Lean mass explains the association between muscular fitness and bone outcomes in 13-year-old boys

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Running head: Fitness and bone outcomes in teenage boys

Abstract

Aim: This study investigated the associations between fitness indices and bone outcomes in young males.

Methods: Data were collected between autumn and winter 2014-2015 on 121 males with a mean age of 13.1 ±0.1 years: 41 swimmers, 37 footballers, 29 cyclists and 14 non-athletes. Participants were recruited from athletic clubs and schools across South West England. Lean mass, areal bone mineral density and hip structural estimates were measured using dual-energy X-ray absorptiometry. The relationships between bone outcomes and the vertical jump, standing long jump and the 20m shuttle run test were analysed using three regression models: model one was adjusted by age and stature, model two added vigorous physical activity and model three then added lean mass.

Results: The boys' performance in the vertical jump and standing long jump was positively associated with the majority of bone outcomes in models one and two, but most of these disappeared in model three. The 20m shuttle run test was positively associated with most bone outcomes in all three models. Lean mass played a key role in the association between muscular fitness and bone outcomes.

Conclusion: Vigorous physical activity did not explain the associations between fitness and bone outcomes, but lean mass did.

Keywords: Body composition; Bone health; Cardiorespiratory fitness; Lean mass; Physical activity

Key notes:

- This study investigated the associations between fitness indices and bone outcomes in young males in South West England.
- Data were collected between autumn and winter 2014-2015 on 121 males with a mean age of 13.1 ±0.1 years: 41 swimmers, 37 footballers, 29 cyclists and 14 non-athletes.
- It found that vigorous physical activity did not explain the associations between fitness and bone outcomes, but lean mass did.

INTRODUCTION

Physical activity (PA) during adolescence has been related to bone health at that age and later in life (1). This appears to be due to the greater sensitivity of the skeleton to adapt to mechanical loading. Previous studies among pre-adolescent boys have suggested that cortical bone mineral content (BMC) and geometry, as assessed by tibial peripheral quantitative computed tomography, are positively associated with vigorous PA (VPA) (2, 3). Data from the Avon Longitudinal Study of Parents and Children showed that habitual levels of high impact activity were positively associated with hip areal bone mineral density (aBMD) and geometry in the adolescent population, but that was not the case for moderate or low impact activity (4). In particular, it has been shown that periods of at least 28 minutes of VPA per day have been associated with high femoral neck aBMD in adolescents (5).

Studies investigating the influence of different sports on bone mass (6) and bone geometry (7) in youth have shown that weight-bearing and high-impact sports, such as football, have a greater positive effect on bones while non weight-bearing and low impact sports, such as swimming and cycling, are associated with lower benefits (8). However, obtaining accurate and objectively measured PA using accelerometers during certain sports, including swimming and cycling, is undeniably challenging.

Physical fitness has been considered an important marker of health and its relationships with bone mass (9) and geometry (10) have been investigated. Muscular and cardiorespiratory fitness have been found to be positively associated with BMC in 16 to 18-year-old boys (11). Similarly, positive associations were found between muscular fitness and bone mass in a group of pre-pubertal boys, but an association between cardiorespiratory fitness and bone mass in the same population was not observed (12). Discrepancies among studies may be due to the inconsistent use of covariates, with some studies omitting important predictors of bone mass development during growth, such as VPA and lean mass (4, 5, 13). Therefore, the role of VPA and, or, lean mass in the association between fitness and bone outcomes requires further investigation.

The objective of this study was to examine the association between muscular and cardiorespiratory fitness with aBMD and hip geometry estimates in young males, including important covariates such as VPA and lean mass.

MATERIAL AND METHODS

Participants

A cross-sectional analysis of baseline data was derived from the PRO-BONE study (trial registration ISRCTN17982776), whose purpose and methodology have been described elsewhere (14). Data were collected between autumn and winter 2014-2015 from 121 male adolescents: 41 swimmers, 37 footballers, 29 cyclists and 14 non-athletes. Male adolescents who were 12 to 14 years old and had engaged in at least three hours per week of football, swimming or cycling for the last three years or more were recruited. This criteria was based on previous research that demonstrated osteogenic benefits with three hours of activity per week among adolescents (6). In addition, 12–14 years males who were not athletes and did not meet the above criteria were also recruited.

The participants were recruited from athletic clubs and schools across South West England. Parental consent and participant assent forms were signed and all participants underwent their first visit at the research centre as part of the study. The methods and procedures conformed to the Code of Ethics of the World Medical Association, namely the Declaration of Helsinki, and were checked and approved by the Ethics Review Sector of Directorate-General of Research of the European Commission, the Sport and Health Sciences Ethics Committee of the University of Exeter and the National Research Ethics Service Committee.

