SPECIFIC SITES OF BONE EXPANSION DEPEND ON THE LEVEL OF VOLLEYBALL PRACTICE IN PREPUBESCENT BOYS

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ABSTRACT: The purpose of this study was to investigate the effect of 18 months of high and low levels of volleyball practice on bone acquisition. 130 prepubescent boys (mean age 11.4 ± 0.7) were divided into a high-level training group (HLG), low-level training group (LLG), and controls. Bone mineral content (BMC) and bone area at the whole body, lumbar spine L2-L4, femoral neck of the dominant leg, and right and left radius were measured using dual-photon X-ray absorptiometry. Enhanced BMC resulted from high-training volleyball activity in all measured sites except the third left and right distal radius, which is not modified by low-level training in prepubescent players but it was accompanied by a bone area expansion in radius and weight-bearing sites for the HLG, and in legs, whole right and left radius for the LLG. Significant improvement of skeletal tissues is associated with the intensity and duration of volleyball training.

KEY WORDS: BMC, bone area, lean mass, prepubescent boys

Accepted for publication 07.04.2013

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INTRODUCTION

Pre puberty is the period immediately prior to the onset of puberty, when growth and changes leading to skeletal maturity occur. Vigilant assessment of factors associated with enhancement of the skeletal tissues during this phase may be considered vital for the prevention of osteoporosis in adulthood [30]. The pre pubertal human skeleton is sensitive to the mechanical stimulation elicited by exercise and there is increasing evidence that regular weight-bearing exercise is an effective strategy for enhancing bone mineral levels throughout growth [4]. It is commonly accepted that physical exercise enhances bone mass gradually around pre puberty, with a peak occurring around late puberty [28], and stabilizes it thereafter.

The beneficial effects of exercise on bone mass have been the focus of intensive research. Indeed, physical activities in an aquatic environment such as swimming do not allow an improvement in bone density; swimmers move in a low gravity environment and this could explain why exercising in water early and regularly over the long term might abolish the osteogenic effect of sport [11]. Some high-level activities play a crucial role in bone development during growth [33,37], Riewald et al. showed that short, intense bouts of load-bearing exercise are more effective at increasing bone mass than are bouts of long-duration, low-intensity exercise [29]. Moreover, high-impact exercise based on jumps could increase the bone mineral content (BMC) in hips [18]. In fact, in prepubescent boys, intensive training, even during shorter periods of 12 minutes with the frequency of 3 times per week during 20 months, induces an increase in whole lean mass and gains in BMC and bone area at the femoral neck compared to controls [24].

Similarly, there was an increase in bone acquisition in hip and lumbar spine among prepubescent boys and girls after a few months jumping 100 times a day from a 61-cm height [15]. The high frequency of strains produced by jumping and the changeability of training impacts seem to improve their osteogenic effect as has been shown among prepubescent tennis players [34]. Volleyball is an osteogenic sport [2,3,7,30] that produces high strain stimulus at the upper and lower limbs due to the short, intensive, explosive, repetitive movement of jumps, lateral movement and ball striking induced not only by muscular contractions pulling on their bony attachments but also by the reaction forces produced by the jumps, which are three to six times the body weight [16]. The ground reaction forces, applied to the skeleton, produced during running are 3 times the body weight [14], and 10 times the body weight in elite gymnastics training [12]. The strain stimulus produced by repeated jumps, as in volleyball practice, promotes bone development better than single maximal jumps [17].

Male elite professional volleyball players have higher BMC in the axial skeleton and the dominant limb, and higher total lean mass compared to controls; they also have higher bone area in the dominant arm compared to the non-dominant arm [7]. In prepubescent boys, BMC was 17% greater in the tennis playing arm compared to the non-playing one [9]. Bone mass acquisition in childhood persists for several years, even when no longer trained; seventy-year-old habitual volleyball players have a larger tibial cross-sectional area than controls [30]. In the same population of our study, a high training level induces enhanced bone formation markers; this can be an indicator of improved acquisition of bone mass [8]. All these studies showed a beneficial effect of volleyball on bone acquisition after the pubescent period. To our knowledge, the effects of practising volleyball on bone mass, and their effects during growth, have not been studied.

The aim of this study was therefore: to study the influence of high and low volleyball practice levels on bone mass development in the growing skeleton among prepubescent boys compared to controls.

MATERIALS AND METHODS

Population. One hundred and thirty Tunisian boys were recruited from several schools and Tunisian volleyball clubs, all resident in the city of Sousse. All subjects, whose ages ranged from 10 to 12, were divided into three groups: 80 were volleyball players in 2 local clubs for at least 18 months in addition to physical education at school; 40 whose ages ranged from 10.9 to 12.1, and who completed 6 to 8 hours of training plus one competitive game per week, constituted the high-level training group (HLG); and 40 whose ages ranged from 10.5 to 11.9, and who completed 3 to 5 hours of training plus one competitive game per week, constituted the low-level training group (LLG). The other 50 subjects, whose ages ranged from 11.1 to 11.5, were assigned to the control group (C). They participated only in the compulsory physical education curriculum at school (two weekly sessions of 50 min each).

