Longitudinal effects of swimming on bone in adolescents: a pQCT and DXA study

INTRODUCTION

Osteoporosis is a skeletal disorder characterized by compromised bone strength, predisposing to an increased risk of fracture [1]. Peak bone mass accumulation, which will contribute to future osteoporosis risk, occurs around the ages of 12 to 14 years and is greater in active children [2]. This is important because active children will attain a higher peak bone mass, and will therefore present a lower risk of suffering osteoporosis later in life. Nevertheless, not all sports have the same effect on bone mass, as non-weight-bearing sports have been reported to be neutral to bone mass [3].

Focusing on swimming, few studies have evaluated bone mass longitudinally in adolescent swimmers comparing them to sedentary controls or other athletes [4-6]. Maimoun et al. [6] compared female adolescent swimmers’ areal bone mineral density (aBMD) acquisition to that of normo-active controls (CG) during one year, finding no differences between swimmers and CG in bone acquisition. Similarly, Czeczelewski et al. [4] registered aBMD acquisition during a 3-year period in female swimmers and a CG, finding no differences in aBMD acquisition. Ferry et al. [5] also compared female adolescent swimmers to other athletes (soccer players), finding that during 8 months of training, swimmers gained less whole body and lumbar spine aBMD than soccer players.

Although dual energy X-ray (DXA) is the gold standard for measuring aBMD, this measurement accounts at best for 60-70% of the variance in ultimate strength of bone tissue [7]. DXA has limitations with respect to the assessment of bone geometric structure and the arrangement of the mineral in the cortical vs. the trabecular compartments of the bone [8], which are distinguished when using peripheral quantitative computed tomography (pQCT). In fact, few studies...
have been carried out on swimmers using pQCT, with most studies being cross-sectional studies in adults, finding that swimmers present similar bone strength indexes when compared to sedentary controls [9-13].

To date, and as far as we know, only one study has described the bone structure of adolescent swimmers using pQCT [14]. This cross-sectional study found that swimmers presented similar strength values when compared to normo-active controls (CG). Other researchers have used DXA hip structural analysis (HSA), finding no differences between male swimmers and CG [15] and lower geometry values in both male and female swimmers when compared to other sports [5, 15]. Nonetheless, HSA is limited by the two-dimensional nature of DXA and does not allow the distinction of cortical and trabecular bone [16]. Moreover, HSA results are greatly influenced by the femur rotation that will affect the projected dimensions from which the geometry is measured [16].

It is of critical importance to evaluate changes both in aBMD and bone strength and structure during a season in adolescent swimmers, as future bone strength will not only depend on bone density but also on bone structure. In addition, bone mass improvements are thought to be mainly related to geometric adaptations and to a lesser extent to changes in aBMD [17]. Thus, it is possible that when comparing aBMD, bone structure and bone strength longitudinally there may be improvements in structure and consequently in strength without improvements in aBMD.

The aims of the present study were therefore; 1) to evaluate swimmers’ aBMD with DXA and bone mass, bone strength and structure with pQCT during a swimming season and compare them to those of normo-active controls (CG), and 2) to evaluate whether practising an extra weight-bearing sport in addition to swimming might confer any aBMD, bone mass, strength or structure benefits.

MATERIALS AND METHODS

Experimental design

Swimmers were recruited from four swimming clubs from the city of Zaragoza, while participants in the control group were recruited from three high schools of the same city.

Participants were requested to visit our laboratory twice. The first evaluation took place between September and November 2012, while the second took place between May and July 2013. Therefore 8 months passed between baseline and post-evaluation.

Written informed consent from parents and verbal assent from the participants were obtained. The study was performed following the ethical guidelines of the Declaration of Helsinki 1961 (revision of Fortaleza 2013). The protocol study was approved by the Ethics Committee of Clinical Research from the Government of Aragón (ref. CP08/2012, CEICA, Spain).

Inclusion criteria

When the study began, inclusion criteria were as follows: participants had to be between the ages of 11 and 18 years, Caucasian, healthy, non-smokers, with no chronic disease or musculoskeletal disorders (fibromyalgia, gout, osteoarthritis, rheumatoid arthritis, tendinitis), bone fractures or medication. Swimmers had to have a history of swimming and competing in regional tournaments for more than 3 years and training for a minimum of 6 hours per week, while CG subjects could not be performing any aquatic activity or more than 3 hours of weight-bearing physical activity per week on a regular basis. Hours of physical activity were determined with a questionnaire. Swimming attendance was individually supervised by trainers. If a swimmer missed many training sessions and consequently did not reach the minimum average of 6 hours per week, he or she was excluded from the study.

