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Effect of maturational timing on bone health in male adolescent athletes engaged in different sports: The PRO-BONE study

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ABSTRACT

Objectives: To describe differences in bone outcomes according to biological age in male athletes participating in osteogenic (OS) or non-osteogenic (NOS) sports.
Design: Longitudinal (12-months).
Methods: 104 adolescents (12–14 years) were measured at baseline and after 1y: OS group (n = 37 football or soccer players) and NOS group (n = 39 swimmers, n = 28 cyclists). Years from peak height velocity (PHV, −2 to +2) was used as a maturational landmark. Bone mineral content (BMC) was assessed using DXA. Hip structural analysis estimated cross-sectional area (CSA), cross-sectional moment of inertia (CSMI) and section modulus (Z) at the femoral neck (FN). Trabecular bone score (TBS) estimated lumbar spine (LS) texture. Quantitative ultrasound measured bone stiffness. Multilevel regression models adjusted by hours of training were fitted.
Results: Compared to NOS, OS had significantly greater total body (less head) BMC from PHV to +2 years from PHV (from 9.5% to 11.3%, respectively); LS BMC from −1 years from PHV to PHV (from 9.8% to 9.9%); hip BMC (from 11.6% to 22.9%), FN BMC (from 12.0% to 15.9%), TBS (from 4.2% to 4.8%) and stiffness index (from 11.9% to 23.3%) from −1 years from PHV to +2 years from PHV; and CSA (from 8.4% to 18.8%), Z (from 5.5% to 22.9%) and CSMI (from 10.6% to 23.3%) from −2 years from PHV to +2 years from PHV. There was a significant trend for the between-group differences to increase with biological age except for LS BMC and TBS.
Conclusions: These findings underline the differential bone response to different sports throughout the years surrounding PHV in male adolescent athletes.
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1. Introduction

Puberty is characterised by changes in the hormonal milieu (e.g. increases in growth hormone, insulin like growth factors and sex hormones) which promote bone accrual.[1] It is an important period to maximise bone accrual as the skeleton suffers rapid changes due to the processes of growth, modelling, and remodelling, with about a 5% additional bone formed by every remodelling cycle compared to resorption.[2] Also, bone mineral accrual depends on level of biological maturity and is site-specific.[3] Previous longitudinal studies have concluded that the timing, pattern and magnitude of bone accrual is a highly-individualised process,[4] and therefore, com-

parisons should be based on biological rather than chronological age.[5]

In this regard, using PHV during growth is a useful alternative to chronological age.[6] PHV is the period of time of maximum growth in stature and years from PHV is considered in terms of time before and time after the PHV. In boys, age at PHV occurs approximately between 13 and 14 years old,[6] and it is considered an appropriate marker of somatic maturity. During the period between −2 to 2 years from PHV, males and females accrue 39% of their adult total body bone mineral content (BMC), 43% of their adult lumbar spine (LS) BMC, 46% of their adult total hip BMC and 33% of their adult femoral neck (FN) BMC.[3]

Similar to bone development during puberty, the type of sport practiced affects the skeleton in a site-specific manner. Intervention studies indicate that during the early pubertal stages, bone mineralisation is high and the skeleton is particularly responsive to exercise stimulus, for example weight-bearing activities.[7] Cross-sectional studies in children[8] and adolescents[9] suggest that those engaged in osteogenic sports (OS, i.e. football, basketball or handball) have higher BMC and areal bone mineral density (aBMD) compared to those engaged in non-osteogenic sports (NOS, i.e. swimming or cycling). This is due to the fact that bone development is dependent on the mechanical load produced during the specific sport practised and the forces applied on the skeleton that trigger bone modelling and remodelling.[10] Sports that provide ground reaction forces greater than 3.5 times of body weight, applied in less than 0.1 s, seem to have the greatest potential to induce additional bone gains during puberty.[11]

Bone strength and fracture risk depends not only on aBMD and BMC, but also on bone structure and strength.[12] In this regard, hip structural analyses (HSA) provides information about bone geometry of the FN, a clinically relevant site related to fracture risk. Another technique such as quantitative ultrasound (QUS) provides useful information about the stiffness of the calcaneus, a robust indicator of bone density.[13] A cross sectional study demonstrated that adolescent athletes who participate in OS have higher CSA, CSMI, Z and bone stiffness compared to NOS.[9] Moreover, the trabecular bone score (TBS) of the LS can predict fracture risk and fragility of the LS [14]. Although most of the knowledge about TBS refers to adult population, TBS usually increases with growth and may provide valuable information about bone quality in young populations.[15] To our knowledge, there is a lack of studies using the combination of these techniques in adolescent male athletes.