In general, training volleyball sessions lasted 1 h 30 min, including about 15-20 min of warm-up, low-intensity games and stretching exercises, 15-25 min of technical volleyball exercises (passing actions, smashing, blocking, and running with fast accelerations), 20-30 min of match practice, and 10 min of active recovery.

Each boy having a chronic disease that might affect either the physical exercise or bone metabolism was automatically excluded from this study. The study was approved by the Independent Ethics Committee of Farhat Hached Tunisian Hospital, and written informed consent was obtained from both parents of each participant.

Anthropometric measurement

Height and weight were measured in light indoor clothing without shoes. Height was measured to the nearest 0.01 m using a fathom; weight was assessed to the nearest 0.1 kg using a balance. Lean mass was measured by DXA, whole body densitometry.

Calcium intake

Calcium intakes of each subject were measured by the method of recording food for three consecutive days. At each meal the patient mentioned what he had eaten, then with the Bilnut SCDA Nutrisoft (Cerelles, France) program we calculated the amount of calcium consumed per day, expressed in $mg \cdot day^{-1}$.

Bone measurement

BMC, bone area and body composition (lean mass) were measured by dual-photon absorptiometry X-rays by DXA (Lunar Prodigy, model DXAP 2004, Madison, WI, USA, software version 3.6) for the whole body, lumbar spine L2-L4, femoral neck of the dominant leg, and right and left radius.

The assessment of efficiency was determined by repeated measurement of BMC, bone area and body composition of 10 children with a short time interval; it is less than 1% in the lumbar spine, femoral neck and whole body, and 2.5% in upper and lower limbs. Parameters of physical activity

VO₂max

The fitness level of children was evaluated by indirect calculation of the maximal oxygen uptake ($\dot{V}O_2max$) through the 20-m shuttle run test as devised by Luc Léger. Subjects were required to run back and forth on a 20-m course and be on the 20-m line at the same time that a beep is emitted from a tape. The frequency of the sound signals increases in such a way that running speed starts at 8.5 km \cdot h⁻¹ and is increased by 0.5 km \cdot h⁻¹ each minute. The length of time the subjects were able to run was recorded to calculate the $\dot{V}O_2max$ [20].

Basal physical activity level

Bratteby's questionnaire estimates the level of daily physical activities during a typical day, without volleyball training; the level of physical activity (PAL) was calculated by the following formula: PAL=TEE / MBR (PAL= physical activity level; TEE= total energy expended; MBR= basal metabolism) [5].

Peak power of lower limbs

We measured the peak power of lower limbs: squat jump (SJ), counter movement jump (CMJ) and horizontal jump (HJ) by using the Sargent test. [1].

Pubertal status

Tanner pubertal status was determined by serum rates of folliclestimulating hormone (FSH), luteinizing hormone (LH), and testosterone [19] and confirmed by a clinical method of recognized validity and reliability [10].

Only children with level of testosterone $< 0.6 \text{ (mUI \cdot ml^{-1})}$, FSH $< 4.6 \text{ (mUI \cdot ml^{-1})}$ and LH $< 4.8 \text{ (mUI \cdot ml^{-1})}$, corresponding to Tanner's stage I, were considered as prepubescent.

Biochemistry

For each participant, blood samples of 18 cc were taken between 8:00 am and 9:30 am and withdrawn following overnight fasting. Immediately, the serum was centrifuged (2100 g for 10 min), then it was isolated and frozen at -80° C.

We measured serum concentration of the hormone gonadotropins (FSH, LH and testosterone). Serum FSH and LH levels were measured in one laboratory by immunoradiometric (IRMA) assay using a commercial kit (IRMA STH IMMUNOTECH FRANCE); the intra-assay coefficients of variation (CVs) were <5% and the inter-assay CVs were <10%. The rates of testosterone were measured by the RIA method (RADIOIMMUNOASSAY kit IMMUNOTECH France). The intra-assay coefficients of variation (CVs) were <4% and the inter-assay CVs were <10%.

Statistical analysis

Anthropometric, age, dietary calcium intake, lean mass, hormonal and physical activity data were analyzed by one-way analysis of variance (ANOVA) followed by Fisher's LSD post hoc test between HLG, LLG and C. The analyses of covariance (ANCOVA) entering weight, height and gonadotropin hormone (FSH) as covariates were performed to evaluate BMC and bone area differences between HLG, LLG and C. Data were expressed as means \pm SD and differences were considered significant at the 0.05 level.