Participant classification

As some of the swimmers who continued with their normal swimming training performed an extra weight-bearing sport on a weekly basis that could modify or mask the results, swimmers were classified as swimmers who swam and performed other sports on a regular basis for more than 3 hours per week (SWI-SPORT) or swimmers that only swam (SWI-PURE). Therefore, for the present study, 3 groups were analyzed and compared: 1) SWI-SPORT, 2) SWI-PURE and 3) CG.

Evaluation of pubertal stage

Pubertal maturation was determined by self-assessment of secondary sexual characteristics according to the criteria devised by Tanner [18]. This method has been reported to be both valid and reliable in assessing sexual maturity among adolescent athletes [19].

Bone parameters

Peripheral quantitative computed tomography (pQCT)

Bone mass, structure and strength were assessed with a Stratec XCT-2000 L scanner (Stratec Medizintechnik, Pforzheim, Germany). The scanning procedure is described in detail elsewhere [14]. Scans were performed in the non-dominant radius and tibia. Radius bone parameters were assessed at 4% and 66% of the forearm length and for the tibia at 4% and 38% of the tibia length. At the 4% site of the radius and tibia total (TOT_Area4%) cross-sectional bone area (mm²), total (TOT_BMC4%) and trabecular (TRB_BMC) BMC (g/cm) were evaluated. At the 66% site of the radius and 38% site of the tibia total (TOT_Area38%) and cortical (CRT_Area) bone cross-sectional area (mm²) and total BMC (TOT_BMC38%) were measured. Cortical thickness (CRT_THK, mm) was also measured at 38% of theibia and 66% of the radius. Bone strength was established with respect to torsion (polar stress strain index, mm³ (SSIPOL)), and bending (fracture load, N (FRC_LOAD_X)) with respect to the X-axis. All the strength indexes were calculated at 38% of the tibia and 66% of the radius.

Dual energy X-ray

Dual energy X-ray (DXA) scans were performed at the whole-body, lumbar spine and hip (trochanter and femoral neck). The protocol
and results for the first evaluation are explained elsewhere [20]. The second evaluation was performed following the same protocol. Lumbar spine, hip and whole body scans were performed, and aBMD was reported for the femoral neck, trochanter, lumbar spine, subtotal body (Whole body – head), arms and legs. ABMD was evaluated with the paediatric version of the software QDR-Explorer (Hologic Corp., Software version 12.4, Bedford, Massachusetts, USA).

Statistical analyses
Sample size calculation. As the main outcome for the present study was tibia bone strength (SSIPOL or FRC_LOAD_X) for the pQCT variables and femoral neck for the DXA variables, sample size was calculated for these 2 variables. For bone strength, data from a previous study evaluating bone strength against torsion and bending in swimmers, controls and other athletes were used to calculate sample size [10]. Due to the lack of studies performed with an adolescent swimming sample, data were obtained from adults. Three groups of data were obtained, data for the SWI-PURE group from a swimming group (1777±316 mm³), data for the SWI-SPORT group from a repetitive low impact group (2063±315 mm³) and data for the CG from a reference group (1646±296 mm³). For femoral neck aBMD data from a study by Taffe et al. [21] were used to calculate the sample size.

As two different statistical tests were performed (analysis of covariance (ANCOVA) and repeated measures ANCOVA), two different sample size calculations were performed in order to guarantee sufficient power.

Firstly, ANCOVA, adjusted by age, Tanner stage and object length, was performed to compare the cross-sectional data of SWI-SPORT, SWI-PURE and CG at pre- and post-evaluation. G*POWER was used selecting ANOVAs to obtain the effect size. For the SSIPOL, a large effect size was obtained ($f = 0.46$), with a required sample of 48 participants. Nevertheless, by adding covariates to the model that are correlated with the outcome variable, the error term is reduced and power is increased with a consequent decrease in the required sample size. Therefore $f$ was adjusted following the Rogers et al. [22] formula adjusted

$$f_a = \frac{f}{\sqrt{1 - r^2}}$$

where $r^2$ is the multiple correlation coefficient between covariates and the dependent variable, which was calculated with linear regression from a pilot study that included 18 swimmers. With an $R^2$ of 0.8 the adjusted $f_a$ was 0.52 with a required sample size of 39 participants (13 participants per group). For the femoral neck the results suggested 8 participants per group. Consequently, including 13 participants per group would allow us to reject the null hypothesis that the population means of the SWI and CG are equal with a power of 0.8 and $\alpha$ of 0.05.

