vertebral body to the DXA scanner tabletop ($r^2 = 0.94$, $p < 0.0001$); this distance is in turn related to fan beam magnification error (FBME) [24]. We used this information to apply a correction for FBME to PABMC, PAAREA and PABMD, as well as all variables derived from this output [24]. Supplemental ANCOVAs evaluated FBME-corrected data to account for this distortion.

Although no studies have reported differences between gymnasts and non-gymnasts for supine lateral lumbar spine DXA parameters, three studies have reported PABMAD differences in Tanner I/II gymnasts vs. non-gymnasts [1,12,13]. Based upon results from these studies, cell sizes of $n=25$ to 37 would be required to achieve 80% power to detect a significant difference based on gymnastic status. Therefore, adequate power should be provided by our sample of EX/GYM (PRE $n=35$, POST $n=25$), and NON (PRE $n=33$, POST $n=21$).

Data for many variables deviated from a normal distribution; thus, all data were converted to natural logarithms for analysis and back-transformed for presentation of results. Two factor analysis of variance (ANOVA) was used to evaluate EX/GYM vs. NON characteristics within the PRE and POST subgroups. Alpha level was set at $p < 0.05$. For the total sample, two-factor ANCOVA was used to evaluate differences related to gymnastic exposure, menarche status and the interaction between these factors, adjusting for chronological age and height as covariates. Exceptions included fracture risk index (adjusted only for age, due to inclusion of height in the formula) and ratios of LATBMC/PABMC and LATDEPTH/PAWIDTH (ANOVA without adjustment, to test for differences in raw ratios). In order to account for potential interactions between menarche status and gymnastic exposure, interaction terms were kept in the model, despite non-significance. Accordingly, reported EX/GYM vs. NON differences account for the main effect of menarche status, the interaction between menarche status and gymnastic exposure, and the effects of age and/or height (as noted above).

Results

Subject Characteristics

For total sample subject characteristics (Table 2), no significant differences were detected between EX/GYM and NON except that EX/GYM did significantly more sit-ups in one minute, exhibited significantly lower percentage body fat and reported significantly higher total non-aquatic physical activity levels over the year prior to the DXA scan than NON ($p < 0.001$). There was a strong trend toward greater height and waist circumference among NON vs. EX/GYM ($p \leq 0.06$). For subject characteristics, the only significant interaction observed between gymnastic exposure and menarche status was for physical activity level (PRE EX/GYM > PRE NON; POST EX/GYM VS. POST NON= NS; PRE EX/GYM > POST EX/GYM; PRE NON VS. POST NON= NS). As would be expected, for age and all anthropometric variables, POST exhibited greater means than PRE. However, on average, PRE exhibited significantly higher mean physical activity levels and calcium intakes than POST ($p < 0.05$). In the POST subsample, significant differences were not detected between EX/GYM and NON for age at menarche or gynecological age (years post-menarche).

On average, PRE EX/GYM had participated in organized gymnastics at a level of greater than 2 h/wk for approximately 4.1 years (sd=2.3, 1.5 yrs to 11.1 yrs). Their current annual mean gymnastic exposure ranged from 6.0 h/wk to 25.0 h/wk. For the year prior to the DXA scan, total annual organized non-aquatic physical activity (including gymnastics) was significantly higher in PRE EX/GYM (annual mean: 6.4 h/wk to 25.0 h/wk) versus PRE NON (0.2 h/wk to 10.6 h/wk)(Table 2).

In contrast, POST EX/GYM exhibited similar levels of physical activity to POST NON; for the majority, this was a function of increasing physical activity in NON and decreasing physical activity among EX/GYM over the previous 8 to 12 years. Gymnastic exposure was remote among most POST EX/GYM, such that only 20% participated in gymnastics within 2 months of the focal DXA scan ($n=5$), and 32% had discontinued gymnastic activity prior to menarche, at an average gynecological age of -1.9 years ($n=8$, sd= 2.2, -6.0 to -0.3 years). The remaining 48% were post-menarcheal “quitters”, with a mean gynecological age at gymnastics cessation of $+2.7$ years post-menarche ($n=12$, sd= 1.7, $+0.0$ to $+5.8$ years). Thus, on average, POST ex-gymnasts discontinued gymnastics at 0.9 years post-menarche ($n=20$, sd= 3.0, -6.0 to $+5.8$ years), 5.6 years prior to the focal DXA scan ($n=20$, sd= 3.5, 0.5 to 10.4). Although exposure was remote for some, most POST EX/GYM achieved a moderately high peak level of gymnastic training during growth. Peak training level was defined as the highest number of hours per week of gymnastic activity maintained for an uninterrupted period of at least 6 months during the longitudinal study period (>6 years of

pre- and post-menarcheal physical activity represented). As such, for POST EX/GYM, mean peak training level was 16.3 h/wk (sd= 5.2, 6 h/wk to 25 h/wk, including current gymnasts).

