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Dimitris Vlachopoulos¹, Alan R. Barker¹, Craig A. Williams¹,
Sigurbjörn A. Arngrímsson³, Karen M. Knapp^{5,6}, Brad S. Metcalf¹,
Ioannis G. Fatouros⁴, Luis A. Moreno², and Luis Gracia-Marco^{1,2}

¹Children's Health and Exercise Research Centre. Sport and Health Sciences, University of Exeter, Exeter, United Kingdom; ²Growth, Exercise, Nutrition and Development Research Group, University of Zaragoza, Zaragoza, Spain; ³Center for Sport and Health Sciences, University of Iceland, Laugarvatn, Iceland; ⁴Department of Kinesiology, Institute for Research and Technology, Physical Education and Sport Sciences, University of Thessaly, Trikala, Greece; ⁵Department of Medical Imaging, College of Engineering, Mathematics and Physical Sciences, University of Exeter, Exeter, United Kingdom; ⁶University of Exeter Medical School, Exeter, United Kingdom

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¹Children's Health and Exercise Research Centre. Sport and Health Sciences, University of Exeter, Exeter, United Kingdom; ²Growth, Exercise, Nutrition and Development Research Group, University of Zaragoza, Zaragoza, Spain; ³Center for Sport and Health Sciences, University of Iceland, Laugarvatn, Iceland; ⁴Department of Kinesiology, Institute for Research and Technology, Physical Education and Sport Sciences, University of Thessaly, Trikala, Greece; ⁵Department of Medical Imaging, College of Engineering, Mathematics and Physical Sciences, University of Exeter, Exeter, United Kingdom; ⁶University of Exeter Medical School, Exeter, United Kingdom

Address for correspondence: Dimitris Vlachopoulos, Children's Health and Exercise Research Centre, Sport and Health Sciences, College of Life and Environmental Sciences, University of Exeter, St. Luke's Campus, Exeter, EX1 2LU, United Kingdom. Email: dv231@exeter.ac.uk

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ABSTRACT

Purpose: Exercise is an effective approach for developing bone mass and adolescence is a key period to optimize bone health. However, sports specific training may have different effects on bone outcomes. This study examined the differences on bone outcomes between osteogenic (football) and non-osteogenic (swimming and cycling) sports and a control group in adolescent males. **Methods:** One hundred twenty one males (13.1 ± 0.1 years) were measured: 41 swimmers, 37 footballers, 29 cyclists and 14 controls. Dual energy X-ray absorptiometry measured bone mineral density (BMD) and content (BMC) at lumbar spine, right and left hip and total body. Hip structural analysis evaluated bone geometry at the femoral neck. Quantitative ultrasound evaluated bone stiffness at both feet. **Results:** Footballers had significantly higher BMD at total body less head (7-9 %), total hip (12-21 %) and legs (7-11 %) compared to all groups and significantly higher BMD at the femoral neck than controls (14 %). Cyclists had higher BMD at the trochanter (10 %) and BMC at the arms (10 %) compared to controls. Geometrical analysis showed that footballers had significantly higher cross-sectional area (8-19 %) compared to all groups, cross-sectional moment of inertia (17 %) compared to controls and section modulus compared to cyclists (11 %) and controls (21 %). Footballers had significantly higher bone stiffness compared to all groups (10-20 %) at the dominant foot and (12-13 %) at the non-dominant foot compared to swimmers and controls. **Conclusions:** Adolescent male footballers exhibited higher bone density, geometry and stiffness compared to swimmers, cyclists and controls. Although swimmers and cyclists had higher bone outcomes compared to controls, these differences were not significant.

Keywords: ADOLESCENCE, BONE MASS, BONE GEOMETRY, BONE STIFFNESS, EXERCISE.

INTRODUCTION

Osteoporosis is a disease characterized by reduced bone mass and deterioration of bone microarchitecture, resulting in increased risk of fragility fractures. Bone mass acquisition during adolescence is not only an important determinant of skeletal growth but also for reducing the risk of osteoporosis later in life (20). In this regard, a 10 % increase in peak bone mass during adolescence might reduce the risk of fracture later in life by 50 % and delay the onset of osteoporosis by 13 years (27). Therefore, early prevention remains one of the most prudent approaches to improve bone health status in later adult life.

It is known that 20 % of the variation in peak bone mass can be explained by lifestyle factors, including physical activity (PA) and diet (i.e. calcium and vitamin D intakes) (18, 36). In terms of PA, a favourable osteogenic response can be obtained when high-impact, intensive and weight-bearing exercise is performed, due to the mechanical load imposed on the bone tissue (14). For example, football is considered an “osteogenic” sport and augments bone mineral density (BMD) and content (BMC) at the weight-bearing sites in early and late pubertal males (19). In contrast, sports such as swimming and cycling have been considered “non-osteogenic” (33), although the supporting evidence is unclear. Previous evidence found that adolescent male swimmers to have lower adjusted BMC and BMD compared to controls (10). A recent systematic review concluded that swimmers have similar bone mass with sedentary controls (11). Similarly, although there are reports of cycling showing no effect on bone-related outcomes in adolescents, some studies suggest cycling during adolescence may negatively impact bone health and compromise the acquisition of a high peak bone mass (22). There is limited evidence evaluating the effects of osteogenic and non-osteogenic sports on bone outcomes in adolescent males and further research is needed to investigate this discrepancy in the literature.

With football, swimming, and cycling among the most popular sports during childhood and adolescence in the United Kingdom, understanding the contribution of these sports to bone health is important. To date, studies evaluating bone-related outcomes in athletic groups have mainly focused in BMD and BMC outcomes provided by Dual energy X-ray Absorptiometry (DXA). But a more comprehensive evaluation of bone structure, as well researched can be obtained using the Hip Structural Analysis (HSA) software from DXA (3). The parameters obtained from HSA software reflect bone strength at the narrow neck site of the clinical important site of the hip. A previous study showed that adolescent female footballers had greater hip strength compared to swimmers and controls, while swimmers had lower bone mass at the narrow neck than footballers and controls (9). Another method to assess bone properties is Quantitative Ultrasound (QUS), which is a non-radiation technique and provides measurements of the bone stiffness changes at the calcaneus site. Currently, there are no studies evaluating bone outcomes in male adolescent athletes using a combination of DXA, HSA and QUS outcomes. Furthermore, there is a lack of consistency when controlling for the use of confounding variables in the assessment of bone outcomes in youth sports. This is important as uncritical use of confounders can lead to size related artefacts (25). Previous studies typically use confounders such as age, height, weight, calcium intake, fat mass, fat-free mass and lean mass (17, 39). However, the most common inconsistencies observed in many studies are the lack of consideration for size adjustments in adolescent participants and the lack of site specific adjustment of the skeletal outcomes. Therefore, more studies are needed to assess the bone outcomes by taking into account the relevant confounders according to participant characteristics.

The PRO-BONE (effect of a PROgram of short bouts of exercise on BONE health in adolescents involved in different sports) study was designed to investigate whether the bone properties, assessed by DXA, HSA and QUS, differ between 12-14 year old males who perform osteogenic (football) and non-osteogenic (swimming, cycling) sports in comparison to a control group after controlling for a comprehensive set of confounders. We hypothesised that adolescent males engaged in football will have higher bone outcomes compared to those engaged in cycling and swimming and compared to a control group, and that adolescent males engaged in cycling and swimming will have similar bone outcomes.