Repeated measures were performed to determine whether both SWI and CG improved over time, and whether a group by time interaction existed. Thus, for a small to medium effect size ($f = 0.20$), and a correlation of 0.7 between the pre- and post-evaluation measurements, the within factors, repeated measures calculation determined a total sample size of 33 (11 per group). For the repeated measures, within-between interaction the required sample size was also 33 (11 per group). Similar results were found for the femoral neck as 11 participants per group were needed for a power of 0.8 and $\alpha$ of 0.05.

Therefore, the ideal sample for the present study would consist of a minimum of 13 SWI-SPORT, 13 SWI-PURE and 13 CG subjects.

Outcome measures treatment. All data were analyzed using SPSS version 15.0 (SPSS, Chicago, IL, USA). Significance level was reported as a level at or below 0.05. Because there were no gender by time interactions, data for girls and boys were pooled and analyzed together.

The Kolmogorov-Smirnov test was used to check for a normal distribution, showing that all variables were normally distributed.

ANOVA with the Bonferroni post hoc test was used to examine baseline and post-evaluation differences between groups (SWI-PURE, SWI-SPORT and CG) for age, height, weight, tibia and radius length. In addition, baseline and post-evaluation aBMD, bone strength, structure, area and BMC values were compared by means of ANCOVA adjusted by age, Tanner stage and height (DXA) or object length (pQCT). Chi-square tests were performed to evaluate differences in Tanner stage before and after the intervention.

ANOVA for repeated measures x 2 (time) was performed between pre- and post-evaluation to determine the effects of swimming on aBMD, bone strength, structure, area and BMC values adjusted by change in height (DXA) or object length (pQCT), initial age and final Tanner stage. When a group by time interaction was significant, further pairwise comparisons were performed (1. SWI-PURE vs. CG; 2. SWI-PURE vs. SWI-SPORT; 3. SWI-SPORT vs. CG).

Effect size calculation. For the within-group improvement, partial Omega squared ($\omega^2$) was calculated from the F and degrees of freedom reported by SPSS. $\rho^2$ is less biased than partial eta squared ($\eta^2$), particularly in small samples like the present one, as when there is no true effect $\rho^2$ from small studies can give the erroneous impression that there is a real small to medium effect may be given, entirely as a result of the bias [23].

For the group by time interactions, as $\omega^2$ is underestimated if there is subject by treatment interaction, $\rho^2$ was used to report effect size of the group by time interactions.

Both $\omega^2$ and $\rho^2$ can be interpreted as: <0.02 small effect, 0.06 moderate effect, >0.14 large effect [24].

RESULTS

Participants
Exclusion and participant loss are detailed in Figure 1.

Physical characteristics of the participants are shown in Table 1. Differences between groups for baseline and post-evaluation were found for height and tibia length, which were higher in SWI-SPORT
than in CG (all $P<0.05$). No other differences were found among groups for any of the variables studied (all $P>0.05$).

**Practised sports**
Regarding SWI-SPORT, five swimmers regularly performed low-impact sports (weight-lifting) while six swimmers were engaged in high-impact sports (2 running, 3 basketball, 1 soccer). For the CG five controls performed low-impact sports (1 cycling, 4 dancing) and six performed high-impact sports (1 basketball, 2 soccer, 1 karate, 1 tennis, 1 gymnastics), always no more than 3 hours per week. None of the SWI-PURE were performing additional sports. All swimmers performed less than one hour per week of dry land training.

**DXA measurements**
Table 2 summarizes the average adjusted values of aBMD at pre- and post-evaluation.
SWI-SPORT presented higher arm aBMD values than CG at baseline and post-evaluation (p<0.05; Table 2).

Within-group improvements
SWI-PURE, SWI-SPORT and CG improved femoral neck ($\omega^2 = 0.297/0.456/0.103$ respectively), lumbar spine ($\omega^2 = 0.535/0.562/0.329$ respectively), subtotal body ($\omega^2 = 0.484/0.833/0.548$ respectively), arms ($\omega^2 = 0.374/0.554/0.666$ respectively) and legs ($\omega^2 = 0.374/0.554/0.666$ respectively) aBMD values (all $p<0.05$; Table 2). Additionally, SWI-SPORT presented improvements in the trochanter ($\omega^2=0.598$).

Group by time interactions
When comparing aBMD acquisition of the 3 groups, a significant group by time interaction was found for the trochanter ($np^2=0.218$; $p<0.05$; Table 2). When focusing on the differences among groups, the posterior pairwise analyses showed two different interactions for the comparisons of SWI-SPORT vs. SWI-PURE ($np^2=0.131$; $p=0.05$) and SWI-SPORT vs. CG ($np^2=0.301$; $p<0.05$), in both cases favouring SWI-SPORT. No group by time interaction was found between SWI-PURE and CG ($np^2=0.06$; $p>0.05$).