Bone Outcomes

For bone outcomes, two-factor ANCOVA and ANOVA detected no significant interaction between gymnastic exposure & physical maturity for any variable. As would be expected, after adjustment for age and height, POST exhibited significantly greater means than PRE for most variables (Table 3). Exceptions included PABMAD, LATBMAD, PALATBMAD, PALATIBS, FRI and LATVHEIGHT; LATVHEIGHT was significantly greater in POST than PRE only if the effects of age and standing height were not considered (ANOVA).

After adjustment for age and height, EX/GYM exhibited significantly greater PABMC, PABMD, PAWIDTH, PAIBS, PACSA and PAVOLUME than NON (Figures 2 & 3, Table 3). For lateral outcomes, EX/GYM only exhibited significantly greater adjusted means for LATBMD, LATBMAD and LATIBS than NON (Figures 2 & 3, Table 3). For paired output, EX/GYM exhibited significantly greater adjusted means for PALATCSA than NON, with an extremely strong trend for greater PALATIBS ($p=0.05$). NON exhibited significantly greater adjusted means than EX/GYM for LATAREA, LATVHEIGHT, LATBMC/PABMC and LATDEPTH/PAWIDTH. Similar results were observed after adjustment for age and height as non-linear terms (quadratic), except that EX/GYM advantages for PALATIBS were statistically significant ($p=0.04$). When Carter's cuboid methods of calculating PAVOLUME_C and PABMAD_C were used [27], EX/GYM did not exhibit larger PAVOLUME_C but did exhibit greater PABMAD_C than NON ($p<0.05$) (data not shown). For evaluation of fracture risk index (FRI), ANCOVA adjusted only for age. NON exhibited significantly higher fracture risk than EX/GYM (Table 2, Figure 3). Nonetheless, as would be expected, no subject exhibited an FRI greater than 1 (indicative of high fracture risk). Similar results were observed after adjustment for age and height as non-linear terms (quadratic).

The ranges and means of waist circumferences in this study were similar to those used in a previously published derivation of a FBME correction factor (46.8–85.8cm; only 3/114 values fell outside the range: 88.7cm, 96.7cm, 100.5cm)[24]. Accordingly, the waist circumference-based FBME correction was applied to data for PAAREA, PABMC and PABMD [24], allowing for evaluation of all derived variables using FBME-corrected data. FBME-corrected ANOVA and ANCOVA results were virtually identical to results based on uncorrected data, except that PALATIBS was significantly greater in EX/GYM than NON, and trends toward interactions between gymnastics exposure and maturity became stronger for all forms of BMAD and IBS.

Discussion

We identified significantly higher areal BMD, greater indices of structural strength in axial compression (IBS) and lower fracture risk index for EX/GYM compared to NON. EX/GYM exhibited wider vertebrae in the medial-lateral plane and greater total cross-sectional area. In contrast, NON had taller vertebrae than EX/GYM, even after accounting for stature and age effects. The overall difference in vertebral shape was reflected by distinct ratios of LATDEPTH/PAWIDTH, which had profound effects on BMAD, as observed in different planes and by different methods. EX/GYM vs. NON comparisons for the ratio LATBMC/PABMC confirmed suspicions that PABMD advantages attributable to gymnastic loading actually reflect posterior element cortical bone adaptation. These results demonstrated the superiority of lateral BMC over PA BMC as a specific index of vertebral body adaptation (confounding posterior elements are excluded). However, we found that EX/GYM lateral areal BMD advantages reflect enlarged vertebral body width rather than an advantage in

volumetric density. Overall, our results emphasized the importance of informed two-plane imaging for assessment of adaptation in response to mechanical loading at the lumbar spine.

BMAD calculations were originally developed to compensate for the two-dimensional nature of areal BMD, generating an index of total bone volumetric density by estimating out-of-plane bone depth [27]. Although BMAD aims to “remove” the effects of bone size variation, our results demonstrate that it actually compounds the effects of disparate bone dimensions. For PABMAD, greater EX/GYM PAWIDTH is incorporated into the denominator twice, once as PAWIDTH and once as estimated out-of-plane bone depth. This artificially inflates EX/GYM relative bone volume. Thus, despite inclusion of greater BMC attributable to the posterior elements, in our cohort, EX/GYM did not demonstrate elevated PABMAD using an ellipsoid model. Conversely, in the lateral view, sole reliance upon LATDEPTH to calculate bone cross-sectional area fails to account for the EX/GYM PAWIDTH advantage (out-of-plane, not detected). Thus, EX/GYM LATBMC is inappropriately attributed to a deflated bone volume, inflating EX/GYM LATBMAD to yield an erroneously significant advantage. When all three linear dimensions are measured via paired scans (PALATBMAD), there is no volumetric density advantage associated with gymnastic exposure.