Despite the established importance of the years surrounding PHV for the accrual of bone mass, there is limited evidence evaluating the effects of OS and NOS on bone outcomes in male adolescent athletes, and combining DXA, HSA, TBS and QUS outcomes. Therefore, the aim of this study was to investigate differences in bone outcomes according to years from PHV in young male athletes participating in OS (football) or NOS (swimming or cycling). We hypothesised that adolescent athletes engaged in OS will not only present greater bone outcomes when aligned against years from PHV compared to those in NOS, but also that the magnitude of the difference will increase with the level of maturity.

2. Methods

The present study shows a 12-month longitudinal analysis of sport participation as part of the longitudinal PRO-BONE (effect of a PROgram of short bouts of exercise on BONE health in adolescents involved in different sports) study, whose purpose, methodology, sample size calculations and inclusion/exclusion criteria have been fully described elsewhere.[16] The inclusion criteria were: male adolescents 12–14 years old, engaged (≥3 h/week) in OS (football or soccer) or NOS (swimming or cycling) for the last 3 years or more.

The exclusion criteria were: (1) not taking part in another clinical trial; (2) not having an acute infection lasting until <1 week before inclusion; (3) to be free of any medical history of diseases or medications affecting bone metabolism; (4) to be white Caucasian.

For the present study, data were obtained at baseline (T0) during autumn/winter 2014/15 and at follow-up (T1) during autumn/winter 2015/2016 (mean difference of visits = 372 days). After exclusion of three participants who dropped out from the study before T1, the study sample was composed by one hundred and four 12–14 year old adolescent male athletes. Baseline anthropometry and bone outcomes did not differ between those who withdrew and those who continued in the study (data not shown).

Participants and parents/guardians were contacted through athletic clubs in the South West of England to participate in the study. Informative meetings were organised to explain the project and answer questions that arose. At the end of these meetings, consent forms and information letters were given for consideration and reminders calls were performed to those that did not send the consent form to check whether they wished or not to participate.

Written informed consent and assent was signed from parents and participants, respectively. The methods of the study have been approved by: (1) the European Commission (no. 618496); (2) the University of Exeter (no. 2014/766) and (3) the National Research Ethics Service Committee (no. 14/SW/0060).

Body mass (kg) and stature (cm) were measured following standard procedures and body mass index (BMI, kg/m^2) was calculated.

Years from PHV was used as a maturational landmark and was predicted using age and height in a validated algorithm in healthy children.[17] Each participant had a chronological age and biological age (calculated as years from PHV) associated with each testing occasion. Biological age categories were constructed using 1-year intervals such that the −1 year from PHV group included observations between −0.49 and −1.50 years from (i.e., before) PHV, as performed in previous studies.[3,5] According to the participants' characteristics, five groups were created (at −2 years from PHV, at −1 year from PHV, at PHV, at +1 year from PHV and at +2 years from PHV).

A Lunar Prodigy DXA scanner (GE Healthcare Inc., Wisconsin, USA) was used to assess BMC (g), and whole body lean mass (g). The whole body (total body less head, TBLH), LS (L1–L4) and the mean of right and left hip scans (total hip, and femoral neck, FN) were used to measure BMC. All DXA scans and subsequent in-software analyses were completed by the same researcher and the GE encore software (2006, version 14.10.022). The coefficients of variation have been reported in previous studies as 0.81% for TBLH BMC and 0.89% for LS BMC in 14–16 years old.[18]

HSA software was used to estimate the hip geometry of the FN (the mean of right and left hip scans) and the following variables were used: CSA (mm^2). Z (mm^3), and CSMI (mm^4). The coefficients of variation of these variables have been reported in previous studies and range from 7.9% to 11.7%.[19]