Bone structure, strength, area and BMC
Table 2 summarizes bone structure, strength, area and BMC values at pre- and post-evaluation. No differences were found among groups for radius or tibia, either at pre- or post-evaluation (all $p>0.05$).

Within-group improvements
For the radius, both SWI-PURE and CG improved CRT_Area, CRT_BMC, TOT_BMC38%, SSIPOL (both $P<0.05$; Table 2) and TOT_Area38% (SWI-PURE $P<0.05$, CG $P<0.06$; Table 2). SWI-PURE also improved TOT_BMC4% ($p<0.05$; Table 2).

For tibia values, the 3 groups improved CRT_Area, CRT_BMC, TOT_BMC38%, FRC_LOAD_X and SSIPOL (all $p<0.05$; Table 2). Improvements were also found from baseline to post-evaluation for the 3 groups for CRT_THK (SWI-PURE and SWI-SPORT both $p<0.05$; CG $p<0.06$) and TOT_AREA38% (SWI-PURE and CG both $p<0.05$; SWI-SPORT $p<0.06$).

Group by time interactions
No group by time interactions were found for the radius values (all $p>0.05$; Table 2).

For the tibia, significant group by time interactions were found for CRT_THK, FRC_LOAD_X, SSIPOL, TOT_BMC38% and CRT_Area among the 3 groups (all $p<0.05$; Table 2). Further pairwise comparisons showed that SWI-SPORT presented a positive tendency towards group by time interactions when compared to SWI-PURE for FRC_LOAD_X and SSIPOL ($pn^2=0.145$ and 0.138 respectively; Figure 2).

When comparing SWI-SPORT to CG, group by time interactions were found for CRT_THK, FRC_LOAD_X, SSIPOL, TOT_BMC38% and CRT_Area among the 3 groups (all $p<0.05$; Figure 2). Further pairwise comparisons showed that SWI-SPORT presented a positive tendency towards group by time interactions when compared to SWI-PURE for FRC_LOAD_X and SSIPOL (both $p=0.08$; $pn^2=0.145$ and 0.138 respectively; Figure 2).

When comparing SWI-SPORT to CG, group by time interactions were found for CRT_THK ($pn^2=0.276$), FRC_LOAD_X ($pn^2=0.252$), SSIPOL ($pn^2=0.281$), TOT_BMC38% and CRT_Area ($pn^2=0.214$) (all $p<0.05$; Figure 2).

Finally, when comparing SWI-PURE to CG, interactions favouring the SWI-PURE were found for CRT_THK ($pn^2=0.299$), SSIPOL ($pn^2=0.231$), TOT_BMC38% ($pn^2=0.170$), and CRT_AREA ($pn^2=0.273$) (all $p<0.05$; Figure 2).
### TABLE 2. Baseline and post-intervention pQCT bone strength indexes adjusted by object length (pQCT) or height change (DXA), Tanner stage and age.