Dual energy x-ray absorptiometry (DXA)

A Lunar Prodigy DXA scanner (GE Healthcare Inc, Wisconsin, USA) was used to measure aBMD in g/cm^2 and lean mass in grams. Four scans were performed to obtain data for the whole body minus the head, the lumbar spine (L1-L4), the right and left hip, including the femoral neck, trochanter and total hip. The mean of the right and left hip scans were used. All DXA scans and subsequent software analyses were completed by a single researcher using the same DXA scanner and enCORE software version 14.10.022 (GE Healthcare Inc, Wisconsin, USA). The DXA equipment was calibrated prior to testing each day using a lumbar spine phantom following the manufacturer's recommendation. Precision studies in 14-16 year olds have shown the coefficient of variation for DXA to be 0.74% for the aBMD of the whole body less head, 0.64% for the lumbar spine aBMD, 0.65% for the total hip aBMD and 1.16% for the femoral neck aBMD (15).

Hip structural analysis

Previous studies have used hip structural analysis software to obtain the geometrical parameters associated with bone strength. This technique, which used the same DXA scanner as the present study, compared favourably with volumetric quantitative computed tomography (16). Analyses were performed using the hip structural software at the narrow neck region across the narrowest point of the femoral neck. The software programme uses the distribution of bone mineral mass in lines of pixels across the bone axis to measure the structural dimensions of bone cross sections. The geometric properties of the bone were obtained and the following variables used: the narrow neck width in mm, the cross-sectional moment of inertia in mm^4, the section modulus in mm^3 and the the cross-sectional area in mm^2. The short-term precision percentage coefficient of variation of these variables has been reported to be between 2.4% and 7.9% (17).

Physical fitness

The fitness tests used in this study have been shown to be reliable and valid in youths (18). The standing long jump and vertical jump in centimetres were used to provide an estimate of lower limb muscular fitness. The vertical jump was performed using a jump mat (Probotics Inc, Alabama, USA), which calculates the height of the jump based on the flight time. Each participant performed three maximal jumps and the best performance was used for the analysis.

Cardiorespiratory fitness was evaluated using the 20m shuttle run test. The participants were asked to run between two lines set 20m apart by following the pace of the audio signals produced from a compact disc player. All participants were equally encouraged to continue the test until they reached maximal effort. The test ended when the participants failed to reach the line on two consecutive occasions and the count of the last completed shuttle run was recorded.

Anthropometry, sexual maturation and physical activity

Stature in centimetres and body mass in kilograms were measured using standard procedures. Body mass index was calculated as body mass in kilograms divided by stature in meters squared. Sexual maturation was self-reported using adapted drawings of the five stages of pubertal hair development (19).

Physical activity was objectively measured using GENEActiv wrist accelerometers (GENEA, Cambridgeshire, UK) for seven consecutive days following the laboratory visit, including during training. Participants were instructed to place the accelerometer on their non-dominant wrist and data was collected at 100Hz. Data were analysed at one-second epoch intervals to establish the time spent at different intensities. The time spent in VPA was calculated using a cut-off point of ≥ 3,600 counts per minute (20). The validity and reliability of the accelerometer has previously been established in children and adolescents (20). The weekly training hours were obtained by face-to-face interviews during the participants' visit to the research centre.

Statistical analysis

The distribution of the variables was checked and verified using Shapiro-Wilk's test, skewness and kurtosis values, visual checks of histograms, Q-Q and box plots. Variables were also checked for collinearity using variance inflation factor values. Descriptive data were reported as means and standard deviations unless otherwise stated. Differences between the groups were determined using analysis of variance (ANOVA) for continuous variables or the chi-square test for categorical variables. Bivariate Pearson's correlations were performed to examine the association between predictors and outcomes. Relationships of physical fitness tests with aBMD and hip geometry estimates were analysed using multiple linear regression models according to three regression model structures. Model one was adjusted by age and stature, model two was adjusted by the covariates included in model one plus VPA and model three was adjusted by the covariates included in model two plus lean mass. These covariates were selected because of their proven correlation with bone outcomes (5, 6, 13). Statistical analyses were performed using SPSS version 22.0 for Windows (IBM Corp, New York, USA). Bonferroni correction was applied to control for multiple testing, which is considered to be the most conservative method of controlling for familywise error rates. Based on a desired alpha level of 0.05 and nine different hypotheses (outcomes), values of $p<0.006$ were considered statistically significant (0.05/9).

