Changes in bone mineral density during puberty

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Abstract

Puberty is the fundamental period for bone mass acquisition. In this period mineralization is found to increase with levels of high bone formation.. The objective of this cross-sectional study was to determine the changes in bone mineral density (BMD) and growth parameters for healthy pubertal males and females at different pubertal stages in Mosul city/Iraq. In addition, we aimed to detect the relationship between BMD, age, pubertal stage and growth parameters, and to reveal the most important determinant of BMD in the pubertal period. BMD of the lumbar spine was performed by dual-energy X-ray absorptiometry in (177) healthy pubertal children and adolescents (96 males, 81 females), aged (9.9-20.2) years. Growth parameters (Weight and height) were measured, BMI were calculated. Pubertal stages were assessed and the subjects were subdivided into 5 stages (Tanner stages of puberty for males and females). There was a significant effect of age and puberty on BMD. Females had significantly higher BMD across all age groups because females enter puberty earlier than males. When gender comparison was done according to pubertal stages, males had higher values for BMD in all pubertal stages , but without significant differences between them except in stage III, which indicated that boys gain more BMD than girls at this stage. Both sexes showed the main increments in BMD between stage IV and V. The major independent determinant of BMD in both sexes was pubertal stage. BMD of males was also highly correlated with growth parameters, but no such correlations for females. values in the study group were significantly lower than Western normative values, with Z scores for girls was (-1.2±1.2) and for boys was (-1.4±1.1). In conclusion, bone mass increased throughout puberty in both sexes and there was a strong evidence that pubertal development was consistent and independent predictors of BMD in healthy children and adolescents.

التغيرات الحاصلة في الكثافة المعدنية للعظام خلال مرحلة البلوغ

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الملخص

تعتبر مرحلة البلوغ أهم فترة يتم فيها اكتساب الكثافة المعدنية للعظام ، لذلك تم دراسة التغيرات الحاصلة في الكثافة المعدنية للعظام في منطقة الفقرات القطنية بواسطة جهاز الدكسا (جهاز فحص الكثافة المعدنية للعظام) لعدد من الأطفال و المراهقين الأصحاء في مدينة الموصل في العراق و بلغ عددهم الكلي (177) مشارك بضمنهم (96) ذكر و (81) إنثى تتراوح أعمارهم ما بين (9-20) سنة و قد تم تصنيفهم حسب العمر و حسب مراحل البلوغ (و هي خمس مراحل بداية من المرحلة الأولى و هي مرحلة قبل البلوغ إلى المرحلة الخامسة و هي مرحلة البلوغ الكامل) . و كان الهدف من هذه الدراسة تحديد التغيرات الحاصلة في الكثافة المعدنية للعظام في مختلف مراحل البلوغ و علاقة هذه التغيرات مع العمر و مراحل البلوغ و مؤشرات النمو (الوزن و الطول و كتلة الجسم) و إيجاد العامل الأساسي الذي يحدد هذه التغيرات . وجد في هذه الدراسة أن هناك زيادة ملحوظة في

الكثافة المعدنية للعظام مع تقدم العمر و تقدم مراحل البلوغ. وعند تصنيف المشاركين حسب الفئات العمرية وجد إن الإناث أكثر كثافة معدنية من الذكور بسبب دخولهن مرحلة البلوغ قبل الذكور . وعند تصنيفهم حسب مراحل البلوغ ، وجد أن الذكور أكثر كثافة من الإناث لكن بدون فرق معنوي بينهم عدا في المرحلة الثالثة . ولوحظ في كلا الجنسين أن الزيادة الرئيسية للكثافة المعدنية للعظام بين المرحلة الرابعة و الخامسة من مراحل البلوغ . و قد وجد أن مرحلة البلوغ هي العامل الأساسي الذي يحدد التغيرات في الكثافة المعدنية للعظام في كلا الجنسين. وأن مؤشرات النمو من العوامل المهمة التي تحدد الكثافة المعدنية للعظام بالنسبة للذكور و ليس الإناث. ولوحظ أن قيم الكثافة المعدنية لعظام الأطفال والمراهقين في مدينة الموصل اقل من قيم الكثافة المعدنية للعظام لدى الأط فال والمراهقين الامريكيين (وهي قيم موجودة في قاعدة البيانات في جهاز الدكسا، حيث يقوم الدكسا بمقارنة القيم المقاسة مع القيم الموجودة في قاعدة البيانات. وتعطى النتيجة على شكل انحراف معياري يسمى (z-score).