METHODS

Study design and participants

The study represents a cross-sectional analysis of the baseline data derived from the PRO-BONE study, which is a 33 month longitudinal design including a 9-month jump intervention programme. The purpose, methodology and sample size of the PRO-BONE study have been justified elsewhere (35). Data were collected between autumn and winter 2014/15 in 121 adolescent males: 41 swimmers, 37 footballers, 29 cyclists and 14 controls. The inclusion and exclusion criteria were: 1) males 12–14 years old, engaged (≥ 3 h/week) in osteogenic (football) and/or non-osteogenic (swimming and cycling) sports for the last 3 years or more; 2) males 12–14 years old not engaged in any of these sports (≥ 3 h/week) in the last 3 or more years (control group); 3) participants not taking part in another clinical trial; 4) participants not having any acute infection lasting until < 1 week before inclusion; 5) participants had to be free of any

medical history of diseases or medications affecting bone metabolism or the presence of an injury; 6) white Caucasian ethnicity.

Participants were recruited from athletic clubs and schools across the South West of England. Written informed consent and assent forms were signed from parents and participants accordingly and all participants completed the first visit at the research centre as part of the study. The methods and procedures of the study have been checked and approved by: 1) the Ethics Review Sector of Directorate-General of Research (European Commission, ref. number 618496); 2) the Sport and Health Sciences Ethics Committee (University of Exeter, ref. number 2014/766) and 3) the National Research Ethics Service Committee (NRES Committee South West – Cornwall & Plymouth, ref. number 14/SW/0060).

Anthropometry and sexual maturity

Stature (cm) and body mass (kg) were measured by using a stadiometer (Harpenden, Holtain Ltd, Crymych, UK; precision 0.1 cm; range 60–210 cm) and an electronic scale (Seca 877, Seca Ltd, Birmingham, UK; precision 0.1 kg; range 2–200 kg) respectively. Body mass index was calculated as body mass (kg) divided by the stature (m) squared. Sexual maturation was self-reported using adapted drawings of the five stages (Tanner) of pubic hair (30).

Dual energy x-ray absorptiometry

A DXA scanner (GE Lunar Prodigy Healthcare Corp., Madison, WI, USA) was used to measure BMD (g/cm^2), BMC (g), bone area (BA, cm^2), fat mass (g) and lean mass (g). Four scans were performed to obtain data for the lumbar spine (LS, L1-L4), right and left hip (including femoral neck, Ward's triangle, trochanter and shaft sub-regions; the mean of right and left hip scans was

used), and the total body scan. The total body scan was then used to obtain data for specific regions such as: arms, legs, pelvis and total body excluding head. All DXA scans and subsequent in-software analyses were completed by the same researcher, using the same DXA scanner and the GE encore software (2006, version 14.10.022). The positioning of the participants and the analyses of the results were undertaken according to International Society of Clinical Densitometry (4).

Hip structural analysis

Using the HSA software, analyses were performed at the narrow neck region across the narrowest point of the femoral neck. The HSA programme uses the distribution of bone mineral mass in line of pixels across the bone axis to measure the structural dimensions of bone cross sections (3). The geometric properties of the bone were obtained and the following variables used: 1) the cortical width neck (mm), which is the narrowest width of the femoral neck; 2) the diameter of the femoral neck (mm); 3) the cross sectional area (CSA, mm³), which is the total bone surface area excluding the soft tissue area and the trabecular; 4) the cross-sectional moment of inertia (CSMI, mm⁴), which is an index of structural rigidity and reflects the distribution of mass in the centre of a structural element; 5) section modulus (mm³), which is an indicator of maximum bending strength in a cross section; and 6) the hip strength index, which is an advanced feature that has been added to the more recent versions of GE enCore software and indicates the risk of fracture forces generated during a fall on the greater trochanter and the CSA short term precision percentage coefficient of variation has been reported to be between 2.4 % and 7.9 % (16).

Quantitative ultrasound

QUS measurements were performed with a Lunar Achilles Insight (TM Insight GE Healthcare, Milwaukee, WI, USA) and the OsteoReport PC (software version 5.x+). The stiffness index is then calculated by a linear combination of broadband ultrasound attenuation (BUA) and speed of sound (SOS) as follows: $\text{Stiffness index} = (0.67 \times \text{BUA}) + (0.28 \times \text{SOS}) - 420$. Both feet were measured twice and the mean of the two measures was used for statistical analyses of the dominant and non-dominant foot. For the purpose of this study only stiffness index values were used. QUS is considered a valid and radiation-free method compared to DXA to assess bone health in children (2).

Physical activity and diet

PA was measured for seven consecutive days by using wrist accelerometers (GENEAActiv, GENEActiv, UK). The validity and reliability of the accelerometer has been established previously in children and adolescents (24). Participants were instructed to place the accelerometer on their non-dominant wrist and data was collected at 100 Hz. Data were analysed at 1 s epoch intervals to establish time spent in different intensities. Time spent in moderate PA and vigorous PA (VPA) was calculated using a cut-off point of 1140-3599 counts per minute and ≥ 3600 counts per minute, respectively (24). Moderate-to-vigorous PA (MVPA) was calculated using a cut-off point of ≥ 1140 counts per minute. Weekly training hours were obtained by face to face questions during the visit of the participants at the research centre.

Dietary calcium, vitamin D and energy intake were assessed using a 24 hour food recall. The validity and reliability of self-reported dietary intake has been previously reported in children

(37). Total energy, calcium and vitamin D intake were estimated using the CompEat Pro software (Nutrition systems, VIS Visual Information Systems Ltd., UK).

Statistical analysis

Statistical analyses were performed using the SPSS IBM statistics (version 21.0 for Windows, Chicago, IL, USA) and descriptive data are reported as mean and SD. The distribution of the variables was checked and verified using Shapiro-Wilk's test, skewness and kurtosis values, visual check of histograms, Q-Q and box plots. The analysis of the data was completed in two stages: 1) raw (unadjusted) data using one-way analysis of variance (ANOVA) with Bonferroni post hoc to detect between-group differences on bone-related outcomes (DXA, HSA and QUS), and 2) adjusted data using one-way analysis of covariance (ANCOVA) with Bonferroni post hoc taking into account the following relevant confounders: age, stature, region-specific lean mass (trunk, total body, arms and legs), calcium intake and MVPA (12, 13, 32, 37). A preliminary analysis showed maturation to have no effect on bone outcomes after accounting for age and thus was not included in the model. Percentages of difference between groups for all variables were used to quantify the magnitude of the differences. Statistical significance level was set at $P < 0.05$ and differences of $P < 0.001$ were also indicated.

RESULTS

Descriptive characteristics of the study sample

Table 1 presents the descriptive characteristics of the participants. Swimmers were older, taller, heavier and had more lean mass than the footballers. Footballers spent more time doing MVPA

and VPA than swimmers and controls. Cyclists were older and spent more time doing VPA than controls and they also spent more time doing MVPA and VPA than the swimmers. In addition, swimmers and footballers trained more hours on average than the cyclists. Finally, controls had more fat mass than all the other groups.