RESULTS

Descriptive characteristics of the participants are shown in Table 1. The swimmers were older, taller and leaner than the footballers and non-athletes and they weighed more than the footballers. The cyclists were older than the non-athletes. The footballers spent the most time doing moderate-to-vigorous PA (MVPA) and VPA, followed by the cyclists, then the swimmers and finally the non-athletes. The footballers had a higher total hip and trochanter aBMD than the swimmers and cyclists and the non-athletes had lower aBMD measurements than all the sporting groups at all sites except the lumbar spine, where only swimmers had significantly greater values. With regard to the hip structural analysis estimates, non-athletes had lower cross-sectional moment of inertia, section modulus and cross-sectional area than the sporting groups. Finally, non-athletes performed worse in all of the fitness tests compared with the sporting groups, except for the vertical jump when they were compared to the cyclists ($p=0.101$).Footballers had the best performance in the 20m shuttle run test when compared to all the other groups.

Bivariate Pearson's correlations between the bone outcomes and predictors are presented in Table 2. Age was positively correlated with stature (r=0.7) but negatively with VPA (r=-0.23). Both stature and lean mass were positively correlated with all bone parameters (r=0.39-0.75 and 0.44-0.84, respectively) except narrow neck width. VPA was positively correlated with cardiorespiratory fitness (r=0.25) and narrow neck width (r=0.22). Muscular fitness was positively correlated with all bone parameters (r=0.35-0.60) and also with cardiorespiratory fitness (r=0.5-0.62). Finally, cardiorespiratory fitness was positively correlated with all bone parameters (r=0.25-0.57).

The adjusted associations between muscular fitness and bone outcomes are shown in Table 3. In model one, performance in the vertical jump and standing long jump were positively associated with aBMD at all sites, with a semi partial correlation of 0.221 to 0.362, except for the vertical jump with femoral neck (p=0.011).They were also positively associated with all hip geometry estimates with a semi partial correction of 0.170 to 0.289, except for the narrow neck width, which was p=0.365 for the vertical jump and 0.096 for the standing long jump. Significant associations remained unchanged after VPA was added to create model two, with a semi partial correction of 0.174 to 0.322. Finally, once lean mass was added into the model to create model three, the most previous significant associations disappeared, except for those related to standing long jump at the total hip, with a semi partial correction of 0.221 and trochanter, with a semi partial correction of 0.225.

The adjusted associations between cardiorespiratory fitness and bone outcomes are presented in Table 4. In model one, performance in the 20m shuttle run test was positively associated with all bone outcomes with a semi partial correction of 0.269 to 0.474, except for narrow neck width (p=0.016). These significant associations remained unchanged after additional adjustment for VPA (model two) with a semi partial correction of 0.263 to 0.425. Finally, after adjusting for lean mass (model three), the significant associations at the lumbar spine (p=0.097) and femoral neck (p=0.008) disappeared, but the rest remained significant (p<0.006).

DISCUSSION

In the present study, VPA did not appear to explain the association between fitness and bone outcomes in young males and the positive associations between muscular fitness and aBMD and hip geometry estimates appeared to be dependent on lean mass.

Previous studies have suggested that objectively measured VPA is a key determinant of bone mass, geometry and strength (2). In children, 25 minutes of VPA per day has been suggested to enhance bone mass and geometry (21), while more than 28 minutes of VPA per day has been reported to predict high aBMD at the femoral neck in adolescents (5). However, other studies have suggested that the association between VPA and bone outcomes is mediated by other factors. A study showed that VPA explained 6.9% of the variability in geometry-related variables in children, such as the cross sectional area and section modulus (22). However, the association was weakened to 3.7% once lean mass was added into the regression model. In addition, another study conducted in boys and girls aged eight to 15 years, observed that the association between PA and femoral neck bone strength disappeared after adjusting for lean mass (23). In this study, objectively measured VPA did not seem to explain fitness associations, namely muscular and cardiorespiratory, with aBMD and hip geometry estimates. In fact, once the multiple linear regressions were adjusted for VPA in model two, the semi partial correlation did not vary for either muscular fitness, which ranged from 0.170-0.362 in model one to 0.174-0.322 in model two, or cardiorespiratory fitness, which ranged from 0.269-

0.474 in model one to 0.263-00.425 in model two. The fact that the participants in the present study spent most of their time doing moderate rather than VPA (Table 1) may partially explain this finding.