TBS is a DXA based technological tool that provides an index of bone microarchitectural texture in the LS. All TBS analyses were performed by the same trained researcher using the TBS iNsight Software (Medimaps, research version 3.0, Pessac, France). The coefficients of variation of TBS in relation to BMC are between 1.1 to 1.9%.[14]

QUS measurements to measure bone stiffness were carried out by Lunar Achilles Insight (TM Insight GE Healthcare, Milwaukee, WI, USA). Both feet were measured twice and the mean of the means was calculated. The precision data for QUS in children has been reported as 1.8% for stiffness.[20]

Statistical analyses were performed using SPSS version 22.0 for Windows (IBM Corp, New York, USA) and the significance level was set at p < 0.05. Data were expressed as mean (standard deviation, SD) unless otherwise stated. Normal distribution of variables

Table 1
Descriptive data at peak height velocity (PHV).

	Osteogenic sport (N = 23)	Non-osteogenic sports (N = 38)
Age (years)	13.7 (0.4)	13.6 (0.4)
Stature (cm)	161.9 (5.7)	163.7 (5.8)
Body mass (kg)	49.4 (5.9)	51.7 (8.5)
BMI (kg/m²)	18.8 (1.4)	19.2 (2.5)
Lean mass (kg)	40.10 (5.32)	39.51 (4.85)
Hours of training	9.4 (1.6)*	6.4 (2.9)

Values presented as mean (SD).
Differences between osteogenic and non-osteogenic sports at PHV * p < 0.001.
BMI, body mass index.

was checked and verified using Shapiro–Wilk's test and visual check of histograms. Independent sample t-tests were performed to assess: descriptive differences between groups (OS and NOS) at PHV; differences in chronological age by years from PHV (from −2 to +2) and; raw differences in bone outcomes between OS and NOS groups by years from PHV (from −2 to +2), respectively. Hierarchical linear models were constructed using a multilevel modelling technique commonly used in the analysis of the repeated measures/longitudinal data. Multi-level modelling accounts for between-child variation by modelling within-child trajectories. This is achieved by entering 'years from PHV' into the model as a random effect, thus allowing the 'years from PHV'-related trajectories to vary for each individual child. In addition, analysis of covariance (ANCOVA) was used to assess mean-adjusted differences in bone outcomes between OS and NOS groups at each category of years from PHV. Hours of training was used as a covariate due to the significant differences observed between OS and NOS at PHV.

3. Results

Descriptive characteristics of the participants at PHV by type of sport are shown in Table 1. The OS group trained more hours per week compared to NOS group (p < 0.001) but there were no significant differences in age, stature, body mass, BMI and lean mass between the OS and NOS groups. In addition, OS and NOS athletes did not differ in chronological age at any PHV (Supplementary Table 1).

Results of unadjusted bone outcomes by years from PHV and type of sport are presented in Supplementary Table 2. All bone outcomes increased during growth both in the OS and NOS groups. The OS group had higher values on all bone outcomes compared to the NOS. More specifically, CSA was higher from −2 to +2 years from PHV; hip BMC, FN BMC, Z, CSMI and stiffness index from −1 to +2 years from PHV; TBS from −1 to +1 years from PHV; LS from −1 year from PHV to PHV and; TBLH at −1, +1 and +2 years from PHV.

Fig. 1 presents BMC-adjusted data by years from PHV and type of sport. Compared to the NOS group, the OS group had significantly greater TBLH BMC from PHV to +2 years from PHV, LS BMC from −1 year from PHV to PHV and, hip and FN BMC from −1 to +2 years from PHV (all p < 0.05). In addition, for TBLH, the interaction coefficient was 47.5 g (p = 0.012), so for every 1 unit increase in years from PHV, the BMC of those in the OS group goes up 47.5 g more than those in the NOS group. For example, −2 years from PHV, the BMC of the OS group was 56.7 g greater than the NOS group, yet +2 years from PHV, the BMC of the OS group was 246.7 g greater than the NOS group. The interaction coefficient for hip was 1.9 g (p = 0.014) and for FN 0.1 g (p = 0.016). However, no interaction was found for LS (p = 0.253).