DXA region-specific BMD, BMC and BA

Table 2 shows the raw differences for the four groups at different sites. Controls had significantly lower BMD and BMC compared with footballers (BMD: ranged from 6.7 % to 30.1 %, BMC: ranged from 18.1 % to 52.4 %), swimmers (BMD: 10.9 % to 17.9 %, BMC: 26.7 % to 57.1 %) and cyclists (BMD: 8.3 % to 17.9 %, BMC: 21.0 % to 40.9 %) for all sites except for the lumbar spine and arms. In addition, controls had significantly lower BA compared to swimmers (BA: 11.6 % to 37.8 %). Footballers had 7.5 %, 10.4 % and 10.1 % significantly higher BMD at total hip, trochanter and Ward's triangle sites than the swimmers. In addition, they had 7.8 %, 10.4 % and 10.4 % significantly higher BMD at total hip, trochanter and Ward's triangle sites than the cyclists. Finally, swimmers had 6.1 % significantly higher BMD and 23.1 % BMC than footballers at the arms as well as 8.9 %, 9.9 % and 17.7 % greater BA at the shaft, lumbar spine and arms, respectively.

Figures 1, 2, and supplementary table 1 (see Table, SDC 1, adjusted data for DXA region-specific BMD, BMC and BA, <http://links.lww.com/MSS/A756>) present adjusted differences for the sports groups at different sites compared to the control group. Once the confounders were controlled for, differences remained significant and higher mainly in the football group compared to the other groups. More specifically, footballers had significantly higher BMD (8.8 % to 25.1 %) and BMC (7.9 % to 29.5 %) than controls at all sites except for the lumbar spine and arms. In

addition, footballers had significantly higher BMD and BMC at all sites except for the lumbar spine and arms than swimmers (BMD: 6.9 % to 13.9 %, BMC: 8.4 % to 20.5 %) and cyclists (BMD: 5.2 % to 12.7 %, BMC: 6.7 % to 18.9 %). BA of footballers was significantly higher at pelvis site compared to the other groups (7.1 % to 8.9 %). Cyclists had 10.3 % significantly higher BMD only at the trochanter, 9.8 % higher BMC and 7.3 % higher BA only at the arms compared to controls. There was no significant difference in the other skeletal sites between cyclists and controls. However, cyclists had non-significant higher bone outcomes (BMD: 3.4 % to 11.0 %, BMC: 1.1 % to 11.8 %) in the most sites of the skeleton. At lumbar spine cyclist had non-significant lower BMC (-1.9 %) compared to controls. No significant difference were found between swimmers and controls at any skeletal sites. However, swimmers had non-significant higher bone outcomes in most skeletal sites (BMD: 0.3 % to 9.7 %, BMC: 0.8 % to 10.8 %). At the lumbar spine swimmers had non-significant lower bone outcomes (BMD: -0.8 %, BMC: -4.6 %) compared to controls. Cyclists and swimmers had similar BMD, BMC and BA (-0.9 % to 5.0 %) with no significant differences at any skeletal site.

Bone geometry - Hip Structural Analysis

The adjusted geometrical differences in narrow neck site between the groups are presented in Figure 3 and the raw and adjusted values are presented in supplementary table 2 (see Table, SDC 2, raw and adjusted data for HSA and QUS, <http://links.lww.com/MSS/A757>). Footballers had a significantly higher CSMI than controls (17.4 %), greater section modulus than cyclists (10.7 %) and controls (21.0 %), significantly higher CSA than swimmers (10.8 %), cyclists (8.7 %) and controls (19.3 %) and a significantly greater hip strength index than swimmers (20.7 %) and controls (38.9 %). Cyclists had only a significantly higher hip strength index compared to

controls (28.6 %). Cyclists had non-significant higher geometrical outcomes compared to controls (CSMI: 6.4 %, Section modulus: 9.3 %, CSA: 9.8 %). Swimmers had non-significant higher geometrical outcomes compared to controls (CSMI: 7.8 %, Section modulus: 10.9 %, CSA: 7.6 %, hip strength index: 15.1 %). Cyclists compared to swimmers had similar geometrical outcomes with minimal differences.

Quantitative ultrasound

The adjusted bone stiffness values of the dominant and non-dominant foot are presented in Figure 4 and the raw differences are presented at supplementary table 2, <http://links.lww.com/MSS/A757>. Footballers had significantly higher bone stiffness in the dominant foot than swimmers (13.4 %), cyclists (10.3 %) and controls (20.1 %). In addition, footballers had significantly greater bone stiffness than swimmers (12.2 %) and controls (12.9 %) at the non-dominant foot. No significant differences were found between dominant vs. non-dominant foot within each group of participants. Cyclists had higher (non-significant) stiffness index compared to controls in both dominant (8.9 %) and non-dominant foot (5.3 %). Swimmers had higher (non-significant) bone stiffness at the dominant (5.9 %) and the non-dominant (0.7 %) foot. Cyclists compared to swimmers had higher (non-significant) bone stiffness at the dominant (2.7 %) and the non-dominant (4.4 %) foot.

DISCUSSION

The key findings from this study are: 1) footballers presented greater adjusted BMD and BMC including clinical relevant sites, an enhanced hip structural geometry at the narrow neck and a

greater bone stiffness index compared to swimmers, cyclists and controls, and 2) swimmers and cyclists had similar bone mass, geometry and bone stiffness and both groups had higher but not significant bone outcomes compared to controls. The impact of osteogenic (football) and non-osteogenic sports (swimming and cycling) on bone-related outcomes has not previously been compared in adolescent male athletes and there is equivocal evidence on the effects of these sports on bone outcomes (10, 22, 34). To date, there are no studies published using a combination of methods such as DXA, HSA and QUS to assess bone outcomes in this population and there are a lack of studies taking into consideration the relevant confounders based on the characteristics of the groups studied. The findings of the present study therefore provide a more comprehensive assessment into the effect of sports participation on bone outcomes in male adolescents.

Bone outcomes in footballers vs controls

Participation in osteogenic sports during adolescence can induce greater adjusted BMD compared to leisure active controls at many sites of the skeleton due to the mechanical loading applied (17). A previous study reported a 10.7 % and 10.5 % higher adjusted BMC at the total hip and lumbar spine respectively, in prepubescent male football players (n= 39) compared to active controls (n= 13) (40). The magnitude of the differences might differ among studies due to the use of different confounders and the characteristics of the participants.

In parallel with the findings for BMD and BMC, the geometrical adaptations examined by HSA at the narrow neck of the femoral neck also supported the higher bone geometry in footballers (Figure 3). One study in oligoamenorrhic female athletes showed that engagement in weight-bearing sports for 4 hours per week resulted in significantly higher CSMI and section modulus

compared to non-athletes (1), which is consistent with the improved structural rigidity we found in footballers.

Previous studies using QUS technique observed positive associations between PA and calcaneal bone stiffness index in a sample of Flemish children and adolescents (5). Our results are in agreement with a study reporting that child and adolescent football players have significantly higher QUS parameters at lower extremities compared with age matched controls (7).

Bone outcomes in swimmers vs controls

A recent meta-analysis of fourteen studies summarized that swimming does not induce improvements in BMD during childhood and adolescence and that swimmers present similar BMD compared to sedentary controls (11). We found similar BMD and BMC between swimmers and active controls concurs with this meta-analysis, which presents neutral effects of swimming on BMD and BMC at most skeletal sites of the skeleton compared to active controls . The latter could be due to the fact that swimmers and controls have similar bone profile as muscle contraction is not enough to produce bone adaptations (17).

The HSA at the narrow neck site showed that adolescent male swimmers have similar bone geometry parameters compared to active controls. To our knowledge there is no previous evidence using the HSA technique in adolescent male swimmers. Only one study have used HSA in elite adolescent female swimmers and showed that they had similar bone geometry compared with controls (17). The latter study highlighted the importance of lean body mass as it was highly correlated with CSA and hip strength index.