| DXA | Baseline | Post-evaluation | Repeated measures | GxT Whithin group |
|-----|----------|----------------|------------------|------------------|
|     | SWI-PURE | SWI-SPORT | CG | SWI-PURE | SWI-SPORT | CG | SWI-PURE | SWI-SPORT | CG | pO2 | pO2 | pO2 | pO2 |
|     | n=23 | n=11 | n=28 | n=23 | n=11 | n=28 | n=23 | n=11 | n=28 |
| Hip | TROCH (g/cm²) | 0.740 ±0.104 | 0.757 ±0.104 | 0.721 ±0.111 | 0.754 ±0.101 | 0.793 ±0.101 | 0.725 ±0.104 | 0.108 | 0.598 <0.001 | 0.218 |
|     | NECK (g/cm²) | 0.807 ±0.108 | 0.866 ±0.108 | 0.833 ±0.110 | 0.826 ±0.107 | 0.885 ±0.108 | 0.844 ±0.110 | 0.297* | 0.456 | 0.103* | 0.080 |
| Spine | LSP (g/cm²) | 0.842 ±0.120 | 0.879 ±0.120 | 0.861 ±0.128 | 0.877 ±0.116 | 0.909 ±0.117 | 0.886 ±0.122 | 0.535* | 0.562 | 0.329* | 0.080 |
| Whole body | SUBT (g/cm²) | 0.873 ±0.071 | 0.912 ±0.076 | 0.889 ±0.076 | 0.934 ±0.072 | 0.905 ±0.075 | 0.484* | 0.833 | 0.548* | 0.078 |
|     | ARMS (g/cm²) | 0.690 ±0.046 | 0.710 ±0.045c | 0.667 ±0.049 | 0.708 ±0.044 | 0.724 ±0.044c | 0.679 ±0.046 | 0.374* | 0.554 | 0.666 | 0.016 |
|     | LEGS (g/cm²) | 1.026 ±0.096 | 1.071 ±0.102 | 1.058 ±0.101 | 1.089 ±0.103 | 1.094 ±0.106 | 0.435* | 0.698 | 0.655* | 0.031 |
| Radius | FRC_LOAD_X (N) | 486.96 ±89.18 | 470.88 ±90.62 | 521.78 ±93.14 | 480.17 ±95.11 | 0.115 | 0.187 | 0.013 |
|     | SSI_POL (mm³) | 259.94 ±42.13 | 256.44 ±42.81 | 285.42 ±51.10 | 262.45 ±52.19 | 0.420* | 0.335 | 0.011 |
|     | CRT_THK (mm) | 2.09 ±0.26 | 2.24 ±0.27 | 2.11 ±0.32 | 2.26 ±0.33 | <0.001 | NA | 0.001 | 0.018 |
|     | CRT_Area (mm²) | 69.64 ±09.41 | 73.33 ±09.61 | 72.98 ±10.44 | 73.88 ±10.66 | 0.464* | NA | 0.001 | 0.001 |
|     | TOT_BMC38% (g/cm) | 0.910 ±0.134 | 0.922 ±0.136 | 0.951 ±0.129 | 0.941 ±0.131 | 0.413* | NA | 0.015 | 0.001 |
|     | TOT.Area38% (mm²) | 127.92 ±16.35 | 125.16 ±16.71 | 134.98 ±16.04 | 128.23 ±16.38 | 0.475* | NA | 0.001 | 0.001 |
| Tibia | TRB_BMC (g/cm) | 0.289 ±0.047 | 0.283 ±0.048 | 0.320 ±0.076 | 0.288 ±0.077 | 0.057 | NA | 0.015 | 0.002 |
|     | TOT_BMC4% (g/cm) | 1.001 ±0.148 | 1.014 ±0.150 | 1.109 ±0.187 | 1.017 ±0.190 | 0.292* | NA | 0.150 | 0.014 |
|     | TOT.Area4% (mm²) | 316.94 ±72.36 | 325.30 ±73.92 | 354.17 ±87.66 | 339.60 ±89.40 | 0.155 | NA | 0.204 | 0.003 |

Data presented as mean±SD; NA=Not applicable due to small sample size; SWIPURE. #Significant differences when compared to the CG. #Within group differences from baseline to post-evaluation p<0.05. *Tendency towards within group differences from baseline to post-evaluation p<0.10. ω²=partial omega squared; p²=partial eta squared; both effect sizes can be interpreted as; <0.01 small. 0.06 moderate, >0.14 large. GxT=Group by time interactions; *Significant group by time interaction. SWI-PURE=Swimmers that only swam; SWI-SPORT=Swimmers that swam and performed and additional weight-bearing sport; CG=Control-group; CRT_THK=Cortical thickness; CRT=Cortical; BMC=Bone mineral content; TOT=Total; TRB=Trabecular; FRC_LOAD_X=Fracture load (axe X); SSI_POL=Polar strength strain index.
FIG 2. Tibia bone structure, strength indexes, bone mineral content and area. Values adjusted by change in object length, baseline age and post-evaluation Tanner stage.
Legend. *=group by time interaction (p<0.05); #=group by time interaction (p=0.08).
DISCUSSION

The present study is the first to combine DXA and pQCT measurements in adolescent swimmers and to assess changes in bone over a swimming season. The main findings of the present study were 1) swimming has no effect on aBMD, radius bone strength, structure or BMC, and minor effects on tibia bone strength, structure and BMC, and 2) practising a weight-bearing sport in addition to swimming is beneficial to tibia strength and structure and to trochanter aBMD.

The only differences found among the 3 groups for DXA and pQCT variables were for the DXA aBMD arm values as SWI-SPORT presented higher pre- and post-evaluation than CG. No other differences were found for any of the studied variables at pre- or post-evaluation among groups, suggesting that swimming has no effect on aBMD. These results are in line with those found in a recent meta-analysis performed in adolescents and children comparing aBMD values between swimmers and CG, finding no differences between groups [25].

Although the present longitudinal study and previously published ones [4, 6, 26] suggest that swimming is not negative to bone mass and therefore should be considered as a “neutral” sport to practise in terms of aBMD, it is important to underline that adolescence is a critical period for bone acquisition. Previous research suggested that physical activity in this period may have greater positive effects on bone mass than many pharmacological interventions undertaken by adults with osteoporosis [27]. Therefore, the fact that adolescent swimmers present similar bone mass acquisition patterns to those not performing weight-bearing activities on a regular basis could entail a negative effect later in life.