Statistical analysis was performed using Statistica software (version 6.0 2001, StatSoft, France).

RESULTS

Anthropometric variables. The average age, height, weight, lean mass, calcium intake and pubertal status data are summarized in Table 1. No age and dietary calcium intake differences were observed among the three groups; however, the high-level trained players were heavier and taller than the low-level trained ones and controls (p < 0.001).

TABLE I. ANTHROPOMETRIC DATA, AGE, CALCIUM INTAKE AND PUBERTAL GONADOTROPINS AND HORMONES IN HIGH-LEVEL TRAINING (HLG), LOW-LEVEL TRAINING (LLG) AND CONTROL (C) GROUPS

MEASURE	HLG	LLG		Comparison between groups		
	N = 40 N = 40	N = 40		а	b	С
Age (year)	11.5 ± 0.6	11.3 ± 0.7	11.3 ± 0.8	ns	ns	ns
Height (m)	1.51 ± 0.06	1.45 ± 0.04	1.43 ± 0.05	a***	b***	ns
Weight (kg)	40.1 ± 6.30	35.50 ± 4.60	33.2 ± 2.80	ns	b***	C***
BMI	17.75 ± 2.22	16.96 ± 1.99	16.13 ± 1.26	ns	b***	ns
Lean mass L.arm (kg)	1.39 ± 0.21	1.29 ± 0.29	1.22 ± 0.18	ns	b**	ns
Lean mass L.leg (kg)	5.19 ± 0.62	4.71 ± 0.52	4.39 ± 0.46	a***	b***	ns
Lean mass R.arm (kg)	1.30 ± 0.19	1.27 ± 0.21	1.09 ± 0.14	ns	b***	C***
Lean mass R.leg (kg)	5.15 ± 0.66	4.61 ± 0.53	4.22 ± 0.39	a***	b***	C**
Whole Lean mass (kg)	29.72 ± 2.76	27.25 ± 2.69	25.39 ± 1.91	a***	b***	C**
Calcium intake (mg · d⁻¹)	712.52 ± 82.75	710.10 ± 117.87	716.88 ± 85.05	ns	ns	ns
FSH (mUI · ml⁻¹)	3.80 ± 0.93	4.07 ± 0.82	3.27 ± 1.11	ns	b**	C***
LH (mUI · ml⁻¹)	2.13 ± 0.40	2.09 ± 0.58	2.08 ± 0.48	ns	ns	ns
Testosterone (mUI · ml ⁻¹)	0.26 ± 0.16	0.25 ± 0.18	0.25 ± 0.15	ns	ns	ns

Note: data represent mean \pm standard deviation, a: comparison between HLG and LLG, b: comparison between HLG and C, c: comparison between LLG and C, *: p <0.05, **: p <0.01, ***: p <0.001, ns: not significant.

TABLE 2. PHYSICAL CHARACTERISTICS OF THE STUDY SUBJECTS

MEASURE	HLG LLG		Control (C)	Comparison between groups		
	N = 40	N =40	N = 50	а	b	с
PAL	35.94 ± 2.76	36.41 ± 2.93	37.02 ± 2.67	ns	ns	ns
VO₂max (ml⋅kg⁻¹ min⁻¹)	50.81 ± 3.55	49.68 ± 2.98	47.65 ± 2.80	ns	b***	C*
HJ (cm)	166.64 ± 16.05	146.14 ± 6.93	137.54 ± 10.83	a***	b***	ns
SJ (cm)	26.74 ± 5.85	23.41 ± 5.36	20.19 ± 5.44	a**	b***	C*
CMJ (cm)	30.34 ± 5.78	26.05 ± 5.13	22.42 ± 4.72	a***	b***	C***

Note: data represent mean \pm standard deviation, a: comparison between HLG and LLG, b: comparison between HLG and C, c: comparison between LLG and C, *: p <0.05, **: p <0.01, ***: p <0.001, ns: not significant, PAL: physical activity level, HJ: horizontal jump, SJ: squat jump, CMJ: counter movement, HLG: high-level training group, LLG: low-level training group, C: control group

When comparing pubertal status between groups, all were similar in LH and testosterone rates, but the training groups had higher FSH rates than the control group. In fact, BMC and bone area results were therefore adjusted for FSH, height and weight.

The increase in lean mass was more marked in volleyball players than controls in all sites except in the non-dominant arm and leg, in which LLG had no difference in lean mass compared to controls. However, when comparing volleyball groups, lean mass in HLG was higher than in LLG only in left and right arms.

Physical fitness

The physical fitness values are displayed in Table 2. The basal physical activity did not show any significant difference among the three groups. Nevertheless, $\dot{V}O_2$ max, SJ and CMJ were slightly greater in the volleyball players than controls, while no difference was observed in HJ between LLG and controls. Based on the comparison between HLG and LLG, HLG had a higher HJ, SJ, and CMJ than LLG, while no difference was observed in $\dot{V}O_2$ max between HLG and LLG.