Regarding QUS, we found similar bone stiffness index in both dominant and non-dominant foot of swimmers compared to controls. A previous study in adolescents reported similar QUS parameters between swimmers and controls and indicated that bone adaptations due to swimming might be counterbalanced by other weight-bearing activities (up to 3 hours per week) (10), however this cannot be the case in our study because the QUS parameters were controlled for MVPA.

Bone outcomes in cyclists vs controls

A systematic review revealed that road cyclists did not have any osteogenic benefits due to the non-mechanical loading character of the sport (23). A previous study conducted in adolescent female cyclists showed they had similar BMD compared to non-athletic controls after adjusting for years since menarche, lean mass and sport specific training (6). According to our study, the skeletal differences between cyclists and active controls are site dependent and more specifically we found significantly greater BMC at the arms after controlling for region-specific lean mass and MVPA among other confounding factors.

There is no previous evidence using HSA technique in adolescent cyclists and only a few studies used volumetric bone parameters. One study conducted in adolescent female cyclists found no significant differences in CSA and CSMI in cyclists compared to controls (6) which is in accordance with our study. However, it should be noted that in our study the HSA revealed cyclists had significantly higher hip strength index than controls (28.6 %), something that was not observed with BMD and BMC at any of the hip variables analysed with DXA. This might be explained by the fact that geometrical bone outcomes may differ from BMD and BMC when using DXA.

The effect of cycling on bone properties using QUS has not been previously evaluated and to the best of our knowledge this study is first that provides evidence for this population. We did not find differences on stiffness index between cyclists and controls, but cyclist had non-significant higher stiffness index in both the dominant (8.9 %) and non-dominant (5.3 %) foot compared to controls. Our results support the findings of previous studies that the loading pattern of sports participation may influence the bone stiffness index in adolescents (8).

Comparison of bone outcomes between footballers, swimmers and cyclists

In the present study the comparison between osteogenic (football) and non-osteogenic sports (cycling and swimming) showed that adolescent male footballers had significantly greater adjusted BMD and BMC compared to swimmers and cyclists at all sites of the skeleton except for the lumbar spine and the arms. A previous study in athletic adolescent females reported that 3 hours per week of football participation induced greater improvements in height and lean mass adjusted BMD and BMC compared to swimmers at femoral neck and other sites of the skeleton (32). Only one study investigated the effects on bone mass between adolescent male footballers and swimmers and reported greater BMD at the femoral neck site in the footballers (28). To our knowledge, no previous evidence exists on the assessment of bone mass between footballers and cyclists in adolescents. Only one study in children reported positive associations between BMD and football participation and negative associations found between BMD and cycling participation (29).

In our study there was no significant difference observed in BMD and BMC between adolescent male swimmers and cyclists at any site of the skeleton. A recent review summarised the impact of sport participation on peak bone mass and it reported that both swimming and cycling may not

be associated with significant improvements in bone health (31). The comparison of BMD and BMC between swimmers and cyclists has been assessed only once in female adolescents, reporting similar values at all skeletal sites after taking into account potential confounders (6). The differences observed in the current study are likely to be explained by the non weight-bearing environment of both swimming and cycling and by the mechanical loading of the skeleton according to the impact of produced by the sport specific patterns. In addition, weight training and the plyometric exercises might induce higher bone mass in adolescent athletes (15). A study in adolescent swimmers has shown that participation of adolescent swimmers in other weight-bearing sports or activities involving plyometric exercises can induce higher BMD and BMC (10). In our study, a subsample of our participants has been asked about weight training and we have shown that almost all footballers were involved in plyometric exercise training. A large number (70.7 %) of the swimmers reported participation in plyometric training, but only a few cyclists (37.9 %) were doing plyometric exercises. The participation in plyometric training or other weight-bearing activities might explain the difference on bone outcomes between adolescent athletes and needs further investigation to quantify the impact of weight training on bone outcomes.

In parallel with the BMD and BMC findings, the bone geometry evaluated by HSA at the narrow neck of the femoral neck was also higher in footballers compared to swimmers and cyclists. Previous research in adolescent female footballers and swimmers showed that the CSA area and section modulus were significantly higher in footballers compared to swimmers at the narrow neck site which is in agreement with our results (9). There is no evidence comparing football and cycling in children and adolescents and the only evidence exists in young females (21 - 28 years)

which found that footballers had approximately 10 % higher CSA compared to cyclists and after adjusting for age, weight and height, these results are in accordance with our findings (21).

In relation to QUS parameters we found improved bone stiffness in footballers compared to cyclists and swimmers at the dominant foot and higher bone stiffness in footballers compared to swimmers at the non-dominant foot. As there is no evidence using QUS in similar age athletic groups, we identified a study in young adults (18-22 years) that reported higher bone stiffness in footballers compared to swimmers at the dominant and non-dominant heels (38) which complies with the findings of the present study.

The strengths of the current study are 1) the investigation of bone outcomes across three male adolescent athletic groups that were not compared before; 2) the combination of DXA, HSA software and QUS outcomes which provides a thorough insight of the differences in BMD, BMC, bone geometry and bone stiffness; 3) the rigorous methodology and strong internal validity to control for specific confounders. It should be noted that the limitation of the cross-sectional study precludes any determination of causality in our findings. Nevertheless, our population had strict age inclusion criteria and sport participation characteristics. The limitations of the self-reported maturation assessment should be noted. Most of the participants of our control group met the physical activity guidelines due to inclusion criteria used, but we know that most adolescents of this age do not meet the guidelines (26). However, in this specific case, it seems reasonable to propose that sport participation could have different effects on bone-related outcomes depending on the characteristics of the sport practiced. Therefore, more studies are needed to focus on the determinants affecting bone health for athletic groups during youth.

CONCLUSIONS

This study is the first to investigate the impact of weight-bearing (football) and non weight-bearing sports on bone outcomes in adolescent males. The findings of this study indicate that participation in weight-bearing sports, such as football, can induce greater improvements in bone mass, bone geometry and stiffness index compared to non weight-bearing sports, such as swimming and cycling and compared to controls. Swimmers and cyclists had similar bone outcomes and both groups had higher bone outcomes compared to controls, but these differences were not statistically significant. These findings add to the sport participation recommendations that specific musculoskeletal training may affect the bone development during adolescence. Further longitudinal analyses of the specific sports are needed for this population in order to identify if these effects will be different after a longer period of sports participation.

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Authors' Contributions

DV obtained and analysed the data and drafted the manuscript under the supervision of LGM (principal investigator), ARB and CAW. BSM, KMK, SAA, IGF and LAM reviewed the draft. All authors have read and approved this work.

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Ethics approval and consent to participate

Ethics approval received from the following committees: 1) the Ethics Review Sector of Directorate-General of Research (European Commission, ref. number 618496); 2) the Sport and Health Sciences Ethics Committee (University of Exeter, ref. number 2014/766) and 3) the National Research Ethics Service Committee (NRES Committee South West – Cornwall & Plymouth, ref. number 14/SW/0060).

Conflict of interest

The authors declare that they have no competing interests.

The results of the study are presented clearly, honestly, and without fabrication, falsification, or inappropriate data manipulation, and the present study do not constitute endorsement by ACSM.