Introduction

BMD is a medical term referring to the amount of matter per square centimeter of bones. Epidemiological studies have consistently shown that BMD is a primary predictor of osteoporosis and fracture risk [(1)]. Osteoporosis is a growing health problem and because there are no safe and effective ways to enhance BMD once osteoporosis has occurred [(2,3)], a primary prevention must aim at increasing bone mass acquisition before late adolescence, this has led to increasing interest in assessing BMD in children and adolescents [(4,5)]. BMD increases during childhood and adolescence until the peak bone mass (PBM) is achieved, PBM is defined as the maximal BMD that is accrued during growth and development plus subsequent consolidation that continues during early adulthood [(5,6)], PBM is regarded as the bone bank for the remainder of life [(7)]. The precise age at which PBM is acquired is still unknown [(8)]**.** Many factors, more or less dependent on each other, are known to influence bone mass accumulation during growth. These determinants classically include: genetic determinants up to 80% [(9)], whereas the remaining 20% is modulated by environmental factors and sex hormone levels during puberty [(10)]. Environmental factors including, amount of weight-bearing physical activity [(11,12)], dietary calcium [(13)], and smoking which is variably associated with a reduced BMD [(3)]. The major systemic hormones involved in the regulation of bone metabolism and growth during childhood and adolescence are: growth hormone, insulin-like growth factor(IGF-I), estrogen and androgen [(14,15)]. Puberty in the human is a unique and integrated transition from childhood to young adulthood. It involves major physical, emotional and psychological changes that culminate in the attainment of fertility [(16,17)]. The dramatic bone growth during puberty encompasses three distinct but integrated processes: accelerated linear growth, bone maturation, and rapid acquisition of bone mass [(18,19)].

Subjects and methods

This study represents a cross-sectional study, carried out from November 2010 to April 2011. Volunteer healthy Asian children and adolescents [177], [96] boys and [81] girls aged [20,2] to [9,9] years, residents of Mosul city, Iraq, the subjects were selected from different sources: outpatient clinic at Ibn-sina teaching hospital, relatives of the staff members in the same hospital, students at the 1st class in the college of medicine and students at the nursing secondary school. These subjects were invited to take part in this study and interviewed with their parents to explain the methods used and seek consent. A questionnaire was administered to all subjects and their families. Exclusion criteria were : any chronic disease associated with low

bone mass, drug consumption (as corticosteroids, anticonvulsants, calcium & vitamin supplements, antacids), smoking which has been linked to low BMD in adolescents [3], doing exercises in a professional manner, since investigations indicate that programmed sporting activities demonstrate the greatest increases in bone mass [20], subjects who were below the 5th percentile or above 95th percentile for weight and for height were excluded [21], irregularity of menstrual periods (history of menstrual irregularities has been consistently associated with lower bone mineral density in premenopausal women)[15], a significant fracture history- The Pediatric Position Development Conference (PDC) of the International Society of Clinical Densitometry- has defined a clinically significant fracture history as "one long bone fracture of the lower extremity, two or more long bone fractures of the upper extremity or a vertebral fracture [22] and family history of osteoporosis, there are evidences for a strong familial resemblance of bone density [9]. Weight was measured with an electronic scale with the subjects wearing light clothes without shoes. Height was measured by asking the subject to stand without shoes and erect, by using a vertical scale.

Body mass index (BMI) was calculated by using the following formula:

BMI (kg/m²) = weight (in kilogram) / height² (in meters)[23].

pubertal development was evaluated by self-assessment of breast and pubic hair stage in girls and genitalia and pubic hair stage in boys, according to the method of Tanner. Subjects were classified as Tanner stage I, II, III, IV, and V [24].

BMD (expressed in g/ cm²) of lumber spine (L1-L4) were measured with dual energy x-ray absorptiometry (DXA). DXA scans were performed using Hologic (Discovery W-S/N 83903,USA). The most commonly used densitometric technique for children throughout the world is DXA due to its precision, short scan times, very low radiation exposure, painless, low cost and robust pediatric reference data [25,26,28,47]. Statistical analysis was carried out by using the statistical Package for the Social Science (SPSS Inc, Chicago, version 11.5). As descriptive statistics, mean, standard deviation (SD), minimum and maximum limits were given for the data. Analyses were performed for males and females separately, differences between the two groups were assessed by independent t-test. The effects of age and puberty on the parameters within each gender were assessed using one-way analysis of variance (ANOVA), followed by Duncan´s multiple range tests for each gender separately. The association between BMD and different predictor variables was tested with multiple regression. P-value lower than 0.05 was considered statistically significant.