Densitometric measurement

BMC adjusted for FSH, height and weight

BMC is summarized in Table 3, Fig. 1A, and Fig. 1B. No significant difference was found in head between the three groups.

The HLG had a higher BMC than LLG and controls in all sites (whole body and lumbar spine, total hip and radius) except the third-left and right-distal radius, where there was no significant difference between groups.

Bone area adjusted for FSH, height and weight

Bone area is summarized in Table 4, Fig. 2A, and Fig. 2B. The bone area in HLG was higher in trochanter, whole femoral neck (Fig. 2A), whole body, lumbar spine (Table 4), ultra-distal and whole radius for the right and left forearms (Fig. 2B) than LLG and controls. No difference of bone area was observed in head (Table 4), or left third-distal radius (Fig. 2B), among the three groups. However, in femoral neck and right third-distal radius, bone area was higher only in HLG

than in controls. Bone area in LLG was higher in whole radius of right and left forearms (Fig. 2B), right and left legs, and whole body (Table 4) than controls.

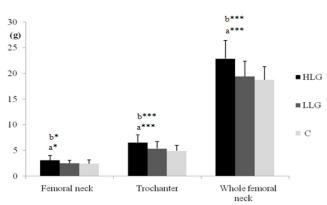
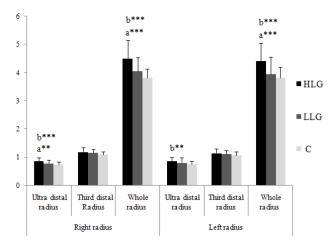
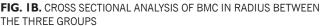


FIG. IA. CROSS SECTIONAL ANALYSIS OF BMC IN TOTAL HIP BETWEEN THE THREE GROUPS

Note: a: comparison between groups HLG and LLG, b: comparison between groups HLG and C; *: p < 0.05, ***: p < 0.001; HLG: high-level training group; LLG: low-level training group; C: control group





Note: a: comparison between groups HLG and LLG, b: comparison between groups HLG and C; **: p <0.01, ***: p <0.001; HLG: high-level training group; LLG: low-level training group; C: control group

TABLE 3. CROSS-SECTIONAL ANALYSIS OF BONE PARAMETERS (BMC) IN WHOLE BODY AND LUMBAR SPINE BETWEENTHE THREE GROUPS

MEASURE	HLG N = 40	LLG N =40	Control (C) N = 50	Comparison between groups		
(g)				а	b	С
Head	351.30 ± 34.25	336.90 ± 29.99	355.30 ± 29.42	ns	ns	ns
Right arm	75.80 ± 15.06	65.09 ± 12.56	62.21 ± 8.78	a***	b***	ns
Left arm	75.63 ± 15.89	68.14 ± 16.14	65.59 ± 8.55	a**	b***	ns
Right leg	299.9 ± 49.73	259.18 ± 37.82	244.10 ± 23.51	a***	b***	ns
Left leg	298.9 ± 54.69	258.28 ± 36.69	243.05 ± 24.72	a***	b***	ns
Whole body	1579.7 ± 190.5	1359.30 ± 151.40	1322 ± 101.82	a***	b***	ns
Lumbar spine	24.31 ± 3.77	21.31 ± 3.46	20.90 ± 2.98	a***	b***	ns

Note: data represent mean \pm standard deviation, a: comparison between HLG and LLG, b: comparison between HLG and C, **: p <0.01, ***: p <0.001, ns: not significant; HLG: high-level training group; LLG: low-level training group; C: control group

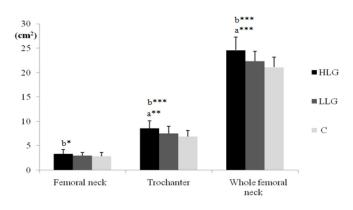


FIG. 2A. CROSS SECTIONAL ANALYSIS OF BONE AREA IN TOTAL HIP BETWEEN THE THREE GROUPS

Note: a: comparison between groups HLG and LLG, b: comparison between groups HLG and C; *: p < 0.05, **: p < 0.01, ***: p < 0.001; HLG: high-level training group; LLG: low-level training group; C: control group

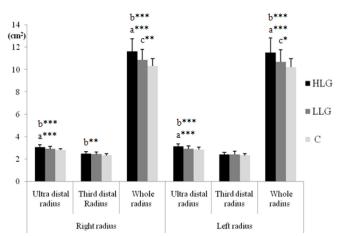


FIG. 2B. CROSS SECTIONAL ANALYSIS OF BONE AREA IN RADIUS BETWEEN THE THREE GROUPS

Note: a: comparison between groups HLG and LLG, b: comparison between groups HLG and C; *: p < 0.05, **: p < 0.01, ***: p < 0.001; HLG: high-level training group; LLG: low-level training group; C: control group