Previous studies have linked VPA to muscular (24) and cardiorespiratory fitness (25) and shown a better correlation to these measures than moderate PA. In the present study, VPA only correlated with cardiorespiratory fitness (r=0.25) but not with muscular fitness. A study in adolescents showed that for a given fitness level, namely muscular and cardiorespiratory, those who met the PA guidelines had higher BMC than their inactive peers and, for a given PA level, those with higher fitness had a higher BMC than those with lower fitness levels (9). These findings can be partially extrapolated to our study, in which positive associations between fitness, namely muscular and cardiorespiratory measures, and bone outcomes were found. However, in the present study, the time spent in VPA did not play a significant role. Our participants exceeded the current PA guidelines of 60 minutes of MVPA per day (101.3 ±33.8), but spent an average of just 16.4 ± 10.4 minutes per day on VPA. The latter value fell below the minimum threshold of 25 minutes of VPA per day in children and 28 minutes of VPA per day in adolescents that has been suggested to enhance bone outcomes and geometry. This could explain the non-significant role of VPA in our study. In addition, it is likely that our PA levels were underestimated, especially in those adolescents engaged in non-weight bearing activities, which is a common limitation of studies using accelerometers.

Previous studies have demonstrated a direct association between muscular fitness and bone outcomes in adolescents (12). However, there are discrepancies in the evaluation of covariates. Poor performance in the standing long jump test and in the handgrip test have been associated with a lower BMC in the whole body and lower limbs in adolescents after adjusting for sex, height, lean mass, calcium intake and pubertal status (9). Similarly, performance in the vertical jump has been positively associated with hip and lumbar bone accretion in pre-pubertal boys using height, body mass and age as covariates (12). A further study found a relationship between BMC and muscular fitness in adolescents, independently of maturation status and lean mass (26). Findings from the present study indicated that muscular fitness was positively related to aBMD and bone geometry estimates in active boys, as a function of lean mass. In this regard, lean mass is considered one of the best predictors of bone mass both during growth and throughout life (5). This relationship can be explained by the mechanostat theory, which states that bone strength is regulated by modelling and remodelling processes that depend on the mechanical forces applied to the skeleton (27). The present findings suggest that lean mass could be a mediator of the association between muscular fitness and bone outcomes (28).

The findings of our study also suggested that cardiorespiratory fitness was positively related to aBMD and hip geometry estimates in active young males. However, most of the associations found between cardiorespiratory fitness and bone outcomes did not seem to be a function of lean mass. We found that the further adjustments for lean mass in model three only attenuated the positive associations found in models one and two in most of the regions. In contrast, those at the lumbar spine aBMD and femoral neck aBMD disappeared. Our results were comparable to another study in which a low performance in the 20m shuttle run test was related to lower whole body BMC in active adolescents, after adjusting for sex, height, lean mass, calcium intake and pubertal status (9). Results from a 15-year longitudinal study showed that the associations between cardiorespiratory fitness, directly measured as the maximum oxygen consumption, and lumbar spine aBMD and hip aBMD in adolescents disappeared after adjusting for sex, stature, weight, skinfolds, biological age and calcium intake (29). These findings were comparable to our results in certain ways because the effect of lean mass was contained within the effect of total body weight and we also observed a function of lean mass in lumbar spine aBMD and a sub-region of the hip, namely femoral neck aBMD.

Limitations and strengths

Our study had a number of strengths and limitations. The data used for these analyses were cross-sectional and that means that we cannot establish cause-effect relationships. In addition, sexual maturation was not used as a covariate in our analyses. However, preliminary analyses showed that age was more strongly associated with bone outcomes and therefore it was used instead. The accelerometer methodology that we used to quantify VPA did not detect any association with bone outcomes. Accelerometers only register accelerations and, therefore, we may have underestimated the intensity of activities with extra weight as well as those that relate to non-weight bearing efforts. Nevertheless, accelerometry has been suggested to be one of the best measures for reporting PA (30). The 20m shuttle run test has been used to assess cardiorespiratory fitness, which may underestimate cyclists and swimmers' aerobic capacity, as they were not necessarily familiar with this type of activity. However, this test has been used worldwide in children and adolescents and it has been shown to be reliable and valid (18). This was the first study in active male adolescents that combined DXA and hip structural analysis software to provide a thorough insight into the differences between aBMD and hip geometry estimates.

CONCLUSION

This study found that objectively measured VPA did not appear to explain fitness associations with bone outcomes and that lean mass played a key role in the association between muscular fitness and bone outcomes in young males. These findings contribute to the considerable body of evidence that suggests that investigations of physical fitness and bone mass must be adjusted for the effects of lean mass.

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LIST OF ABBREVIATIONS

aBMD: areal bone mineral density; BMC: bone mineral content; DXA: Dual energy x-ray absorptiometry; PA: physical activity; VPA: vigorous physical activity; .

CONFLICT OF INTEREST

The authors have no conflicts of interest to declare.

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