Fig. 2 presents HSA, TBS and stiffness index-adjusted data by years from PHV and type of sport. The OS group showed significantly greater values in CSA, Z and CSMI from −2 to +2 years from PHV compared to the NOS group. The OS group had significantly

greater scores in TBS and stiffness index from −1 to +2 years from PHV compared to the NOS group. Moreover, for CSA, the interaction coefficient was 5.7 mm² (p = 0.013), so for every 1 unit increase in years from PHV, the CSA of those in the OS group goes up 5.7 mm² more than those in the NOS group. The interaction coefficient for Z was 38.0 mm³ (p = 0.006), for CSMI was 642.0 mm⁴ (p = 0.014) and for stiffness index was 4.0 units (p = 0.023). However, no interaction was found for TBS (p = 0.712).

4. Discussion

The main findings are: (1) OS athletes had greater BMC, HSA estimates, TBS and stiffness index at a given years from PHV compared to NOS athletes; (2) the differences in bone outcomes between OS and NOS groups increase with biological age.

In this study, the OS and NOS groups showed a linear increase in all bone outcomes from −2 to +2 years from PHV, supporting the idea that bone accrual occurs because the remodelling activity is greater than the resorption activity during puberty,[2] and also, due to the influence of sex steroids and body composition changes during puberty and adolescence.[21] For BMC-adjusted outcomes, differences between groups favouring the OS group became evident from −1 year from PHV at hip and FN, and from PHV at TBLH. In this regard, the lack of significant differences at −2 years from PHV might be affected the small sample size of each group at this PHV. The percentage of difference between groups from −2 to +2 years from PHV ranged from 5.7 to 11.3% for TBLH, from 4.8 to 22.9% for hip and from 9.7 to 15.9% for FN. Our results did not show an interaction effect for LS BMC, but significant differences between groups were only observed from −1 year from PHV to PHV, favouring the OS group. In addition, we observed an almost significant trend in the differences between OS and NOS groups at +1 years from PHV (p = 0.052). The trend can be due to the fact that the differences in bone tissue of each bone site are influenced by specific mechanical stimulus induced from the movements of each sport.[10] In our study, the OS group was football, which involves high strains and ground reaction forces applied at lower limbs that receive a greater mechanical load compared to LS site.[22] Besides, a previous study reported that the peak accrual in LS BMC occurs slightly later (approximately +0.7 years from PHV) compared to other sites, such as FN BMC that occurs at +0.5 years from PHV.[4] Based on this, it could also be that the bone accrual at LS may not have occurred at the same pace as the other skeletal sites in the present study.