MEASURE	HLG	LLG	Control (C)	Comparison between group		groups
(cm ²)	N = 40	N =40	N = 50	а	b	С
Head	230.20± 38.60	233.09 ± 54.79	229.30 ± 32.87	ns	ns	ns
Right arm	111.60 ± 19.33	95.43 ± 14.35	91.88 ± 10.94	a***	b***	ns
Left arm	110.40 ± 19.09	96.48 ± 16.14	96.31 ± 11.19	a***	b***	ns
Right leg	300.10 ± 40.74	275.10 ± 29.18	257.10 ± 20.32	a***	b***	C***
Left leg	301.80 ± 40.11	271.80 ± 23.91	255.80 ± 17.6	a***	b***	C***
Wholebody	1650.90 ± 151.10	1476.70 ± 127.90	1417.30 ± 81.76	a***	b***	C**
Lumbar spine	31.44 ±3.29	28.83 ± 3.68	28.03 ±2.71	a***	b***	ns

Note: data represent mean ± standard deviation, a: comparison between HLG and LLG, b: comparison between HLG and C, *: p <0.001, ns: not significant; HLG: high-level training group; LLG: low-level training group; C: control group

DISCUSSION

The current investigation shows that the effect of volleyball participation on BMC and bone area is already beneficial in prepubescent boys, depending on the frequency and the intensity of practice. The main finding in the present study was that young volleyball players trained with higher intensity and frequency than their counterparts, and show higher lean mass, BMC and bone area in most sites. Moreover, as expected, prepubescent LLG had higher lean mass in whole body and right arm and leg, and higher bone area in whole right and left radius than controls. In this study, there was no significant difference in dietary activities and calcium intake between groups at the time of measurement. Nevertheless, volleyball players were significantly taller, and heavier than their sedentary counterparts; thus, there may have been bias introduced into the study based on selection. To account for this potential confounding and bias associated with selection, several important variables (height, weight and FSH) to bone mineral accrual in prepubescents were controlled for, in which the HLG, LLG and controls may have differed when being measured. This was done by incorporating these variables as covariates that describe the relationship between volleyball practice and bone response.

The practice of weight-bearing exercise, especially volleyball activity, exerts positive effects on the skeleton, highlighted by the increase of BMC and bone area in the most used sites [32]. Likewise, tennis participation is associated with increased lean mass and bone mass in the playing arm [34]. Volleyball training is a succession of exercises based on vertical and horizontal jumps, which develops physical characteristics. The enhanced VO2max and horizontal and vertical jumps rise in parallel with the enhanced height values of the training frequency. The LLG's physical performance of jumps and $\dot{V}O_2$ max is slightly improved by low-intensity training, while the HLG generates significant high $\dot{V}O_2$ max and jumps compared to controls. There is no significant difference in dietary activities and calcium intake between groups.

Like many sports, the practice of volleyball in adolescence and adulthood leads to an increase in lean mass and to the development of BMC and bone area in the most used sites [7]. The variation of this gain is proportional to the intensity of the practice.

The beneficial effect of volleyball practice on bone development is well documented in lean mass among adults [2,3,7]. Our study provides further evidence indicating that this latter develops disproportionately

among high and low training groups, and increases more importantly in all sites of HLG compared to controls, but for the LLG this development is obviously seen only in the most used sites – the dominant leg and arm. Similar results were found by Vincent Rodriguez et al.; after one year of soccer practice, there was an increase in total body lean mass among prepubescent players compared to controls [35].

In addition to previous knowledge, a significant bone gain is evidenced by BMC and bone area. Except for the third left and right distal radius, there was an enhanced BMC in all measured sites, after intensive training; nevertheless, low-intensity training is not known to have a greater impact on BMC compared to controls. This enhanced BMC is the outcome of an 18-month period of intensive training. Therefore, our results are in agreement with those reported by McWhannell et al., who observed an increase in BMC in total body among prepubescent children; however, our results were partially not in accordance with the latter data, which showed no variations in BMC, in lumbar spine and femoral neck after high-level exercise during 9 weeks [26]. Thus, the gain of BMC in the lumbar spine and femoral neck needs more than a 9-week period of intensive training.