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Figure captions

Figure 1. Difference (%) in adjusted bone mineral density (BMD), content (BMC) and bone area (BA) between the sports groups and controls at the total body less head and hip sites. Letters denote a significant difference with: a (swimmers), b (footballers), c (cyclists) and d (controls). a,b,c,d $p<0.05$ and aa,bb,cc,dd $p<0.001$.

Figure 2. Difference (%) in adjusted bone mineral density (BMD), content (BMC) and bone area (BA) between the sports groups and controls at the lumbar spine, pelvis, arms, and legs. Letters denote a higher significant difference with: a (swimmers), b (footballers), c (cyclists) and d (controls). a,b,c,d $p<0.05$ and aa,bb,cc,dd $p<0.001$.

Figure 3. Percentage of difference in adjusted geometrical parameters of the narrow neck site between groups. Neck Width (mm), CSA: Cross sectional area, CSMI: Cross sectional moment of inertia, FN: femoral neck. * $p<0.05$ and ** $p<0.001$.

Figure 4. Difference (%) in adjusted stiffness index (SI) (mean \pm SE) between the sports groups and the controls at the dominant and non-dominant foot. Letters denote a higher significant difference with: a (swimmers), c (cyclists) and d (controls). a,b,c,d $p<0.05$ and aa,bb,cc,dd $p<0.001$. No differences observed between dominant and non-dominant foot at the groups.

Supplemental Digital Content files

Supplementary digital content table 1.docx Adjusted data for DXA region-specific bone mineral content (BMC, g), density (BMD, g/cm^2) and area (BA, cm^2) of all participants

Supplementary digital content table 2.docx Raw and adjusted data for Hip Structural Analysis (HSA) and Quantitative Ultrasound (QUS) parameters of all participants