For comparison and discussion purposes, years from PHV from other investigations has been estimated using validated algorithms for boys and girls (both R = 0.90).[17] In a previous cross-sectional study with adolescent athletes from this cohort we showed that the footballers at −1 year from PHV had 5–7% more TBLH aBMD and 10–12% more hip aBMD compared with swimmers and cyclists at PHV.[9] Another cross-sectional study in adolescent athletes at +2 years from PHV concluded that a NOS group (swimmers) had lower BMC in the total body, lower limbs and LS compared to OS (gymnastics, basketball, and handball).[23] Moreover, adolescent male cyclists at +3 years from PHV showed a 10% lower BMC in the lower limbs compared to an active control group.[24] A cross-sectional study conducted in female swimmers at −1 year from PHV showed 5–17% lower aBMD at FN, pelvis and hip compared to footballers at −1 year from PHV.[8] Similarly, an 8-month longitudinal study[25] comparing female swimmers at +1 year from PHV but footballers at +2 years from PHV showed swimmers had 25.3% lower aBMD at the hip than footballers. These results in NOS groups mostly agree with ours in swimmers and cyclists, who had lower BMC values not only at the hip and FN but also at TBLH compared to the OS group (footballers).
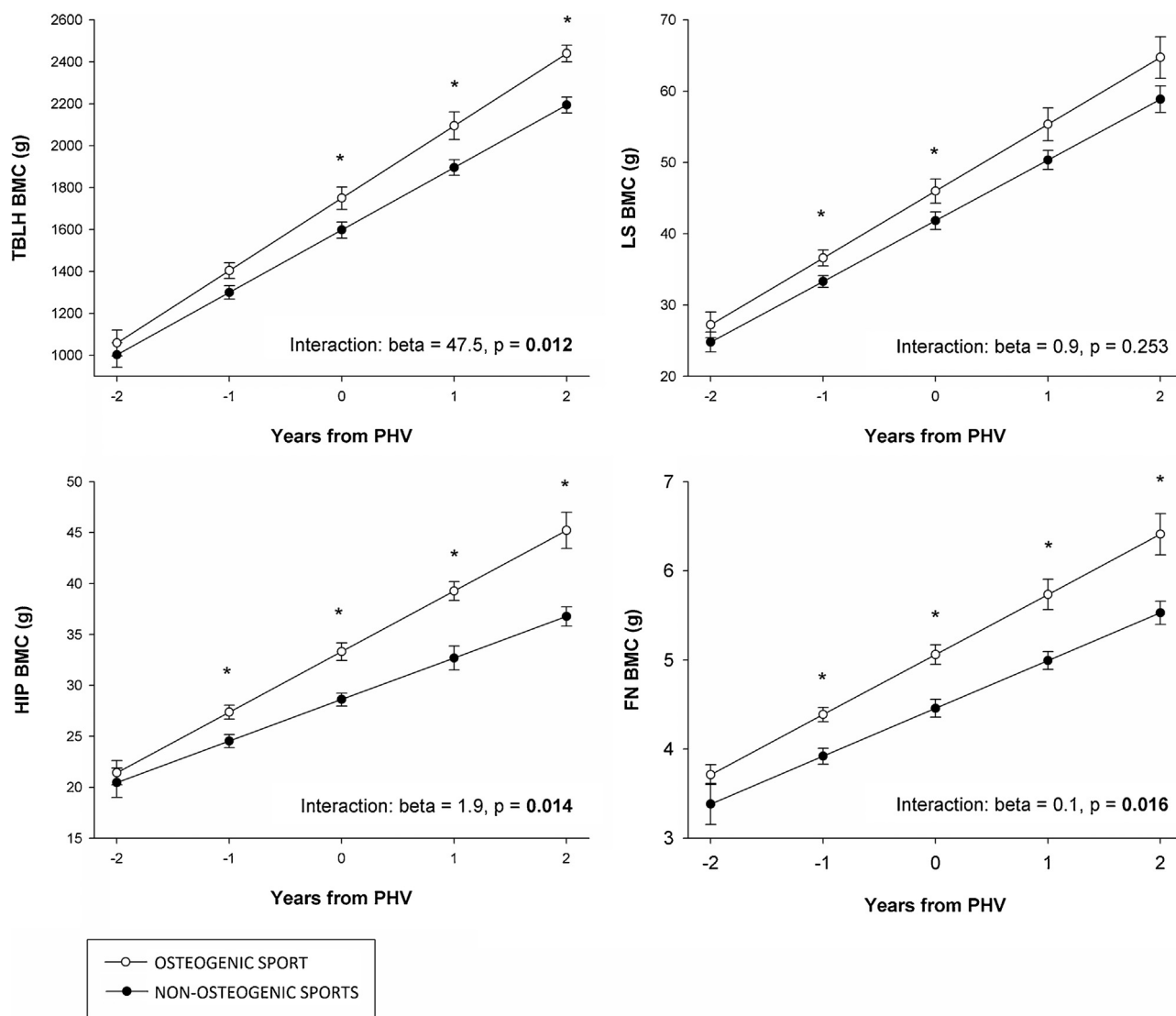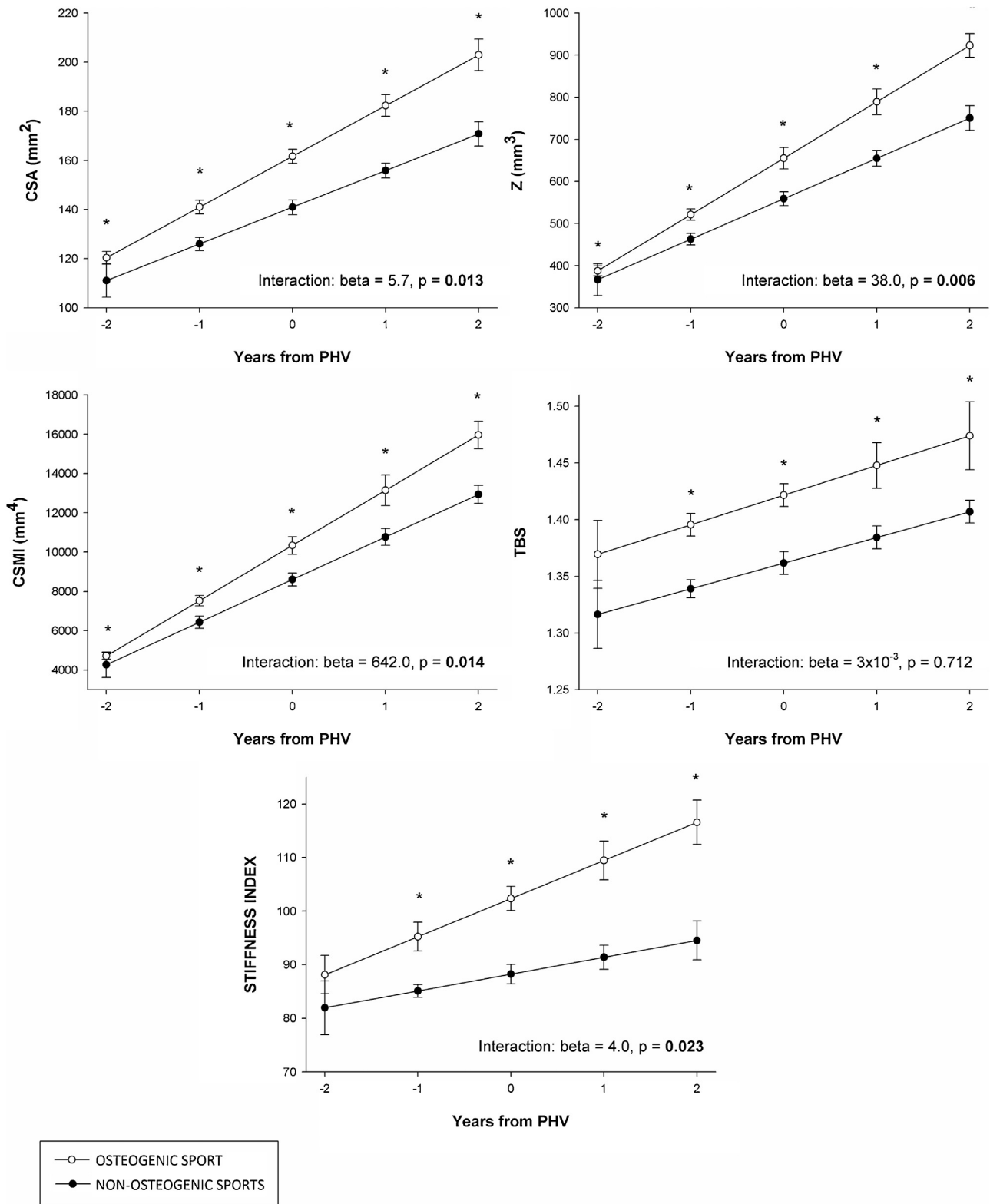
Fig. 1. Bone mineral content (BMC) according to type of sport (osteogenic vs. non-osteogenic) aligned by years from peak high velocity (PHV), where 0 is the PHV. Results (mean and SEM) are adjusted by hours of training. TBLH, total body less head; LS, lumbar spine; FN, femoral neck. Asterisk shows significant differences between type of sports at each biological age category (p < 0.05).