Nonetheless, Fuchs et al. also found an increase in BMC among prepubescent children in the femoral neck (4.5%) and lumbar spine (3.1%) after a high-level exercise of jumps during a 7-month period [15]. Similarly, Linden et al. found among prepubescent girls outstanding development of BMC in lumbar spine L2-L4 and legs owing to a 2-year training programme based on osteogenic exercises compared to controls [21]. The gain in BMC was also controlled by the nature of the practised sport. Lethonen-Veromaa et al. found an increase of BMC in the femoral neck and lumbar spine among prepubescent gymnasts compared to runners and controls [22]. Similarly, Nanyan et al. found an increase in BMC among prepubescent girls, practising gymnastics, judo, and dance in the third-distal of non-dominant radius compared to controls [27]. In contrast to our results, one year of soccer practice induced no variations in BMC in prepubescent boys aged 10-12 years in skull, dominant arm, supporting leg, kicking leg, whole body, total hip and lumbar spine [38].

High and low level of osteogenic exercises in prepubescent children not only develops BMC but also can increase bone area [23]. This significant bone gain is explained by an increase in bone area after intensive training in all measured sites, except the femoral neck and the third-distal radius. Moreover, as expected, a substantially low exercise volume contributes to enhanced bone area in whole body, right and left legs, and right and left whole radius.

In general, there are numerous studies that have showed increases of BMC and bone area in the upper limbs as well as in the weight-bearing bones associated with prepubertal stage in various sports [24, 28, 34], but the originality of the present work concerns specifically bone mass in prepubescent volleyball players at different levels of practice. The high BMC and bone area of the upper limbs and the weight-bearing sites observed in the prepubescent volleyball players suggest that bone stimulation was produced by a high-impact loading physical exercise during the prepubertal stage. Therefore, our results support the idea that volleyball has an osteogenic effect [3,7] and agree with those reported by Laing et al., who found increases in the dominant forearm's bone area among high-level trained and low-level trained prepubescent girls, who had been practising artistic gymnastics for two years, compared to their chronological age counterparts [23]. Also, the high-level training group promoted more additional gains in the forearm bone area compared to the low-level training group. However, the rise in the level of volleyball training enhanced bone area acquisition. Moreover, our data suggest that volleyball may elicit bone expansions in the whole body, lumbar spine, femoral neck and radius similar to those described in other studies performed in young female soccer players, who clearly benefited from the improvement of the trabecular bone area at the trochanter [6, 31]. Some studies on intervention children showed that 8 months of less vigorous bone-loading programme enhanced areal bone at the trochanter (4.4% vs 3.2%) [25]. However, other studies showed no obvious bone area variations in prepubescent soccer players [38], suggesting that the intensity of soccer training could be considered insufficient for optimal bone expansion.

As far as radius is concerned, the usual findings are either no variation in bone area, as in prepubescent soccer players, or an increase of bone area in the third-distal radius of the dominant arm, as in prepubescent female gymnasts [13], and in prepubescent male gymnasts [36]. Our results, which showed an increase in bone area among HLG compared to controls in all measured sites except at the third-distal radius of the non-dominant arm, were in agreement with these latter data. Unlike what we found, Ward et al. noted no variations in radius distal area and lumbar spine among prepubescent gymnasts compared to controls [36]. Similarly, there were no obvious differences in bone area in lumbar spine after a 7-month jumping programme [15]. The difference of results was revealed by the variation of mechanical stress induced by every sport modality in the stimulated region. We can therefore conclude that the bone area response was more directly related to the intensity of bone deformations rather than the training duration.

The trabecular bone appears to be particularly more sensitive to the mechanical stress elicited by volleyball actions than cortical bone, as shown by the lack of change in bone area at the femoral neck and the third-distal radius, which are predominantly composed of cortical compact bone, whereas at the trochanter and ultra-distal radius, bone is mainly composed of trabecular bone. Our results are in line with those of Ducher et al., in which tennis practice presented similar impact characteristics as volleyball practice [9]. They compared children's bone response at trabecular (ultra-distal region) and cortical (the mid and third-distal region) skeletal sites, to that of young adults at the ultra-distal radius. Children and adult tennis players found similar side-to-side differences in BMC and bone area, but at the mid and third-distal radius the asymmetry was much greater in adults than in children [9]. This affinity of trabecular cells to external impacts generated by the practice allowed the development of epiphyseal bone lengthening and increasing volume, which could contribute to the tallness of young volleyball players compared to controls.

When considering high and low levels of prepubertal groups, both BMC and bone area values were significantly higher for the volleyball players than the controls without variation in BMD. This can be explained by the lack of a balanced diet in general and particularly calcium intake (approximately 800 mg·day⁻¹), which is lower than the Official Institute of Medicine (IOM) recommendations $(1300 \text{ mg} \cdot \text{day}^{-1})$, and may not enhance hormonal impregnations in the prepubescent period.