Our results for PABMAD are in direct contrast with those of numerous other studies that report significant gymnast and ex-gymnast advantages in lumbar spine PABMAD (7–12%) [1,12–14,18]. In each case, these investigators utilized the cuboid formula for PABMAD_C calculations [27], rather than the ellipsoid cylinder formula used in our calculations [19]. When we applied the cuboid formula to our data, we obtained an EX/GYM PABMAD_C advantage similar to prior reports (7.2%). As both cuboid and ellipsoid PABMAD formulae use PABMC, they differ only in their calculations of vertebral volume (Table 1). The cuboid formula assumes equal projected area in all three dimensions [27]. However, our results demonstrate that vertebral dimensions are not equal in all planes, nor do they vary proportionally or exhibit consistent relationships within and between individuals. The inaccuracy of this assumption is likely the chief source of the discrepancy between cuboid and ellipsoid calculations. In particular, the cuboid approach incorporates any difference in vertebral height as though it applies to all three dimensions. Even though EX/GYM exhibit an advantage in PAWIDTH that is also applied to all three dimensions, this raw advantage is smaller than the raw height deficit, so the net effect of the cuboid PABMAD_C formula is to deflate gymnast volume and inflate gymnast BMAD. In contrast, our ellipsoid PABMAD formula accounts for vertebral height (LATVHEIGHT); its chief inaccuracy is the assumption that vertebral width and depth are identical, inflating volume and comparatively deflating PABMAD. PALATBMAD rectifies these shortcomings and improves outcomes further by isolating vertebral body BMC from the posterior elements. It is unlikely that these bone geometric issues are uniquely influential in our study, as gymnasts frequently exhibit shorter sitting heights than non-gymnasts [1,36]. Thus, for any previously reported gymnast vs. non-gymnast comparisons, cuboid PABMAD_C results are likely to be invalid.

Numerous cadaver studies indicate that indices of BMAD and volumetric BMD provide incomplete assessments of theoretical axial compressive strength and fracture risk when isolated from bone geometry [34,37,38]. Roux and colleagues demonstrated that lateral areal BMD is superior to lateral BMAD for prediction of L3 trabecular thickness, cortical thickness, cortical curvature, failure load and work to failure [34]. Edmondston and colleagues reported that their most effective model for L1–L5 failure load incorporated DXA lateral areal BMD, QCT volumetric BMD and QCT vertebral CSA, explaining over 80% of the variance, with each variable contributing significant predictive value [38]. Cheng et al. reported that DXA areal BMD and CT IBS (trabecular volumetric BMD × CSA) yielded higher predictive value for L3 ultimate failure load than trabecular vBMD alone

[37]. Furthermore, for in vivo comparisons of subjects with vertebral fractures versus unaffected subjects, low vertebral width was associated with vertebral fracture in both men and women [39–41]. All of the above suggest that indices incorporating bone geometry, such as areal BMD, IBS and FRI, should take precedence over BMAD and volumetric BMD in evaluation of vertebral fracture risk and adaptation to interventions.

To evaluate fracture risk within our cohort, we generated a fracture risk index (FRI) based upon equations reported by Duan et al. [28]. EX/GYM demonstrated significant advantages in areal BMD (7.9% to 8.4%) and axial compressive strength (IBS, 9.6% to 17.6%), as well as lower fracture risk index (FRI, –10.4%). To date, despite its flaws, DXA PABMD remains a potent indicator of vertebral fracture risk [19], likely because it encapsulates gross assessments of both bone geometry and density. Similarly, work by Roux et al. supports the value of lateral areal BMD in prediction of L3 vertebral failure load, stiffness and work to failure [34]. FRI has also been shown to be a useful predictor of fracture risk, as FRI greater than 1 has been associated with vertebral fracture in men and women [28]. In our analyses, loading associated differences in IBS (PALAT 9.6%, PA 17.6%, LAT 16.4%) and our modified version of FRI (10.4%) are of greater magnitude than areal BMD advantages (PA 8.4%, LAT 7.9%). We would expect the derivation of IBS and FRI to improve sensitivity to detect mechanical loading adaptation and DXA fracture prediction through the incorporation of bone cross-sectional area and BMAD.

Our results demonstrate the influence of vertebral depth/width ratio in both maturity and gymnastic loading comparisons, as the relationship between vertebral width and depth is associated with both age [19] and loading exposure. Without adjustment for age and height, most variables from PA, lateral and paired scan output were greater in post-menarcheal than pre-menarcheal subjects and were significantly positively correlated with age and body size, indicating vertebral growth in proportion to overall growth. However, PAWIDTH and PACSA were not correlated with age and body size parameters but were significantly greater in EX/GYM than NON, suggesting that medial-lateral vertebral expansion may be the predominant marker of gymnastic loading exposure.

The observation of muted differences in vertebral depth deserves discussion. We speculate that the necessity for antero-posterior vertebral expansion (LATDEPTH) is diminished by tensile resistance from the posterior elements and their ligamentous connections. In contrast, the comparative absence of medial-lateral accessory support may require expansion of vertebral PAWIDTH to resist mechanical loading. Furthermore, direct psoas muscle forces may stimulate medial-lateral vertebral expansion, as these muscles originate from the lateral aspects of the lumbar vertebrae. This hypothesis is supported by correlations of bone parameters vs. sit-up performance, which reflects psoas functional muscular endurance (negative correlation: sit-ups vs. fracture risk index; positive correlations: all but LATVHEIGHT and PALATBMAD (data not shown)).