Figure 1

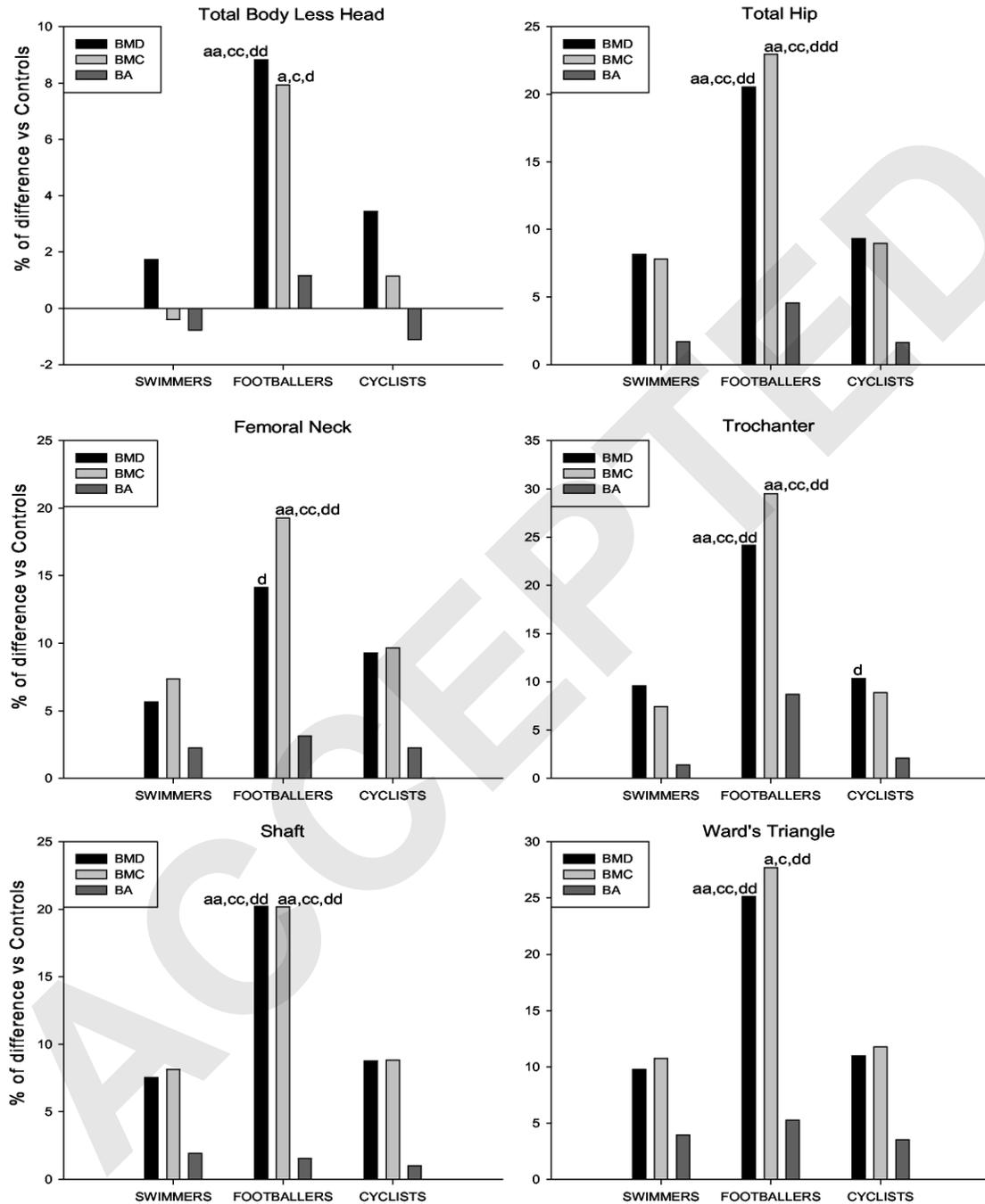


Figure 2

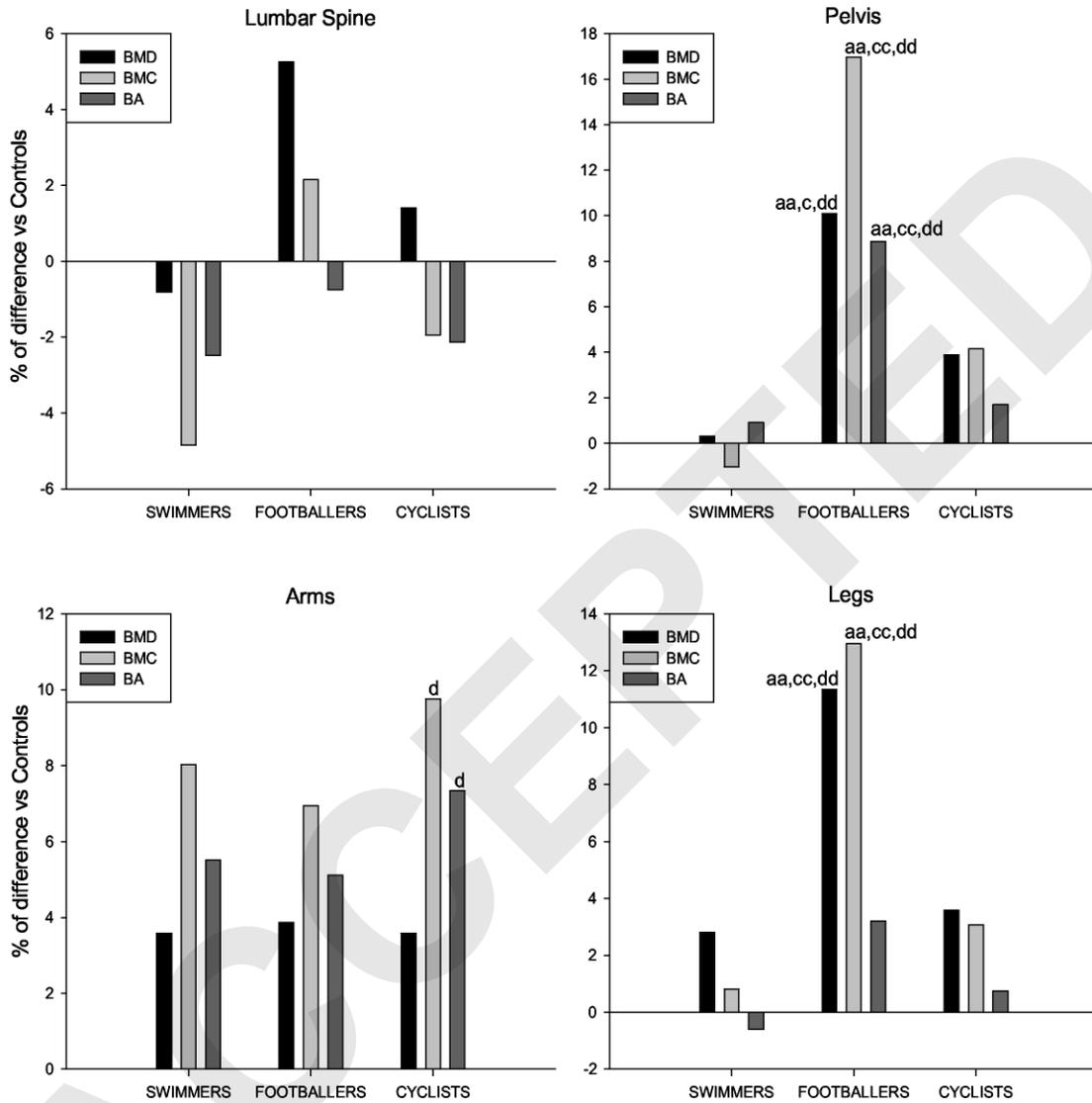
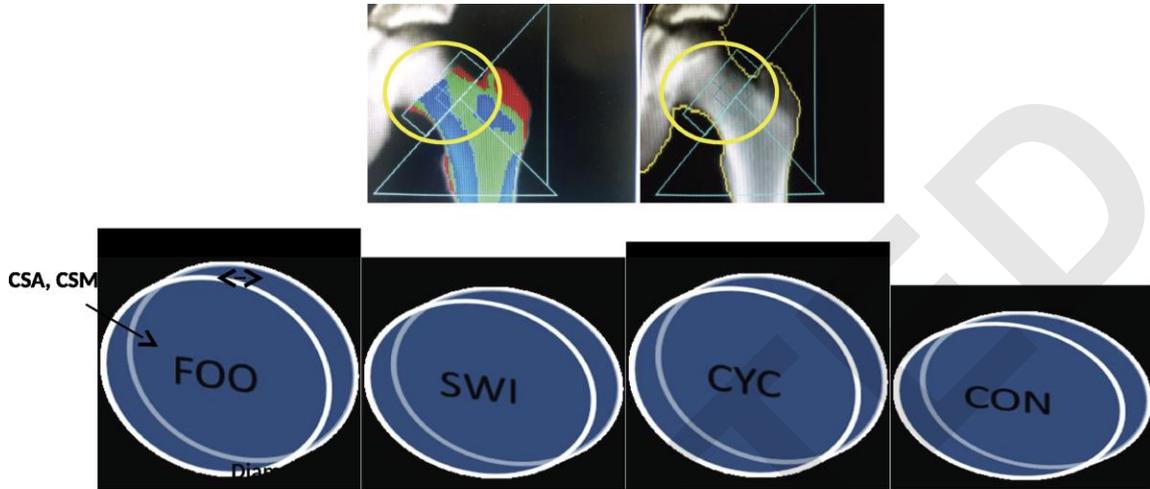


Figure 3



	FOO vs SWI	FOO vs CYC	FOO vs CON	SWI vs CYC	SWI vs CON	CYC vs CON
Neck Width	10.0%	3.1%	13.8%	-6.3%	3.5%	10.3%
Diameter FN	0.7%	1.0%	3.0%	0.3%	2.3%	2.0%
CSMI	9.0%	10.4%	17.4%*	1.4%	7.8%	6.4%
Section modulus (Z)	9.2%	10.7%*	21.0%**	1.5%	10.9%	9.3%
CSA	10.8%**	8.7%*	19.3%**	-1.9%	7.6%	9.8%
Hip strength index	20.7%*	8.0%	38.4%**	-10.5%	15.1%	28.6%*