In relation to bone geometry and bone quality, our results are in line with those of a cross-sectional study with this cohort,[9] in which footballers at −1 year from PHV had higher CSA, CSMI, Z and stiffness index (8–21%) compared with swimmers and cyclists at PHV. In the present study, the percentage of difference between groups from −2 to +2 years from PHV ranged from 8.4 to 18.8% for CSA, from 5.6 to 22.9% for Z, from 10.6 to 23.3% for CSMI and from 7.5 to 23.3% for SI. According to a previous review in 10–30 year-old athletes, the adaptations observed in bone geometry outcomes consequence of sports practice are different depending on the type of sport. This is due to the fact that the skeleton is adapted to the load resulting from sport-specific actions.[10] As for the LS BMC, our results did not show an interaction effect for TBS which can be explained by the reasons mentioned above but significant differences between groups were observed from −1 year from PHV. Since TBS assesses DXA images of the LS scans the same reasons as highlighted for LS BMC may explain the lack of interaction. TBS is a novel bone score parameter[26] of bone microarchitectural texture in the LS and little is known about its use in children. A recent cross-sectional study in women, showed that footballers, squash players and power lifters had about 2%, 3% and 4% higher TBS, respectively, compared with swimmers [26]. Sim-

ilar to our findings, a longitudinal study in girls at +4 years from PHV found that CSA in the FN increased more in footballers (3.2% vs. 2.3%) than swimmers after 8 months of sport participation.[25] In other sports, Maïmoun et al. showed that young girls at +1 year from PHV engaged in artistic gymnastics (OS) had greater CSA and Z (20.3% and 21.8%, respectively) compared to swimmers (NOS) at +2 years from PHV.[22] These findings could be extrapolated to our study, in which the practice of OS promotes a higher CSA, CSMI, Z, TBS and stiffness index compared to that of NOS before and after PHV.

A recent meta-analysis found that the differences between swimmers and the athletes of osteogenic sports increased with age.[27] Similarly, our findings show that the difference in BMC outcomes, geometry outcomes and stiffness index between OS and NOS groups increase with biological age, from −2 to +2 years from PHV. This suggests that participation in NOS may affect the acquisition of a high peak bone mass (compared to that of OS) during adolescence. On average, 26% of adult total body BMC is accrued during the 2 years around peak BMC velocity[4] and achieving a high peak bone mass is essential to protect against future bone fractures and diseases.[28] It has also been suggested that sport stimuli dur-

Fig. 2. Hip structural analysis (HSA) of the femoral neck (FN), trabecular bone score (TBS) of the lumbar spine (LS) and stiffness index of the calcaneus according to type of sport (osteogenic vs. non-osteogenic) aligned by years from peak high velocity (PHV), where 0 is the PHV. Results (mean and SEM) are adjusted by hours of training. CSA, cross sectional area; Z, section modulus; CSMI, cross sectional moment of inertia. Asterisk shows significant differences between type of sports at each biological age category ($p < 0.05$).

ing childhood and adolescence may provoke a permanent change on bone metabolism that promotes enhanced accrual throughout growth.[29] Therefore, we suggest the practice of OS during the years surrounding PHV (from −2 to +2), since it is an important period characterised by significant linear growth and BMC accrual[4] in order to contribute to the prevention of osteopenia and/or osteoporosis later in life.

This is the first longitudinal study in male adolescent athletes investigating the differences in bone quantity, bone geometry, bone texture and bone quality between OS (football) and NOS (swimming or cycling) according to biological age. The combination of these techniques provides a thorough insight of bone health during adolescence. To date, the number of studies using TBS in adolescent male population is very limited and further research is needed to better understand its use in young populations. The number of scans in the −2 years from PHV is relatively small and results should be treated with caution. Despite the present study covers the range of −2 to +2 years from PHV, future studies with longer follow-up periods will help to better understand bone changes in response to sport participation throughout adolescence. Our findings allow us to compare male adolescents athletes involved in OS and NOS, and not to compare to a bone status (lack of control group). Future studies in girls are needed as the timing of peak BMC accrual occurs at different periods between sexes,[15,30] and it is influenced by different hormonal status[21] and body composition changes.[30]

5. Conclusion

Participation in OS during adolescence seems to promote a greater improvement in bone quantity (BMC), bone geometry (HSA estimates), bone texture (TBS) and bone quality (stiffness index) compared to the practice of NOS. These findings underline the differential bone response to different sports throughout the years surrounding PHV in male adolescent athletes.

Practical implications

- This study provides evidence that osteogenic sport athletes (football) had better bone health compared to non-osteogenic sport athletes (swimming and cycling) at a given year from PHV.
- Interestingly, the differences increase with biological age, which may have important implications for the achievement of a high peak bone mass in non-osteogenic sport athletes.
- This has been explored by measuring bone quantity, geometry, texture and quality, which adds novelty to this research question.
- From a public health and sports medicine perspective, this is especially important, as football, swimming and cycling are among the most practiced sports worldwide.

Acknowledgements

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.jsams.2018.08.009.

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