Our vertebral geometric findings echo observations in EX/GYM relative to NON at the distal radius [29–32, 42–43]. At both the radial metaphysis and the lumbar spine, larger bone geometry and greater IBS underlie EX/GYM areal BMD advantages. However, distal radius advantages appear to be of a greater magnitude, presumably due to more direct force application and lower levels of force dampening at the distal radius than at the lumbar spine [42]. In a related cohort, pQCT-measured distal radius trabecular volumetric BMD was greater in post-menarcheal EX/GYM than non-gymnasts with no concomitant advantage in total volumetric BMD (BMAD), potentially due to large geometric differences (larger denominator for EX/GYM BMAD equation) [31]. Thus, the lack of EX/GYM L3 BMAD advantages may not rule out advantages in core trabecular volumetric BMD, as the latter

cannot be detected by DXA. Nonetheless failure to detect a PALATBMAD advantage may reflect deterioration of benefits among post-menarcheal ex-gymnasts after training cessation.

Dietary composition was not a major focus of the current study. Nonetheless, we evaluated food frequency questionnaire results (including supplement use) to account for the potential influence of calcium intake. Differences were not detected between EX/GYM AND NON, but post-menarcheal girls' intakes were lower than intakes for pre-menarcheal girls. Grand mean intake was below the current recommended daily intake of 1300mg/day for girls aged 9–18 yrs [44], and the range of intakes was broad. Nonetheless, there was no significant correlation between calcium intake and any bone variable. Because the strongest trend was for LATVHEIGHT ($r^2 = 0.033$, $p = 0.066$), we entered calcium intake as a covariate into a supplemental ANCOVA for LATVHEIGHT. Even in this case, calcium intake was not influential ($p > 0.10$), and results were qualitatively unchanged. Accordingly, in this context, exposure to mechanical loading appears to exert a more potent influence upon lumbar vertebral body characteristics than calcium intake.

Limitations

First and foremost, this is a cross-sectional analysis in which pre- and post-menarcheal subjects are not the same individuals. Accordingly, inter-subject genetic and environmental variability may have been influential in pre-menarche vs. post-menarche comparisons. This design issue may have confounded evaluation of maturity-specific effects. Furthermore, inter-individual differences in childhood loading exposure may have affected evaluation of interactions between maturity and loading.

Second, because this is an observational study and not a randomized controlled trial, it is possible that the reported advantages do not reflect adaptation to loading. Furthermore, evaluation of gymnastics $\times$ maturity interactions and POST EX/GYM vs. NON differences may have been affected by lack of peri-menarcheal exposure in pre-menarcheal quitters and/or the lapse between gymnastic exposure and study assessment. At the time of the DXA scans, the majority of POST EX/GYM had not trained for over a year, and some had discontinued gymnastics more than one year before menarche. The majority of our ex-gymnasts quit for non-medical reasons, such as changing to other activities and graduating from high school. However, three ex-gymnasts quit secondary to back injuries; none of these involved vertebral body fracture. In addition, 11 of the 68 pre-menarcheal girls achieved menarche within one year following the focal DXA scan, potentially reducing maturity group differences. Overall, gymnastic exposure was low compared to other study cohorts, providing a conservative assessment of loading associations. This conservatism suggests that lumbar spine fracture risk may be reduced with relatively low gymnastic exposure, and benefits may still persist well after training cessation. Thus, although interactions between exposure and maturity were not detected in this analysis, longitudinal studies of high-level gymnasts who continue training throughout puberty may demonstrate more profound advantages and/or maturity-specific benefits.

Third, PA DXA scans of the lumbar spine may be affected by fan beam magnification error (FBME), leading to erroneously low values for PABMC and geometric measurements in individuals whose vertebral distance is higher from the table top (larger body size/waist circumference). As NON exhibited significantly greater waist circumferences than EX/GYM, we were concerned that this may have affected our results. To address this issue, we applied an FBME correction to our study data; results were qualitatively unchanged or differences were augmented (IBS). On this basis, it is unlikely that FBME was the sole source of observed differences in bone dimensions and IBS.

Fourth, although the majority of DXA's shortcomings were alleviated by use of paired PA and lateral scans, it is important to note that DXA outcomes are gross indicators of bone mass and size, incapable of evaluating bone microarchitecture or tissue composition. Thus, subtle adaptations in trabecular structure, material density or structural variability would not have been detected. It is possible that gymnast vertebrae may have larger, more numerous trabeculae with greater connectivity and lower spacing, as suggested by micro-MRI results at the tibia [45]. In this scenario, BMAD differences may be moderated by lower material volumetric BMD as a function of higher bone turnover rates (as suggested by pQCT studies in gymnasts [31,42,43,46]). Furthermore, it is possible that spatial distribution of bone mineral content within the vertebral body differs between EX/GYM and NON, yielding benefits in fracture resistance without exhibiting a detectable BMAD advantage. Future micro-MRI studies may elucidate micro-architectural parameters without undesirable truncal radiation exposure.