CONCLUSIONS

The present investigation indicates that enhanced BMC resulting from high-impact volleyball activity is not mediated or promoted by low-level training in prepubescent players but is accompanied by a little muscular development in lower and dominant upper members, as well as by bone area expansion in legs, total right and left radius. However, despite the lack of calcium intake, our study implies that high-level-trained volleyball players have increased BMC, bone area and lean mass in arms and legs, whose magnitude depends on the number of weekly hours devoted to volleyball. Indeed, it is recommended for prepubescent volleyball players to have sufficient means of calcium intake leading to optimal skeleton mineralization. Hence, the significant improvement of both skeletal and muscle tissues is associated with the intensity and duration of volleyball training, which is evidence of the unique aspect of our findings that were not provided elsewhere.

Acknowledgements

The authors gratefully acknowledged the volleyball players and sedentary boys who participated in this study with great enthusiasm. Financial support was obtained from the Laboratory of Physiology and Functional Explorations, Faculty of Medicine Sousse, and the Department of Rheumatology, Farhat Hached Hospital, Sousse, Tunisia.

Conflicts of interest: no conflicts of interest

REFERENCES

- 1. Anderson F.C., Pandy M.G. Storage and utilization of elastic strain energy during jumping. J. Biomech. 1993;26:1413-1427.
- 2. Alfredson H., Nordstrom P., Lorentzon R. Bone mass in female volleyball players: a comparison of total and regional bone mass in female volleyball players and nonactive females. Calcif. Tissue Int. 1997;60:3383-42.
- 3. Alfredson H., Nordstrom P., Pietila T., Lorentzon R. Long-term loading and regional bone mass of the arm in female volleyball players. Calcif. Tissue. Int. 1998;62:303-308.
- 4. Burrows M. Exercise and bone mineral accrual in children and adolescents. J. Sport. Sci Medic. 2007;6:305-312.
- 5. Bratteby L.E., Sandhagen B., Fan H., Samuelson G. A 7-day activity diary for assessment of daily energy expenditure validated by the doubly labelled water method in adolescents. Eur. J. Clin Nutr. 1997;51:585-591.
- 6. Bobbert M.F., Huijing P.A., Van Ingen Scheanau G.J. Drop jumping. I. The influence of jumping technique on the biomechanics of jumping. Med. Sci. Sport. Exer. 1987;19:332-338.
- 7. Calbet J.A., Diaz Herrera P., Rodriguez L.P. High bone mineral density in male elite professional volleyball players. Osteoporos. Int. 1990;10:468-474.
- 8. Chaari H., Zouch M., Denguezli M., Bouajina E., Zaouali M., Tabka Z. A high level of volleyball practice enhances bone formation markers and hormones in prepubescent boys. Biol. Sport 2012;29:303-309.
- 9. Ducher G., Daly R.M., Bass S.L. Effects of 17. Gruodyte R., Jurimae J., Saar M.,

repetitive loading on bone mass and geometry in young male tennis players: a quantitative study using MRI. J. Bone Miner. Res. 2009;24:1686-1692.

- 10. Duke P.M., Litt I.F., Gross R.T. Adolescents' self assessment of sexual maturation. Pediatrics 1980;6:918-20.
- 11. Długołęcka B., Czeczelewski J., Raczyńska B. Bone mineral content and bone mineral density in female swimmers during the time of peak bone mass attainment. Biol. Sport 2011;28:69-74.
- 12. Daly R.M., Rich P.A., Klein R., Bass S. Effects of high-impact exercise on ultrasonic and biochemical indices of skeletal status: A prospective study in young male gymnasts. J. Bone Miner. Res. 1999;14:1222-1230.
- 13. Dowthwaite J.N., Flowers P.P., Spadaro J.A., Scerpella T.A. Bone geometry, density, and strength indices of the distal radius reflect loading via childhood gymnastic activity. J. Clin. Densitom. 2007;10:65-75.
- 14. Engsberg J.R., Lee A.G., Patterson J.L., Harder J.A. External loading comparisons between able-bodied and below-kneeamputee children during walking. Arch. Phys. Med. Rehabil. 1991;72:657-666.
- 15. Fuchs R.K., Bauer J.J., Snow C.M. Jumping improves hip and lumbar spine bone mass in prepubescent children: a randomized controlled trial. J. Bone. Miner. Res. 2001;16:148-156.
- 16. Grimston S.K., Willows N.D., Hanley D.A. 22. Lehtonen-Veromaa M., Mottonen T., Mechanical loading regime and its relationship to bone mineral density in children. Med. Sci. Sport. Exer. 1993; 25:1203-1210.

Maasalu M., Jurimae T. Relationships between areal bone mineral density and jumping height in pubertal girls with different physical activity patterns. J. Sport. Med. Phys. Fitness 2009;49:474-479.