Results

Table (1): anthropometric characteristics of the study group.

parameter	min	max	Mean ±SD
Age (year)	9.9	21.2	13.92±2.8
Weight (Kg)	22.8	92.5	47.7±13.5
Height (cm)	129	184	154±10.6
BMI (Kg/m²)	13.37	27.29	19.75±3.45
BMD (g/cm²)	0.403	1.160	0.721±0.16

Table (2): comparison of BMD values between males and females in each pubertal stage

parameter	Male	Female	p-value
Stage I	0.518±0.03	0.512±0.04	0.673 (NS)
Stage II	0.598±0.03	0.583±0.03	0.165 (NS)
Stage III	0.702±0.05	0.658±0.04	0.005 *
Stage IV	0.801±0.06	0.776±0.06	0.234 (NS)
Stage V	0.927±0.11	0.928±0.07	0.975 (NS)

There was no significant differences between both sexes across all pubertal stages except in stage III.

Table (3): Comparison of parameters between different pubertal stages in male subjects :

parameter	I	II	III	IV	V
age(year)	10.98 (a)	12.66 (b)	13.96 (c)	15.89 (d)	17.7 (e)
Weight(Kg)	33.61 (a)	43.6 (b)	51.02 (c)	57.64 (d)	66.6 (e)
Height(cm)	142.39 (a)	150 (b)	158.95 (c)	164.85 (d)	168.56 (e)
BMI(Kg/m²)	16.53 (a)	19.32 (b)	20.2 (b)	21.14 (b)	23.34 (c)
BMD(g/cm²)	0.518 (a)	0.598 (b)	0.702 (c)	0.801 (d)	0.927 (e)

The means that have similar letters mean that there was no significant difference between them according to Duncan´s test at the subset for alpha=0.05. In table (3), all parameters increased with increments in pubertal stages, and they exhibited significant differences across all pubertal stages.

Table (4): Comparison of parameters between different pubertal stages in female subjects :

parameter	I	II	III	IV	V
age(year)	10.4 (a)	11.48 (b)	11.9 (b)	13.66 (c)	17.05 (d)
Weight(Kg)	32.67 (a)	37.18 (a,b)	42.29 (b,c)	45.43 (c)	52.68 (d)
Height(cm)	140.11 (a)	146 (b)	148.65 (b,c)	152.11 (c)	156.62 (d)
BMI(Kg/m²)	16.53 (a)	17.37 (a,b)	19 (b,c)	19.6 (c,d)	21.4 (d)
BMD(g/cm²)	0.512 (a)	0.583 (b)	0.658 (c)	0.776 (d)	0.928 (e)

Table (4) females showed an increase in BMD that parallel an increase in age, body weight, height, and BMI. Subjects in either Tanner stage IV or V had higher values for BMD than those at earlier stages of sexual development.

Table (5): Comparison of BMD between males and females in each age group

	male		female		
Age groups(years)	No. of subjects	BMD g/cm²	No. of subjects	BMD g/cm²	p-value
< 10	2	0.504	3	0.486	0.346(NS)
10-10.9	11	0.536	13	0.598	0.011*
11-11.9	5	0.595	13	0.618	0.771(NS)
12-12.9	8	0.613	12	0.658	0.123(NS)
13-13.9	18	0.638	11	0.756	0.009*
14-14.9	7	0.675	5	0.796	0.037*
15-15.9	12	0.770	8	0.808	0.688(NS)
16-16.9	11	0.806	11	0.915	0.050*
17 and >	21	0.924	5	0.920	0.837(NS)

In table (5) the BMD values was described according to age groups for each gender separately, they were subdivided into nine age groups at one-year intervals in each. In this study, there were no significant differences in BMD between the age 17 and more than 17 years(the upper limit of this study was 22.2 year), so the age of 17 years and more was considered as one group. The fact that after the age of 16 years, most children have completed their skeletal growth may perhaps explain the low variation in this age group[29]. This table shows a comparison of BMD between males and females in each age group by using independent sample t-test. There were no significant differences between boys and girls until the age of 10. After the age of 10, females presented higher values for BMD than males in all other age groups, probably because of the earlier onset of puberty in females, the significant differences were at the age of 10, 13-15 and at 16 years. By the age of 17 years and more, the male values were greater than female, but the difference was not significant. The effects of age on the BMD values within each gender were assessed using one-way analysis of variance (ANOVA), followed by Duncan´s multiple range tests for each gender separately. We found that BMD increased significantly with age. For males, the increase occurred especially after 15 years. There were another periods of acceleration in lumbar spine BMD, in the age group 16-17 year for males. The lowest variation was present in the age group 11-12 year. For female subjects, regular increase in BMD values in the different age groups was found. The lowest variation was present in the age group 10-11 and 16-17 year, while the maximal increase occurred around the age of 13 years in girls (approximately two years later in boys, around the age of 15). There were another periods of acceleration in lumbar spine BMD, at the beginning of puberty (after 10 years age) and from 15-16 years. Multiple regression analysis was performed in order to evaluate the

impact of the combination of several parameters on BMD values. Age, weight, height, BMI and Pubertal stages were entered into each regression model as continuous independent predictors, and BMD as dependent variable. In both sexes, the major independent determinant of BMD was the pubertal stage (p-value <0.001). For males, in addition to pubertal stage, weight, height and BMI were the factors with significant influence on BMD of lumber spine.