Fifth, although we feel that a PALAT ellipsoid cylindrical model equates true vertebral body shape better than a PALAT cuboid model, neither model captures true vertebral shape. CT or MRI imaging is necessary in order to accurately assess vertebral body shape, but neither of these modalities was appropriate for the current study due to excessive radiation dose and cost (respectively). It is important to consider that as long as height, width and depth are assessed and accounted for independently, ellipsoid cylinder and cuboid models differ only by multiplication by a constant ($\pi/4$ or $4/\pi$). Thus, regardless of PALAT model form, ellipsoid cylinder and cuboid results will be perfectly correlated, potentially underestimating and overestimating true volume, respectively. As long as all three vertebral dimensions are taken into account, estimates of vertebral geometry and volumetric density will be improved relative to Carter PA cuboid results.

Finally, although we report food frequency results for assessment of habitual calcium intake, we acknowledge the shortcomings of this dietary instrument. It is notoriously difficult to represent variability in daily nutrient intake in humans, and this was not a primary aim of our study. Our assessments were intended primarily to evaluate general sufficiency of habitual intake and to account for systematic intake differences in EX/GYM versus NON. The questionnaire that we used may rely unduly on dairy foods for assessment and is therefore unlikely to account for confounding issues such as acid balance and associated calcium retention/wasting. Issues of specific nutrient intake and dietary composition are best addressed in nutritional studies designed primarily to address these concerns.

Conclusion

Gymnastic exposure during growth is associated with greater vertebral compressive strength and areal BMD; both are primarily a function of large vertebral cross-sectional geometry. This advantage is observed in both immature girls of relatively low gymnastic exposure and mature young women who had discontinued gymnastics an average of five years prior. Critically, comparisons of PA, lateral and paired scan output show that results vary considerably by scan plane. For more accurate assessment of skeletal adaptation to mechanical loading, outcomes should be evaluated based upon paired scan output, comparing and contrasting geometric and densitometric results from each DXA plane. The gymnastic model of mechanical loading is associated with expansion in vertebral dimensions that emphasizes PA width over lateral depth, suggesting that bone geometry plays a critical role in vertebral fracture resistance. Accordingly, future studies should specifically evaluate lumbar spine geometry and skeletal strength indices in addition to standard bone outcomes.

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Abbreviations

DXA	dual energy X-ray absorptiometry
ANCOVA	analysis of covariance
EX/GYM	ex/gymnasts
NON	non-gymnasts
PRE	pre-menarcheal
POST	post-menarcheal
L3	lumbar vertebra 3
PA	Posteroanterior DXA scan
LAT	Lateral DXA scan
PALAT	paired PA & LAT DXA scan output
CSA	cross-sectional area
FBME	fan beam magnification error
BMD	areal bone mineral density
BMC	bone mineral content
Area	projected area
LATVHEIGHT	vertebral body height, measured by lateral scan
BMAD	bone mineral apparent density
PAVOLUME_C	volume derived using Carter et al. cuboid formula
PABMAD_C	BMAD derived using Carter et al. cuboid formula
IBS	index of bone strength in axial compression
FRI	fracture risk index
nbFFM	non-bone fat-free mass
WABMD	width-adjusted BMD, a form of BMAD
WAVOLUME	width-adjusted volume

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For 3 dimensions without posterior element BMC:
 Combine Postero-Anterior Width with
 Lateral Depth, Lateral Height and Lateral BMC

Figure 1.

Simplified Geometric Model of Lumbar Vertebral Body L3: Mean width derived from the PA DXA scan is combined with mean height and mean depth derived from the Lateral DXA scan, yielding a three-dimensional model of the L3 vertebral body (elliptical cylinder). Using this model, mean width, depth and height are allowed to vary independently. This contrasts with the cuboid model of vertebral volume published by Carter et al. [27], in which PA projected area is cubed, assuming that mean width, mean height and mean depth are equal.

Lumbar Spine: PA vs. Lateral Outcomes

Figure 2.

Percent advantages are presented for ex/gymnasts relative to non-gymnasts in graphical form. The zero line represents the mean for non-gymnasts. Reported outcomes account for the main effect of menarche status, the interaction between menarche status and gymnastic exposure, and the effects of age and height. Pale grey bars represent PA values; dark grey bars represent Lateral values. a= $p < 0.05$; b= $p \leq 0.01$; c= $p \leq 0.001$.

Lumbar Spine: PA, Lateral and PA/Lateral Outcomes

Figure 3.