- 18. Gunter K., Baxter-Jones A.D., Mirwald R.L., Almstedt H., Fuchs R.K., Durski S., Snow C. Impact exercise increases BMC during growth: an 8-year longitudinal study. J. Bone Miner. Res. 2008;23:986-993.
- 19. Jaakkola T., Ding Y.Q., Kellokumpu-Lehtinen P., et al. The Ratios of Serum Bioactive/Immunoreactive Luteinizing Hormone and Follicle-Stimulating Hormone in Various Clinical Conditions With Increased and Decreased Gonadotropin Secretion: Re-evaluation by a Highly Sensitive Immunometric Assay. J. Clin. Endocrinol. Metab. 1990;70:1496-1505.
- 20. Leger L.A., Mercier D., Gadoury C., Lambert J. The multistage 20 metre shuttle run test for aerobic fitness. J. Sport. Sci. 1988;6:93-101.
- 21. Linden C., Ahlborg H., Besjakov J., Gardsell P., Karlsson M. A school curriculum-based exercise program increases bone mineral accrual and bone size in prepubertal girls: two-year data from the pediatric osteoporosis prevention (POP) study. J. Bone. Miner. Res. 2006;21:829-835.
- Irjala K., Nuotio I., Leino A., Viikari J. A 1-year prospective study on the relationship between physical activity, markers of bone metabolism, and bone acquisition in peripubertal girls. J. Clin.

Endocrinol. Metab. 2000;85:3726-3732.

- Laing E.M., Wilson A.R., Modlesky C.M., O'Connor P.J., Hall D.B., Lewis R.D. Initial years of recreational artistic gymnastics training improves lumbar spine bone mineral accrual in 4- to 8-year-old females. J. Bone. Miner. Res. 2005;20:509-519.
- MacKelvie K.J., Petit M.A., Khan K.M., Beck T.J., McKay H.A. Bone mass and structure are enhanced following a 2-year randomized controlled trial of exercise in prepubertal boys. Bone 2004;34:755-764.
- 25. McKay H.A., Petit M.A., Schutz R.W., Prior J.C., Barr S.I., Khan K.M. Augmented trochanteric bone mineral density after modified physical education classes: a randomized school-based exercise intervention study in prepubescent and early pubescent children. J. Pediatr. 2000;136:156-162.
- McWhannell N., Henaghan J., Foweather L., Doran D., Batterham A., Reilly T., Stratton G. The effect of a 9-week physical activity programme on bone and body composition of children aged 10-11 years: an exploratory trial. Int. J. Sports Med. 2008;29:941-947.

- 27. Nanyan P., Prouteau S., Jaffre C., Benhamou L., Courteix D. Thicker radial cortex in physically active prepubertal girls compared to controls. Int. J. Sports Med. 2005;26:110-115.
- Nebigh A., Rebai H., Elloumi M., Bahlous A., Zouch M., Zaouali M., Alexandre C., Sellami S., Tabka Z. Bone mineral density of young boy soccer players at different pubertal stages: relationships with hormonal concentration. Joint Bone Spine 2009;76:63-69.
- 29. Riewald S., Roetert E. Bone of Contention: What Exercises Increase Bone Strength? Strength Cond. J. 2004;26:46-47.
- Rantalainen T., Linnamo V., Komi PV., Selanne H., Heinonen A. Seventy-yearold habitual volleyball players have larger tibial cross-sectional area and may be differentiated from their age-matched peers by the osteogenic index in dynamic performance. Eur. J. Appl. Physiol. 2010;109:651-658.
- Singleton M.C., LeVeau B.F. The hip joint: structure, stability, and stress. Phys. Ther. 1975;55:957-973.
- 32. Schwarz P., Courteix D., Karlsson M. Exercise and bone. Eur. J. Sport Sci. 2006;6:141-144.

- 33. Silva C.C., Goldberg T.B., Teixeira A.S., Dalmas J.C. The impact of different types of physical activity on total and regional bone mineral density in young Brazilian athletes. J. Sport. Sci. 2011;29:227-234.
- Sanchis-Moysi J., Dorado C., Olmedillas H., Serrano-Sanchez J.A., Calbet J.A. Bone mass in prepubertal tennis players. Int. J. Sports Med. 2010;31:416-420.
- Vicente-Rodriguez G., Ara I, Perez-Gomez J., Serrano-Sanchez J.A., Dorado C., Calbet J. High femoral bone mineral density accretion in prepubertal soccer players. Med. Sci. Sport. Exerc. 2004;36:1789-1795.
- Ward K.A., Roberts S.A., Adams J.E., Mughal M.Z. Bone geometry and density in the skeleton of pre-pubertal gymnasts and school children. Bone. 2005;36:1012-1018.
- Shin Y.H., Jung H.L., Kang H.Y. Effects of taekwondo training on bone mineral density of high school girls in korea. Biol. Sport 2011;28:195-198.
- Zouch M., Jaffre C., Thomas T., Frere D., Courteix D., Vico L., Alexandre C. Long-term soccer practice increases bone mineral content gain in prepubescent boys. Joint Bone Spine 2008;75:41-49.