Ethnic differences: Z scores in the study group were derived through the densitometer software using a American database as reference. For girls, the mean Z scores was: (-1.2±1.2). For boys, the mean Z scores was: (-1.4±1.1). these Z scores demonstrated that mean BMD in healthy subjects in Mosul city is lower than that of age- and gender-matched American children and adolescents.

Discussion

This study provided gender-specific lumber spine BMD values, expressed in discrete age and pubertal stage subgroups, were measured in pubertal males and females aged between 9.9-21.2 years. The well-described pubertal increments in BMD at the lumber spine were observed, concomitant with the significant increase in body dimensions and its relationship with maturation of secondary sexual characteristics. A number of different factors are listed as being important for maximum growth of bone mineral density during puberty. Among these factors, those that stand out are contributions of a genetic nature, alterations to the body's dimensions, weight and stature, the hormonal profile leading to skeletal and sexual maturity, the practice of physical activity by the adolescents and sufficient calcium ingestion during this age range and that are reflected in intense bone mineralization [30,31]. It is evident that during puberty two events occur almost simultaneously. One phenomenon is the sustained burst in physical growth characterized by substantial increases in stature and the other is the attainment of peak bone mass. Apparently both situations are mediated by a similar hormonal cascade including GH, IGF-1 and sex steroids [30]. In our study, we found that the main increase in height for the male subjects occurred between stage II and III, while for females between stage I and II, and both sexes showed the main increase in BMD between stage IV and V, that is, there was a delay between the maximum gain in height and BMD in both sexes, and this in agreement with findings of Theintz et al [31], Fournier et al [32] and Bailey et al [33] who noticed a one-year delay between peak height velocity and BMD accrual. This delay occurs between Tanner stages II to III and III to IV, or between the ages of ~12 and 13 in girls, and ~14 and 15 in boys. They also showed that about 26% of peak bone mass is acquired during the 2-year period across peak height velocity, suggesting a clear relationship between the increase of sex steroids, linear growth and bone growth. Also we found that weight, height and BMI were the factors with significant influence on BMD of males, but not for females. These results were similar to what was previously reported by Silva et al [34], Boot et al [34], Ahmed et al [36] who correlated BMD with anthropometric variables, body weight and height in the boys. Other studies observed the dependence between BMD and growth parameters, but in different pattern; Goksen et al [37] found the correlation of BMD with height in both sexes, while Arabi et al [38], Hasanoğlu et al [39] and Yilmaz et al [40] found that

weight was one of the important factors influencing BMD in both sexes. They stated that body weight is the main source of bone load and this determines the mechanical strength of the bone. Discrepancies in the effect of height, weight and BMI on BMD between studies, may be explained by a variety of factors. These include the fact that some studies did not take into account the pubertal stages, and the different techniques for analyzing the data, in addition, DXA devices from different manufacturers might not give identical results, because of differences in calibration. For males as well as females, a progressive increase in BMD values in the different age groups was found, this in agreement with the findings of other authors [35,39,41], this increase of BMD with age, suggesting that the increase in lumbar spine BMD does reflect a real increase in mineralization, and is not merely a result of accelerated growth [3]. There was a variation in the velocity of mineral gain that occurred at different ages. After an initial period with slight increases in BMD values, a period of rapid growth and accumulation of bone mass in the lumbar spine was observed, especially striking around 13 and 15 years of age in girls and boys, respectively. This acceleration of bone mass gain in females, occurring later in males, has been extensively documented in the literature [31, 33, 35, 40, 42] and seems to be associated with pubertal growth, as females enter puberty earlier than males. Because of this increase in bone mass was not steady, with periods of distinct rates of bone mineralization being observed , the correlation of BMD with age was not significant as seen by stepwise multiple regression analysis. Although age is a major temporal indicator for alterations occurring in adolescence, it is limited in relation to the constant modifications occurring in puberty due to maturation level variability in individuals of the same age [43]. It is well known that the rate of bone accretion increases dramatically during puberty and is a function of pubertal stage, rather than chronological age [41]. Bone mineralization parameters were compared with degree of sexual maturity, in an attempt to identify those puberty stages that indicated the greatest increment in bone mass. A significant increase in BMD values according to puberty was observed between all pubertal stages, and occurred earlier in girls than boys with no significant differences between them in all stages except in stage III. Both sexes showed the main increase in BMD between stage IV and V, during these late stages of puberty the deceleration of the growth spurt occurs and adult levels of sex steroids can be attained [44]. similarly to what was previously reported by Sluis et al [5], Boot et al [35], Ahmed et al [35], Hasanoğlu et al [39], Yilmaz et al [40], De Schepper et al [44]. In contrast to the our findings, Gracia-Marco et al [45] showed the most important BMD increase between Tanner stages III and IV in both sexes. This cross-sectional study confirmed the powerful independent effect of puberty on BMD, by stepwise multiple regression analysis, Tanner stage in boys and girls had a significant and independent correlation with BMD and this in agreement with other studies [34, 35, 38, 42]. The timing of bone mass acquisition differs between boys and girls apparently due to the different onset and progression of puberty according to sex steroid production and their response at target tissues [46]. The pattern of bone growth in boys differs from that in girls in 2 ways. First, boys have 2 more years of prepubertal growth because of a later onset of puberty (age 13, rather than 11 as in girls). Second, their pubertal growth