Regarding the pQCT variables, no differences were found either at pre- or at post-evaluation among the 3 groups. Focusing on the radius, no group by time interactions or differences in the within-group improvements (swimmers improved the same variables as CG) were found, suggesting that the improvements could be due to biological growth, as maturity-related differences in bone geometry, density, and strength in boys and girls have been previously well documented [28]. This lack of differences for radius values has been previously described in cross-sectional studies performed in adolescents [14] and adults [12] that have also compared swimmers to CG, finding no differences in radius strength indexes between groups. The lack of radius improvements was surprising, as unlike the tibia or the femoral neck, which are constantly submitted to gravitational forces while walking and performing daily activity tasks, the forearm bones do not have to sustain loads during habitual daily tasks. It could therefore be expected that improvements in swimmers’ radius when compared to CG would be found, as swimmers are constantly pushing with their hands against the water, a movement that involves the forearm muscles that react with contractions and thus produce bone strain. Nevertheless, from the lack of improvements found in the present study it would appear that the push against the water executed while swimming is not sufficient to reach the minimum effective strain for modelling, which would result in bone gain [29]. For the tibia variables, the group by time interaction found for cortical thickness, cortical area and total BMC at the shaft in SWI-SPORT when compared to CG was in line with previous studies that also found improvements in these parameters in jumpers when compared to CG, as during impact exercises, the rate of loading is much higher, and thus is associated with thickened cortical bone [9, 11, 30]. This increase in cortical thickness could explain the improvements in SSIPOL and FRAC_LOAD_X, both strength indexes that are influenced by cortical size, and that have previously been described as higher in weight-bearing athletes than in controls [10, 31].

Surprisingly, SWI-PURE also presented group by time interactions for cortical thickness, SSIPOL, total BMC and cortical area, all measured at the shaft. Liu et al. [9] also found that female adult swimmers presented higher tibia strength strain indexes values than controls. In contrast to our findings, Nikander et al. [32] did not find any differences between adult swimmers and CG in cortical thickness of the femoral neck measured with MRI. Nevertheless, the femoral neck might not support as much impact as the tibia while swimming, as swimmers are constantly pushing against the wall, an action that involves the calf muscles, although this is just a hypothesis that has not been tested in any previous studies. Swimmers develop their sport in a hypogravitational medium with no impacts until they perform a turn and use the wall to push themselves off and regain speed. This push against the wall may be compared to the takeoff phase while jumping although with much less force. Therefore, these “pushes”, that do not generate as much reaction force as a jump, might be able to generate sufficient forces to reach the minimum stimulus necessary to have some effect on the tibia bone. Although these pushes would generate very small forces, they are generated very often, as swimmers in the present sample train on average 10 hours per week, in 25-metre swimming pools with average training sessions of 3000 metres (120 pushes against the wall per session). The fact that no differences exist for the radius and some minor benefits do exist for the tibia could be due to this elevated number of minor impacts performed while pushing, which would range from 600 to 800 per week. Nevertheless, this is just a hypothesis that should be tested in future studies that compare swimmers who perform the exact amount of training in different types of swimming pools (25 metres vs. 50 metres).

The significant group by time interactions found for the tibia values among the 3 groups suggest that swimmers and CG are evolving in different ways. These interactions could be partly explained by the previously mentioned pushes but could also be due to differences in maturational stage, as although there were no differences in pre- or post-evaluation for Tanner stage, the CG had a larger number of participants in Tanner stage 5. It is probable that these participants would not improve as much as participants with lower maturation status. This hypothesis is supported by Figure 2, as when comparing SWI-PURE to CG, SWI-PURE at baseline was always lower than the CG (although not significantly), and although there was a group by
time interaction suggesting that SWI-PURE improved more than the CG, these improvements were always trending to equalize the CG values (although they always remained below). In contrast, the group by time interactions between SWI-SPORT and CG, which also suggest that SWI-SPORT improved more than the CG, showed that SWI-SPORT started and ended at a higher point, suggesting that the differences between groups increased (although they were non-significant at pre- and post-evaluation). The fact that these differences between SWI-SPORT and CG at pre- and post-evaluation were non-significant could be due to the sample size of SWI-SPORT (n=10) when the ideal sample size would have been 13. Effect sizes for the comparison between the 2 groups supported this idea, as they were on average medium ($pn^2=0.07$).