Figure 4

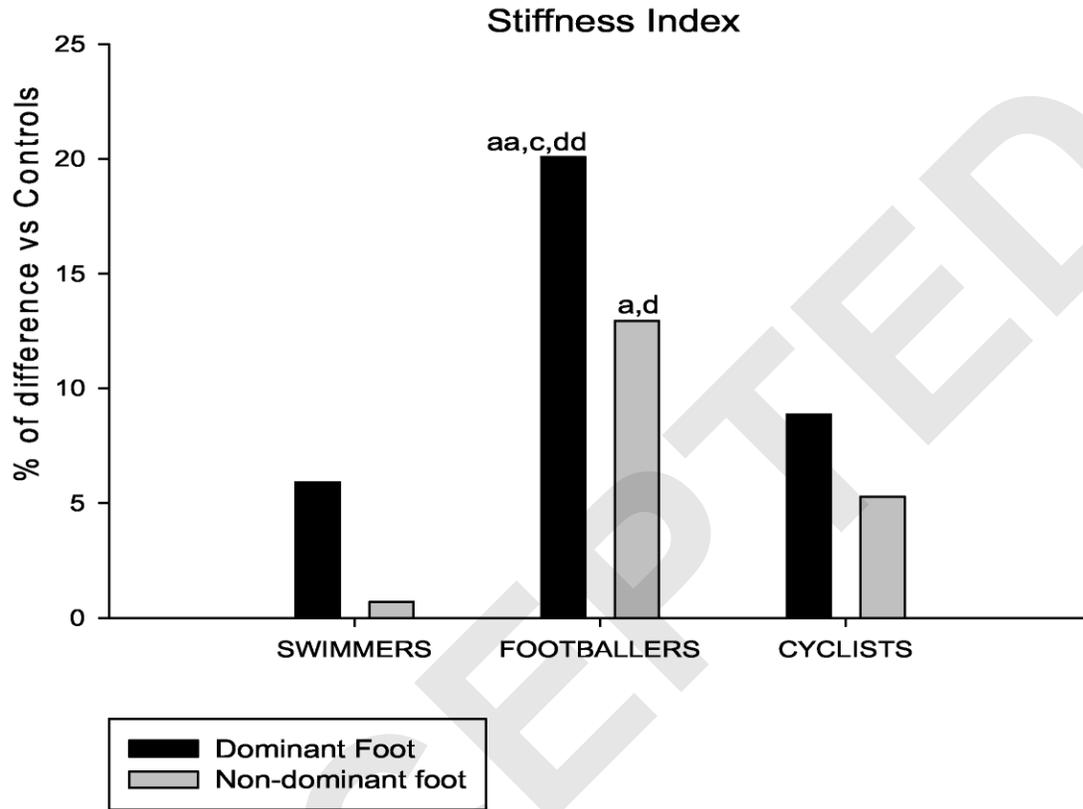


Table 1. Descriptive characteristics of the participants

Parameters	Swimmers (n=41)	Footballers (n=37)	Cyclists (n=29)	Controls (n=14)
Age (yrs)	13.4 (1.0) ^{b,dd}	12.8 (0.9)	13.2 (1.0) ^d	12.3 (0.5)
Stature (cm)	165.5 (9.7) ^{bb,d}	155.2 (9.3)	160.8 (9.9)	154.5 (9.9)
Body mass (kg)	52.4 (9.0) ^{bb}	44.3 (7.9)	49.5 (12.3)	48.3 (13.0)
BMI (kg/m ²)	19.0 (1.7)	18.3 (1.4)	18.9 (3.3)	20.0 (3.4)
Lean mass (kg)	41.6 (9.1) ^{b,dd}	35.4 (7.2)	37.7 (7.5)	31.7 (5.5)
Fat mass (kg)	8.3 (3.2)	6.6 (2.4)	8.6 (7.2)	14.1 (8.5) ^{a,bb,c}
Percentage of body fat (%)	17.1 (7.1)	15.8 (5.6)	17.8 (8.9)	29.0 (10.5) ^{aa,bb,cc}
Weekly training hours (h)	9.5 (5.1) ^{cc}	10.0 (2.3) ^{cc}	5.1 (2.1)	-
Pubertal maturation (I/II/III/IV/V) (%)	(15/25/13/45/2)	(24/35/24/16/0)	(14/28/28/27/3)	(29/21/21/29/0)
MVPA (min/day)	85.9 (30.4)	119.8 (29.7) ^{aa,d}	107.2 (33.3) ^a	83.2 (26.8)
VPA (min/day)	11.9 (7.3)	22.5 (9.0) ^{aa,dd}	18.5 (12.8) ^{a,d}	8.9 (4.0)
Energy intake (kcal/day)	2084.5 (560.6)	2093.7 (755.4)	2219.8 (843.4)	1748.9 (434.6)
Calcium intake (mg/day)	988.8 (429.7)	1017.9 (504.5)	1014.9 (601.9)	881.5 (380.7)
Vitamin D intake (µg/day)	1.84 (1.5)	1.82 (1.89)	1.95 (1.69)	1.52 (1.39)

Values presented as mean ± SD. BMI: Body mass index, MVPA: Moderate to vigorous physical activity, VPA: Vigorous physical activity.

Superscript letters denote a higher significant difference with: a (swimmers), b (footballers), c (cyclists), d (controls),
^{a,b,c,d} p<0.05, ^{aa,bb,cc,dd} p<0.001.

Table 2. Raw data for DXA region-specific bone mineral content (BMC, g), density (BMD, g/cm²) and area (BA, cm²) of all participants

Parameters		Swimmers (n=41)	Footballers (n=37)	Cyclists (n=29)	Controls (n=14)
TBLH	BMD	0.918 (0.067) ^d	0.931 (0.071) ^{dd}	0.905 (0.086) ^d	0.828 (0.071)
	BMC	1630.66 (333.56) ^d	1473.49 (338.60)	1498.27 (362.08)	1234.38 (347.86)
	BA	1762.63 (250.99) ^{b,d}	1564.89 (248.35)	1636.10 (261.69)	1469.43 (300.17)
Total hip	BMD	0.962 (0.107) ^{dd}	1.034 (0.085) ^{a,c,dd}	0.959 (0.114) ^{dd}	0.830 (0.116)
	BMC	28.87 (5.52) ^{dd}	28.78 (6.18) ^{dd}	27.59 (5.97) ^d	21.12 (5.55)
	BA	29.86 (3.83) ^d	27.60 (4.21)	28.54 (3.86)	25.14 (3.79)
Femoral Neck	BMD	0.948 (0.098) ^d	1.001 (0.081) ^{dd}	0.975 (0.192) ^d	0.832 (0.118)
	BMC	4.46 (0.65) ^{dd}	4.53 (0.74) ^{dd}	4.40 (0.79) ^d	3.52 (0.73)
	BA	4.70 (0.43) ^d	4.51 (0.46)	4.61 (0.43) ^d	4.21 (0.45)
Ward's triangle	BMD	0.928 (0.111) ^d	1.022 (0.096) ^{a,c,dd}	0.926 (0.127) ^d	0.799 (0.120)
	BMC	2.31 (0.49) ^d	2.40 (0.59) ^{dd}	2.23 (0.55) ^d	1.64 (0.45)
	BA	2.48 (0.43) ^d	2.34 (0.44)	2.40 (0.42)	2.04 (0.36)
Trochanter	BMD	0.799 (0.089) ^{dd}	0.882 (0.078) ^{aa,cc,dd}	0.799 (0.108) ^{dd}	0.678 (0.098)
	BMC	9.08 (2.41) ^d	9.31 (2.67) ^{dd}	8.61 (2.39) ^d	6.11 (2.17)
	BA	11.26 (2.30) ^d	10.41 (2.34)	10.66 (2.15)	8.82 (2.12)
Shaft	BMD	1.100 (0.140) ^d	1.170 (0.109) ^{dd}	1.090 (0.132) ^d	0.941 (0.150)
	BMC	15.33 (2.62) ^{dd}	14.94 (2.97) ^{dd}	14.58 (2.98) ^d	11.49 (2.74)
	BA	13.91 (1.36) ^{b,d}	12.67 (1.65)	13.27 (1.51)	12.11 (1.49)
Lumbar Spine	BMD	0.892 (0.114) ^d	0.883 (0.095)	0.867 (0.122)	0.791 (0.101)
	BMC	43.26 (11.21) ^d	38.54 (8.93)	39.50 (11.04)	32.64 (8.67)
	BA	47.94 (7.37) ^{b,d}	43.17 (8.82)	44.88 (6.99)	40.79 (6.99)
Arms	BMD	0.784 (0.071) ^{b,dd}	0.736 (0.047)	0.747 (0.069) ^d	0.690 (0.049)
	BMC	244.93 (64.87) ^{bb,dd}	188.34 (48.05)	212.89 (59.27) ^d	155.89 (40.58)
	BA	308.22 (58.14) ^{bb,dd}	253.62 (51.89)	281.00 (58.00) ^d	223.71 (45.67)
Legs	BMD	1.091 (0.010) ^d	1.124 (0.106) ^{dd}	1.077 (0.116) ^d	0.975 (0.103)
	BMC	779.05 (141.65) ^d	747.84 (175.02)	745.39 (179.21)	612.28 (179.74)
	BA	709.83 (80.64) ^d	657.46 (96.32)	684.24 (108.91)	617.50 (102.23)
Pelvis	BMD	0.994 (0.087) ^d	1.025 (0.103) ^{dd}	0.989 (0.130) ^d	0.888 (0.087)
	BMC	246.55 (57.43) ^d	238.35 (63.80) ^d	227.93 (63.75) ^d	174.81 (45.97)
	BA	245.85 (41.36) ^{dd}	229.19 (40.89) ^d	226.69 (37.38)	194.07 (34.96)

TBLH: Total body less head. Values are presented as mean ± SD. Superscript letters denote a higher significant difference with: a (swimmers), b (footballers), c (cyclists) and d (controls). ^{a,b,c,d} p<0.05 and ^{aa,bb,cc,dd} p<0.001.