spurt lasts for 4 years rather than the 3 years in girls [47,48]. In the current study, In both sexes bone mass progressively increased during childhood, with a rapid gain during puberty. However, some gender differences in the accrual of bone mass were evident. when gender differences have been compared depending on age groups, There were no significant differences between boys and girls until the age of 10. After the age of 10, females had greater BMD at lumber spine across all age groups, as the results of the earlier onset of the puberty, whereas in the late adolescence (around the age of 17) total bone mass in boys exceeds that measured in girls, but without significant differences. Similarly some studies have reported spine BMD to be higher in girls than in boys [35,39,40,41], until late adolescence; and it has been suggested that ultimately these gender differences during adolescence at the spine disappear as boys catch up with puberty and growth [4]. When gender differences have been compared depending on pubertal stages, males had higher values for BMD in all pubertal stages , but without significant differences between them except in stage III, this indicated that boys gain more BMD than girls at this stage. As mentioned before, in stage I (prepubertal years) there was no significant difference in BMD between males and females and this coincides with the results of Boot et al [35], De Schepper et al [44], This indicates that the development of BMD before puberty is not dependent of sex steroids. In view of these results, one may conclude that the accepted explanation attributing the gender differences in bone density in adolescents to the differences in bone size only is unlikely, and that the mechanisms underlying this effect may possibly be different at cortical and trabecular sites. At the trabecular sites, such as the lumbar spine, gender differences in BMD may be explained by the earlier attainment of puberty in girls, whereas, at the cortical sites, they may be explained by other factors, such as size, muscle mass, and the difference in the level of physical activity [33]. Studies in animals suggested the existence of sex-linked genes mediating the gender difference in BMD [50]. Z scores in the study group were derived through the densitometer software using a American database as reference**,** it has been found that our pediatric and adolescent population have lower BMD values than Americans. For girls, the mean Z scores was -1.2±1.2 and for boys, the mean Z scores was -1.4±1.1. There is established ethnic difference in BMD [41,51]; BMD in Asians is reported to be lower than in other people that are because of their smaller bodies [52]. Little is known, however, about the factors that contribute to racial variations in bone mass or the time of life when such differences become manifest. Some explained ethnic differences, in part, by the differences in lifestyle or in anthropometric measurements [53].

Conclusion

BMD increases with age, with a higher increment during puberty and this increase in bone mineralization during puberty occurs at the same time as significant increases in body dimensions. There was a strong evidence that pubertal development was consistent and independent predictors of BMD (p-value <0.001) in healthy children and adolescents. So densitometry data should be adjusted for pubertal status since both growth and puberty influence bone accretion, and this will be of particular significance in the evaluation of children and adolescence with pubertal

or growth disorders. The increase in levels of serum testosterone and E2 were significant at different pubertal stages in males and females, respectively, until adult levels were achieved at the end of puberty, and this increase was accompanied by an increment in BMD values. So testosterone level was a positive predictor for BMD in males and E2 was a positive predictor in females. anthropometric parameters also had a significant influence on BMD of males only and not for females. There was gender difference in values of BMD by age group, females had significantly greater BMD than males, this due to the early onset of puberty in females. By the age of 17 (the end of the puberty), males presented higher BMD values, although the difference was not significant. while the gender differences in values of BMD by pubertal stages were not significant except in stage III.

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