Percent advantages are presented for ex/gymnasts relative to non-gymnasts in graphical form. The zero line represents the adjusted mean for non-gymnasts. Reported outcomes account for the main effect of menarche status, the interaction between menarche status and gymnastic exposure, and the effects of age and height, except for FRI which is adjusted only for age. Pale grey bars represent PA values; dark grey represents lateral scan values; striped bars represent PALAT (paired scan) values; scan outcomes are placed in this order from left to right. a= $p < 0.05$; b= $p \leq 0.01$; c= $p \leq 0.001$.

Table 1

Formulae for Lumbar Spine Geometric and Densitometric Derivations

Scan Type	Variable	Formula (Reference)	Units
Postero- Anterior (PA)	PACSA	$(\pi/4) * ((\text{PAAREA}/\text{LATVHT})^2)$	cm ²
	PAVOLUME	$(4 * \text{LATVHT})/(\pi * \text{PAAREA}^2)$	cm ³
	PAVOLUME _c (cuboid, not reported)	$(\text{PAAREA})^{1.5}$ {27}	cm ³
	PABMAD	$\text{PABMC}/(\pi^4 * (\text{PAAREA}/\text{LATVHT}) * (\text{PAAREA}/\text{LATVHT}) * (\text{LATVHT})) \text{ OR } (4 * \text{PABMC} * \text{LATVHT})/(\pi * \text{PAAREA}^2)$ {19}	g/cm ³
Lateral (LAT)	PABMAD _c (cuboid, not reported)	$\text{PABMC}/(\text{PAAREA})^{1.5}$ {27}	g/cm ³
	PAIBS	$4/\pi * \text{PABMD}^2 \text{ OR } \text{PABMAD}^2 * \text{PACSA}$ {26}	g ² /cm ⁴
	LATCSA	$(\pi/4) * (\text{LATAREA}/\text{LATVHT})^2$	cm ²
	LATVOLUME	$(4 * \text{LATVHT})/(\pi * \text{LATAREA}^2)$ {19}	cm ³
Paired (PA/LAT)	LATBMAD or Hologic WABMD	$(4 * \text{LATBMC} * \text{LATVHT})/(\pi * \text{LATAREA}^2)$	g/cm ³
	LATIBS	$4/\pi * \text{LATBMD}^2 \text{ OR } \text{LATBMAD}^2 * \text{LATCSA}$ {26}	g ² /cm ⁴
	PALATCSA	$(\pi/4) * \text{PAW IDTH} * \text{LATDEPTH}$	cm ²
	PALATVOLUME	$(\pi/4) * \text{PAWIDTH} * \text{LATDEPTH} * \text{LATVHT}$ {19}	cm ³
Fracture Risk Index (FRI)	PALATBMAD	$\text{LATBMC}/((\pi/4) * \text{PAWIDTH} * \text{LATDEPTH} * \text{LATVHT})$ {19}	g/cm ³
	PALATIBS	$\text{PALATBMAD}^2 * \text{PALATCSA}$ {26}	g ² /cm ⁴
	FRACTURE RISK INDEX (FRI)	$((0.455 * \text{WT}/\text{PALATCSA}) * (0.186 \text{ STHT}/\text{LATDEPTH}))/ (8515 * \text{PALATBMAD})^{1.6}$ (modified, substituting PALAT for LAT measures, as appropriate, 28)	

PA= Postero-anterior; BMAD= bone mineral apparent density; BMC= bone mineral content; LAT= lateral; HOLOGIC WABMD= for output from Discovery A software version 12.7.3.2.3, we found WABMD to be equivalent to our derivation of LATBMAD; LATVHT= lateral vertebral height (roughly equivalent to PA vertebral height); π = pi; AREA= vertebral projected area; IBS= index of structural strength in axial compression; BMD= areal BMD; CSA= derived cross-sectional area; WABMD= width-adjusted BMAD; WT= total body mass (kg); STHT= total body stature (m).