Supplementary table 1: Adjusted data for DXA region-specific bone mineral content (BMC, g), density (BMD, g/cm²) and area (BA, cm²) of all participants

Parameters		Swimmers (n=41)	Footballers (n=37)	Cyclists (n=29)	Controls (n=14)
TBLH	BMD	0.888 (0.008)	0.950 (0.008) ^{aa,cc,dd}	0.903 (0.008)	0.873 (0.013)
	BMC	1462.44 (21.27)	1584.97 (22.09) ^{a,c,d}	1485.07 (23.13)	1468.36 (35.64)
	BA	1628.11 (13.78)	1659.72 (14.31)	1622.53 (14.98)	1640.69 (23.08)
Total hip	BMD	0.942 (0.015)	1.050 (0.016) ^{aa,cc,dd}	0.952 (0.016)	0.871 (0.025)
	BMC	26.67 (0.49)	30.42 (0.50) ^{aa,cc,dd}	26.96 (0.52)	24.74 (0.79)
	BA	28.12 (0.25)	28.88 (0.26)	28.10 (0.27)	27.65 (0.41)
Femoral Neck	BMD	0.934 (0.019)	1.009 (0.020) ^d	0.966 (0.020)	0.884 (0.031)
	BMC	4.23 (0.08)	4.70 (0.08) ^{aa,c,dd}	4.32 (0.08)	3.94 (0.12)
	BA	4.56 (0.05)	4.60 (0.05)	4.56 (0.05)	4.46 (0.08)
Ward's triangle	BMD	0.910 (0.018)	1.037 (0.019) ^{aa,cc,dd}	0.920 (0.019)	0.829 (0.030)
	BMC	2.16 (0.06)	2.49 (0.06) ^{a,c,dd}	2.18 (0.06)	1.95 (0.10)
	BA	2.37 (0.05)	2.40 (0.05)	2.36 (0.05)	2.28 (0.08)
Trochanter	BMD	0.786 (0.014)	0.890 (0.014) ^{aa,cc,dd}	0.791 (0.014) ^d	0.717 (0.022)
	BMC	8.23 (0.22)	9.92 (0.22) ^{aa,cc,dd}	8.34 (0.23)	7.66 (0.36)
	BA	10.35 (0.19)	11.10 (0.20)	10.42 (0.20)	10.21 (0.31)
Shaft	BMD	1.069 (0.019)	1.195 (0.020) ^{aa,cc,dd}	1.081 (0.020)	0.994 (0.031)
	BMC	14.21 (0.25)	15.79 (0.26) ^{aa,cc,dd}	14.30 (0.27)	13.14 (0.42)
	BA	13.23 (0.12)	13.18 (0.12)	13.13 (0.13)	12.98 (0.19)
Lumbar Spine	BMD	0.850 (0.014)	0.902 (0.015)	0.869 (0.015)	0.857 (0.024)
	BMC	38.29 (0.89)	41.11 (0.93)	39.46 (0.98)	40.24 (1.51)
	BA	44.49 (0.53)	45.28 (0.55)	44.65 (0.58)	45.62 (0.90)
Arms	BMD	0.752 (0.006)	0.754 (0.007)	0.752 (0.007)	0.726 (0.011)
	BMC	211.40 (3.15)	209.28 (3.19)	214.77 (3.41) ^d	195.68 (5.22)
	BA	276.16 (3.12)	275.13 (3.16)	280.95 (3.38) ^d	261.75 (5.17)
Legs	BMD	1.060 (0.011)	1.148 (0.011) ^{aa,cc,dd}	1.068 (0.012)	1.031 (0.018)
	BMC	713.53 (10.85)	799.47 (11.13) ^{aa,cc,dd}	729.54 (11.51)	707.81 (17.72)
	BA	666.45 (6.82)	691.97 (6.99)	675.52 (7.23)	670.46 (11.13)
Pelvis	BMD	0.954 (0.012)	1.047 (0.013) ^{aa,c,dd}	0.988 (0.014)	0.951 (0.021)
	BMC	216.33 (4.61)	255.68 (4.78) ^{aa,cc,dd}	227.69 (5.03)	218.61 (7.76)
	BA	224.41 (2.76)	242.12 (2.86) ^{aa,cc,dd}	226.18 (3.01)	222.39 (4.65)

TBLH: Total body less head

Values are presented as mean ± SE. Superscript letters denote a higher significant difference with: a (swimmers), b (footballers), c (cyclists) and d (controls).

^{a,b,c,d} p<0.05 and ^{aa,bb,cc,dd} p<0.001.

Adjusted for age, stature, calcium intake, MVPA and region-specific lean mass.

Supplementary table 2. Raw and adjusted data for Hip Structural Analysis (HSA) and Quantitative Ultrasound (QUS) parameters of all participants

Parameters	Swimmers (n=41)	Footballers (n=37)	Cyclists (n=29)	Controls (n=14)
Neck Width (mm)	5.9 (1.9)	6.7 (1.8)	6.5 (2.0)	5.3 (1.3)
Adjusted Neck Width (mm)	6.0 (0.3)	6.6 (0.3)	6.4 (0.3)	5.8 (0.5)
Diameter of FN (mm)	31.4 (2.7) ^d	30.5 (2.8)	30.9 (2.6) ^d	28.4 (2.5)
Adjusted Diameter of FN (mm)	30.7 (0.3)	30.9 (0.3)	30.6 (0.3)	30.0 (0.5)
CSMI (mm ⁴)	8944 (2574) ^d	8471 (2607) ^d	8403 (2552) ^d	6021 (2673)
Adjusted CSMI (mm ⁴)	8212 (244)	8947 (250) ^d	8102 (259)	7618 (398)
Section Modulus (mm ³)	558.3 (121.4) ^{dd}	548.1 (116.7) ^{dd}	530.8 (123.3) ^{dd}	395.0 (123.4)
Adjusted Section Modulus (mm ³)	523.6 (12.0)	571.5 (12.3) ^{c,dd}	516.1 (12.7)	472.2 (19.6)
CSA (mm ²)	137.2 (20.2) ^{dd}	140.9 (20.4) ^{dd}	135.9 (22.7) ^d	109.8 (21.0)
Adjusted CSA (mm ²)	131.1 (2.3)	145.3 (2.4) ^{aa,c,dd}	133.7 (2.4)	121.8 (3.8)
Hip Strength Index	1.45 (0.35)	1.75 (0.37) ^{a,dd}	1.62 (0.34) ^d	1.26 (0.37)
NA				
SI Dominant	91.6 (13.2) ^d	100.4 (12.4) ^{a,dd}	92.7 (11.5) ^d	81.3 (11.2)
Adjusted SI Dominant Foot	89.6 (2.0)	101.6 (2.1) ^{aa,c,dd}	92.1 (2.2)	84.3 (3.3)
SI Non-Dominant Foot	89.6 (11.9)	97.8 (10.5) ^{a,dd}	92.7 (13.8)	83.1 (12.2)
Adjusted SI Non-Dominant Foot	87.9 (1.9)	98.6 (1.9) ^{a,d}	91.9 (2.0)	86.0 (2.9)

Values presented as mean ± SD. CSA: Cross sectional area, CSMI: Cross sectional moment of inertia, FN: femoral neck. NA: Not Available adjustment for Hip Strength Index. SI: Stiffness Index. Superscript letters denote a higher significant difference with: a (swimmers), b (footballers), c (cyclists), d (controls). ^{a,b,c,d} p<0.05, ^{aa,bb,cc,dd} p<0.001. Adjusted for age, stature, calcium intake, MVPA and region-specific lean mass.