In addition, when comparing SWI-SPORT to SWI-PURE, a tendency towards a group by time interaction between the two groups was found, suggesting that the SWI-SPORT subjects improved their tibia FRC_LOAD_X and SSIPOL more than the SWI-PURE subjects. Again, it is possible that this group by time interaction did not reach significance due to sample size. The large effect size of 0.145 and 0.138 makes us hypothesize that if we had reached 13 participants per group we would have found significant group by time interactions between SWI-SPORT and SWI-PURE for the 2 bone strength indexes. This reinforces the idea that extra-weight-bearing training in addition to swimming is of critical importance to adolescent swimmers.

The controversial results found in current literature evaluating aBMD in adolescent swimmers [33] might be explained by differences in physical activity involvement by the CG or the extra activity performed by some of the swimmers included in those studies. The results of the present study support the latter hypothesis, as swimmers who performed an extra activity (SWI-SPORT) showed higher aBMD trochanter acquisition when compared to both SWI-PURE and CG, suggesting that the additional aBMD gains were due to the impacts while performing other weight-bearing activities. SWI-SPORT was the only group that improved aBMD of all the measured areas. Therefore, future studies with swimmers should always report additional weight-bearing activities performed by the swimmers, as pooling together swimmers who only swim with swimmers who perform other sports could mask the results.

The main limitation of the present study is the grouping of both males and females due to the limited sample size (although there was no gender by time interaction, suggesting that both improved similarly). Another important limitation was the quantification of additional practised sports, as they were self-reported and not objectively measured, and therefore some participants might have overestimated their extra-physical activity while others might have underestimated it. Nonetheless, the present study has several strengths such as the use of two bone measurement techniques to evaluate both bone mass and bone structure and the longitudinal design of the study.

**CONCLUSIONS**

It seems that swimming has no effect on radius bone strength and induces minor benefits on tibia bone strength and trochanter aBMD. Swimmers who complemented swimming with an extra weight-bearing activity presented more improvements in aBMD and tibia structure and strength than any of the other groups, suggesting that impacts are determinant for improving bone strength.

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**Conflicts of interest**

Authors declare that they have no conflicts of interest that may affect the contents of this work.

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**REFERENCES**

1. Department of Health and Human Services US. Bone Health and Osteoporosis: A Report of the Surgeon General.; 2004.
2. Vicente-Rodriguez G. How does exercise affect bone development during growth? Sports Med. 2006;36(7):561-9.
3. Guadalupe-Grau A, Fuentes T, Guerra B, Calbet JA. Exercise and bone mass in adults. Sports Med. 2009;39(6):439-68.
4. Czeczelewski J, Długolecka B, Czeczelewski E, Raczyńska B. Intakes of selected nutrients, bone mineralisation and density of adolescent female swimmers over a three-year period. Biol Sport. 2013;30(1):17-20.
5. Ferry B, Lespesailles E, Rochcongar P, Duclos M, Courtex D. Bone health during late adolescence: Effects of an 8-month training program on bone geometry in female athletes. Joint Bone Spine. 2013 January;80(1):57-63.
6. Maimoun L, Coste O, Mura T, Philibert P, Galtier F, et al. Specific bone mass acquisition in elite female athletes. J Clin Endocrinol Metab. 2013 Jul;98(7):2844-53.
7. Dougherty G. Quantitative CT in the measurement of bone quantity and bone quality for assessing osteoporosis. Med Eng Phys. 1996 Oct; 18(7):557-68.
8. Bouxsein ML, Kasaski D. Bone geometry and skeletal fragility. Curr...
Osteoporos Rep. 2006 Jun; 4(2):49-56.

9. Liu L, Maruno R, Mashimo T, Sanka K, Higuchi T, et al. Effects of physical training on cortical bone at midtibia assessed by peripheral QCT. J Appl Physiol. 2003 Jul;95(1):219-24.

10. Nikander R, Kannus P, Rantalainen T, Uusi-Rasi K, Heinonen A, et al. Cross-sectional geometry of weight-bearing tibia in female athletes subjected to different exercise loadings. Osteoporos Int. 2010 Oct; 21(10):1687-94.

11. Nikander R, Sievanen H, Uusi-Rasi K, Heinonen A, Kannus P. Loading modalities and bone structures at nonweight-bearing upper extremity and weight-bearing lower extremity: A pQCT study of adult female athletes. Bone. 2006;39(4):886-94.

12. Schipilow JD, Macdonald HM, Liphardt AM, Kan M, Boyd SK. Bone micro-architecture, estimated bone strength, and the muscle-bone interaction in elite athletes: an HR-pQCT study. Bone. 2013 Oct;56(2):281-9.