Table 2

Subject Characteristics: Means and 95% Confidence Intervals

Variable	Total Sample		Pre-Menarche		Post-Menarche	
	Ex/Gym n= 60	Non-Gym n= 54	Gym n= 35	Non-Gym n= 33	Ex/Gym n= 25	Non-Gym n= 21
Chronological Age (yrs)	14.2 (13.7 to 14.7)	13.8 (13.3 to 14.4)	10.6 (10.1 to 11.1)	9.9 (9.4 to 10.4)	19.1 (18.0 to 20.2)	19.3 (18.2 to 20.6)
Tanner Breast Stage (I–V, Percent Frequencies)	I=35.0%; II=16.7%; III=8.3%; IV=6.7%; V=31.7%	I=35.2%; II=22.2%; III=7.4%; IV=11.1%; V=24.1%	I= 60.0%; II= 28.6%; III= 8.6%	I= 57.6%; II= 36.4%; III= 6.1%	III= 8.0%; IV= 16.0%; V= 76.0%	III= 9.5%; IV= 28.6%; V= 61.9%
Height (cm)	149.0 (146.3 to 151.7)	152.9 ($p<0.06$) (149.9 to 155.9)	138.2 (135.1 to 141.5)	140.8 (137.4 to 144.2)	160.6 (156.3 to 165.2)	166.0 (161.1 to 171.1)
Weight (kg)	43.6 (41.1 to 46.2)	45.9 (43.1 to 48.9)	33.6 (31.2 to 36.2)	35.4 (32.8 to 38.3)	56.6 (51.8 to 61.9)	59.4 (53.9 to 65.6)
DXA FEM (kg)	32.3 (30.7 to 34.1)	32.1 (30.4 to 34.0)	25.4 (23.7 to 27.1)	24.8 (23.1 to 26.5)	41.2 (38.0 to 44.6)	41.7 (38.2 to 45.5)
BMI (kg/cm ²)	19.6 (18.9 to 20.3)	19.6 (18.9 to 20.5)	17.5 (16.7 to 18.4)	17.9 (17.0 to 18.8)	21.9 (20.7 to 23.2)	21.6 (20.3 to 23.0)
Percent Body Fat (%)	21.8 (20.7 to 23.0)	26.4^a (24.9 to 27.9)	20.2 (18.9 to 21.7)	26.0^a (24.3 to 27.9)	23.5 (21.7 to 25.5)	26.7^a (24.4 to 29.2)
Waist Circumference (cm)	64.5 (62.6 to 66.4)	67.3 ($p<0.06$) (65.2 to 69.5)	58.8 (56.6 to 61.1)	62.9 (60.4 to 65.4)	70.8 (67.6 to 74.1)	72.0 (68.6 to 75.7)
Sit-ups (1 minute count)	48.4^a (45.6 to 51.4)	39.1 (36.7 to 41.5)	45.6^a (42.3 to 49.2)	36.0 (33.3 to 38.8)	51.4^a (46.9 to 56.3)	42.4 (38.5 to 46.6)
Age at Menarche (yrs)	---	---	N/A	N/A	13.4 (12.9 to 13.9)	13.3 (12.7 to 13.8)
Gynecological Age (yrs)	---	---	N/A	N/A	4.9 (3.8 to 6.3)	5.8 (4.4 to 7.6)
Physical Activity (h/wk)	7.4^a (6.1 to 9.0)	3.0 (2.4 to 3.7)	11.5^a (9.0 to 14.8)	2.7 (2.1 to 3.5)	4.8 (3.6 to 6.5)	3.3 (2.4 to 4.6)
Gymnastic Activity (h/wk) (current gymnasts only)	10.5 (8.7 to 12.6)	---	10.7 (9.3 to 12.4)	---	10.2 (7.2 to 14.4)	---
Calcium Intake (mg/day) n=104	838.0 (719.1 to 976.5)	826.3 (702.0 to 973.6)	895.2 (741.0 to 1081.4)	1001.2 (821.4 to 1220.5)	784.5 (617.1 to 998.2)	682.0 (525.8 to 884.5)

DXA FFM= Total Body Non-bone Lean Mass; BMI= Body Mass Index; Gynecological Age= Years post-menarche; Physical Activity= Total organized activity (includes gymnastics, excludes aquatics).

^a Bold type indicates significantly greater mean in Ex/Gym vs Non-Gym comparisons ($p<0.05$).[‡] Significant maturity $\times$ gymnastics interaction: PREGYM > PRENON; PREGYM > POSTGYM; PRENON $\approx$ POSTNON; POSTGYM $\approx$ POSTNON.Post-menarcheal means are significantly greater than pre-menarcheal means, except gymnastics, physical activity & calcium (PRE > POST, $p<0.05$).

Table 3
Two-Factor ANCOVA (activity $\times$ maturity): Lumbar Spine (L3) Results Adjusted for Chronological Age and Height.

DXA Plane	Variable	Mean (95% CI)		Mean (95% CI)	
		Ex/Gym n=60	Non n=54	Pre-Men n=68	Post-Men n=46
Postero- Anterior (PA)	PABMD (g/cm ²)	0.882 (0.854 – 0.911) ^b	0.814 (0.787 – 0.841)	0.769 (0.733 – 0.806)	0.934 (0.875 – 0.998) ^c
	PABMC (g)	10.86 (10.40 – 11.35) ^b	9.81 (9.37 – 10.25)	8.65 (8.13 – 9.23)	12.30 (11.26 – 13.44) ^c
	PAAREA (cm ²)	12.32 (12.03 – 12.60)	12.05 (11.76 – 12.33)	11.26 (10.88 – 11.66)	13.17 (12.55 – 13.80) ^c
	PAWIDTH (cm)	6.80 (6.57 – 7.04) ^c	6.27 (6.06 – 6.49)	6.15 (5.85 – 6.47)	6.93 (6.46 – 7.43) ^a
	PABMAD (g/cm ³)	0.165 (0.158 – 0.173)	0.165 (0.157 – 0.173)	0.159 (0.148 – 0.171)	0.172 (0.156 – 0.189)
	PAIBS (g ² /cm ⁴)	0.991 (0.929 – 1.058) ^c	0.843 (0.790 – 0.900)	0.752 (0.684 – 0.827)	1.112 (0.974 – 1.267) ^c
	PACSA (cm ²)	36.31 (33.89 – 38.90) ^b	30.91 (28.82 – 33.15)	29.73 (26.87 – 32.85)	37.75 (32.82 – 43.47) ^a
	PAVOLUME (cm ³)	65.69 (62.30 – 69.34) ^b	59.38 (56.20 – 62.68)	54.43 (50.35 – 58.91)	71.66 (64.26 – 79.84) ^b
	LATBMD (g/cm ²)	0.748 (0.725 – 0.771) ^c	0.693 (0.672 – 0.715)	0.681 (0.651 – 0.712)	0.760 (0.714 – 0.809) ^a
	LATBMC (g)	5.53 (5.28 – 5.80)	5.32 (5.08 – 5.58)	4.85 (4.53 – 5.20)	6.07 (5.52 – 6.68) ^a
Lateral (LAT)	LATAREA (cm ²)	7.40 (7.23 – 7.59)	7.68 (7.50 – 7.88) ^b	7.12 (6.88 – 7.38)	7.99 (7.61 – 8.40) ^b
	LATDEPTH (cm)	4.09 (4.02 – 4.16)	4.00 (3.94 – 4.08)	3.89 (3.79 – 3.99)	4.21 (4.06 – 4.36) ^b
	LATVHEIGHT (cm)	1.81 (1.77 – 1.86)	1.92 (1.87 – 1.97) ^c	1.83 (1.77 – 1.90)	1.90 (1.81 – 2.00)
	LATBMAD (g/cm ³)	0.233 (0.226 – 0.240) ^b	0.220 (0.214 – 0.227)	0.223 (0.214 – 0.233)	0.230 (0.217 – 0.244)
	LATIBS (g ² /cm ⁴)	0.711 (0.668 – 0.757) ^c	0.611 (0.574 – 0.651)	0.590 (0.540 – 0.647)	0.736 (0.649 – 0.834) ^a
	LATCSA (cm ²)	13.13 (12.68 – 13.60)	12.59 (12.15 – 13.04)	11.89 (11.30 – 12.50)	13.90 (12.96 – 14.92) ^b
	LATVOLUME (cm ³)	23.78 (22.97 – 24.61)	24.17 (23.34 – 25.03)	21.76 (20.70 – 22.87)	26.42 (24.63 – 28.30) ^c
	PALATBMAD (g/cm ³)	0.140 (0.134 – 0.146)	0.141 (0.135 – 0.147)	0.141 (0.133 – 0.150)	0.140 (0.128 – 0.152)
	PALATIBS (g ² /cm ⁴)	0.427 (0.400 – 0.457)	0.390 (0.365 – 0.417)	0.374 (0.339 – 0.411)	0.446 (0.390 – 0.510)
	PALATCSA (cm ²)	21.82 (21.76 – 22.94) ^b	19.73 (18.75 – 20.76)	18.80 (17.48 – 20.23)	22.90 (20.70 – 25.36) ^a
Paired PA/LAT	PALATVOLUME (cm ³)	39.53 (38.02 – 41.10)	37.86 (36.42 – 39.41)	34.43 (32.52 – 36.45)	43.51 (40.17 – 47.09) ^c
	FRACTURE RISK INDEX ^{\$}	0.160 (0.150 – 0.170)	0.182 (0.171 – 0.194) ^b	0.190 (0.174 – 0.208) ^a	0.153 (0.135 – 0.174)

DXA Plane	Variable	Mean (95% CI)		Mean (95% CI)	
		Ex/Gym n=60	Non n=54	Pre-Men n=68	Post-Men n=46
	LATBMC/PABMC [#]	51.8 (50.4 – 53.3)	55.1 (53.5 – 56.8) ^b	52.5 (51.1 – 53.8)	54.5 (52.7 – 56.2) ^b
	LATDEPTH/PAWIDTH [#]	60.9 (59.6 – 62.3)	64.9 (63.4 – 66.5) ^c	59.4 (58.2 – 60.7)	66.5 (64.9 – 68.2) ^c

There were no significant interactions between maturity and activity for any dependent variable.

All variables were analyzed as natural logarithms; results are presented in antiln form.

For all results other than ratios, means and 95% confidence intervals account for the main effect of menarche status, the interaction between menarche status and gymnastic exposure, and the effects of age and height (except as noted below).

[§] Results for Fracture risk index are not adjusted for the linear effect of height (ANCOVA), due to inclusion of height in the formula.

[#] Results for ratios LATBMC/PABMC and LATDEPTH/PAWIDTH reflect simple ANOVA (no adjustment), but still account for the main effect of menarche status and the interaction between menarche status and gymnastic exposure.

^a p<0.05,

^b p≤ 0.01,

^c p≤ 0.001