13. Shaw CN, Stock JT. Habitual throwing and swimming correspond with upper limb diaphyseal strength and shape in modern human athletes. Am J Phys Anthropol. 2009;140(1):160-72.

14. Gomez-Bruton A, Gonzalez-Aguero A, Gomez-Cabello A, Matute-Llorente A, Casajús JA, Vicente-Rodríguez G. The effects of swimming training on bone tissue in adolescence. Scand J Med Sci Sports. 2015;25(6):e589-602.

15. Vlachopoulos D, Barker AR, Williams CA, Arngrimson SA, Knapp KM, et al. The Impact of Sport Participation on Bone Mass and Geometry in Adolescent Males. Med Sci Sports Exerc. 2016 Sep 14.

16. Beck TJ. Extending DXA beyond bone mineral density: understanding hip structure analysis. Curr Osteoporos Rep. 2007 Jun;5(2):49-55.

17. Ashizawa N, Nonaka K, Michikami S, Mizuki T, Amagai H, et al. Tomographical description of tennis-loaded radius: reciprocal relation between bone size and volumetric BMD. J Appl Physiol. 1999 Apr;86(4):1347-51.

18. Tanner JM, Whitehouse RH, Takaishi M. Standards from birth to maturity for height, weight, height velocity, and weight velocity: British children, 1965. II. Arch Dis Child. 1966 Dec;41(220):613-35.

19. Leone M, Comtois AS. Validity and reliability of self-assessment of sexual maturity in elite adolescent athletes. J Sports Med Phys Fitness. 2007 Sep;47(3):361-5.

20. Gomez-Bruton A., Gonzalez-Aguero A., Gomez-Cabello A., Matute-Llorente A., Casajús JA., Vicente-Rodríguez G. The effect of swimming training on bone tissue in adolescence. Scand J Med Sci Sports. 2015;25(6):e589-602.

21. Taaffe DR, Snow-Harter C, Connolly DA, Robinson TL, Brown MD, et al. Differential effects of swimming versus weight-bearing activity on bone mineral status of eumenorrheic athletes. J Bone Miner Res. 1995 Apr;10(4):586-93.

22. Rogers WT, Hopkins KD. Power estimates in the presence of a covariate and measurement error. Educational and Psychological Measurement. 1988;48:647-56.

23. Lakens D. Calculating and reporting effect sizes to facilitate cumulative science: a practical primer for t-tests and ANOVAs. Front Psychol. 2013;4:863.

24. Cohen D. Statistical Power Analysis for the Behavioral Sciences. 2nd ed. ed. Hillsdale, NJ: 1988.

25. Gomez-Bruton A, Montero-Marín J, Gonzalez-Aguero A, García-Campayo J, Moreno LA, et al. The Effect of Swimming During Childhood and Adolescence on Bone Mineral Density: A Systematic Review and Meta-Analysis. Sports Med. 2015 Nov 26; 46(3):365-79.

26. Diugolęcka B, Czecelewski J, Racyzynska B. Bone mineral content and bone mineral density in female swimmers during the time of peak bone mass attainment. Biol Sport. 2011;28(1):63-74.

27. Gunter KB, Almstedt HC, Janz KF. Physical activity in childhood may be the key to optimizing lifespan skeletal health. Exerc Sport Sci Rev. 2013 Jan; 40(1):13-21.

28. Kontulainen SA, Macdonald HM, Khan KM, McKay HA. Examining bone surfaces across puberty: a 20-month pQCT trial. J Bone Miner Res. 2005 Jul;20(7):1202-7.

29. Skerry TM. One mechanostat or many? Modifications of the site-specific response of bone to mechanical loading by nature and nurture. J Musculoskeletal Neuronal Interact. 2006 Apr-Jun;6(2):122-7.

30. Heinonen A, Sievanen H, Kyrolainen H, Perttunen J, Kannus P. Mineral mass, size, and estimated mechanical strength of triple jumpers’ lower limb. Bone. 2001 Sep; 29(3):279-85.

31. Rantalainen T, Nikander R, Daly RM, Heinonen A, Sievanen H. Exercise loading and cortical bone distribution at the tibial shaft. Bone. 2011 Apr 1; 48(4):786-91.

32. Nikander R, Kannus P, Dastidar P, Hannula M, Harrison L, et al. Targeted exercises against hip fragility. Osteoporos Int. 2009 Aug; 20(8):1321-8.

33. Gomez-Bruton A, Gonzalez-Aguero A, Gomez-Cabello A, Casajús JA, Vicente-Rodríguez G. Is bone tissue really affected by swimming? A systematic review. PLoS One. 2013;8(8):e70119.