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BONE MINERAL DENSITY, 25-HYDROXYVITAMIN D LEVELS AND CHRONIC LIVER DISEASE IN PEDIATRIC PATIENTS

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ABSTRACT

Bone health of children and adolescents has been the focus of studies in recent years, since it is necessary to understand the low bone mass and / or hypovitaminosis D in pediatric patients with chronic disease, in order to propose a preventive and therapeutic strategy for promote healthy skeletal growth. This study sought to describe bone mineral density (BMD) and serum vitamin D levels in pediatric patients, followed up at referral outpatient clinics for liver disease. This is a descriptive study with patients aged 5-19 years, seen at outpatients with liver disease. Variables studied: body mass index, sexual maturation stage, serum levels of 25-hydroxyvitamin D and BMD by dual emission X-ray absorptiometry. Hypovitaminosis D values <20 ng / ml and low BMD were considered, Z scores ≤ -2 standard deviations. The patients were divided into two groups: one with chronic liver disease under conservative treatment and the other with transplant patients. The data were analyzed using the tests: Student's t, Mann-Whitney, Pearson's chi-square, Fisher's exact test and logistic regression analysis. P <0.05 were considered significant. Were studied 132 patients, 98 (74.2%) under conservative treatment and 34 (25.8%) transplanted. Comparing the two groups, it was observed that those undergoing clinical treatment were older [12.4 (3.5) vs 10.5 (3.1); p = 0.007], had a greater severity of the disease (27.6% vs 5.9%; p = 0.009) and a higher percentage of patients with low BMD (23.5% vs 2.9%; p = 0.007). In the conservative treatment group, those with low BMD had a higher frequency of hypovitaminosis D when compared to those with normal BMD (56.5% vs 29.7%; p = 0.019), even after adjusting for the severity of the disease (p = 0.038). Patients under conservative treatment had a lower BMD when compared to those undergoing liver transplantation and when analyzed separately, in the first group an association was observed between BMD and serum vitamin D levels.

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INTRODUCTION

In recent years, interest in studying bone health in children and adolescents has grown[1,2]. Identifying low bone mass and / or hypovitaminosis D in pediatric patients with chronic disease can be an important preventive and therapeutic strategy to be adopted to promote healthy skeletal growth[3,4]. The impairment of bone

mineralization is one of the extrahepatic complications known in chronic liver disease (DHC), which can silently, linked to late diagnosis, impair the healthy development of the skeleton, increasing the risk of fracture at a more advanced stage of the disease. Few data are available to identify measurements of bone mineral density (BMD) and vitamin D levels in pediatric patients, especially in the early stage of liver disease[4]. Studies carried out in pediatric liver disease patients in the Brazilian population found a high prevalence of low BMD (61.5%) [5] and hypovitaminosis D (36%) [6]. These findings demonstrate that DHC can compromise the quantity and quality of bone, leading to bone fragility. Dual emission X-ray absorptiometry (DXA) is an appropriate tool to identify children and adolescents with bone deficit and to complement the clinical information necessary for better monitoring of these patients bone health. In addition, the early detection of this deficit allows the adoption of preventive measures against fractures and related morbidities[7,10]. The vitamin D acts on the metabolism of calcium and phosphorus and plays a fundamental role in bone mineralization, particularly in childhood, when there is a greater acquisition of bone mass[11]. In the pediatric population, the main risk groups for hypovitaminosis D are those with chronic diseases, such as liver disease, because in this condition there is impairment in one of the stages that involve the activation of the vitamin D metabolite, in addition to the pharmacological interference of glucocorticoids used in which contribute to low absorption of vitamin D[11,13]. In this sense, vitamin D deficiency, as well as impaired bone mineralization, can have a negative impact on the health, growth and development of pediatric patients with DHC. Thus, the goal of the present study was to describe BMD and serum vitamin D levels in pediatric patients, followed up at referral outpatient clinics for liver disease.

MATERIALS AND METHODS

Study carried out with pediatric patients followed at the outpatient clinics for the treatment of chronic liver disease (DHC) at the Pediatric Gastroenterology and Hepatology Service, at the Professor Edgard Santos University Hospital Complex (C-HUPES), at the Federal University of Bahia (UFBA, Brazil), conducted from February 2016 to March 2017. Patients were divided into two groups: one of patients under conservative treatment and the other of patients who underwent liver transplantation. This project was approved by the Ethics Committee of the HUPES Complex under nº 1.360.091/2015. The parents and/or guardians who agreed to participate, signed the Free and Informed Consent Form and the adolescents signed the Informed Consent Form. The study included patients aged 5 to 19 years, of both sexes, with or without cirrhosis, who met the necessary criteria for performing DXA of the entire body. Children under 5 years of age were not included because they had difficulty keeping in position for the time necessary to perform DXA[7]. The patients who participated in the study had the following diagnoses: biliary atresia, choledochal cyst, sclerosing cholangitis, Alagille syndrome, Budd-Chiari syndrome, autoimmune hepatitis / HAI, viral hepatitis, Alpha1 antitrypsin deficiency, Wilson's disease, Lysosomal Acid Lipase (LAL-D) deficiency, Niemann Pick, Tyrosinemia, Glycogenoses. Patients with kidney disease, primary bone disease, celiac disease, inflammatory bowel disease, cystic fibrosis, short bowel syndrome, patients with ascites and those with vitamin D supplementation were excluded.

Information on age, sex, clinical data (diagnosis of chronic liver disease, liver biopsy, use of medications, supplements) were collected from the patient's medical record. Children and adolescents were weighed in light clothing and with bare feet on an electronic platform scale (Filizola® São Paulo, Brazil). Height was measured standing, with a vertical stadiometer attached to the platform scale, according to the Guidance Manual of the Brazilian Society of Pediatrics (SBP)[14]. To assess the anthropometric status, the body mass index (BMI / I) was calculated, considering the classification recommended by Sisvan (Technical Standard of the Food and Nutritional Surveillance System-2011)[15], adapted from the World Health Organization (WHO-2007 -)[16], considering: thinness (z score <-2); eutrophy (z score ≥ -2 and ≤ +1), overweight (z score ≥ +1 and <+2); obesity (z score ≥ +2). Adolescents were classified as prepubertal (male: sexual maturation stages 1 and 2; female: stage 1), pubescent (male: stage 3; female: stages 2 and 3) and post-pubertal (stages 4 and 5 in both sexes), according to the sexual maturation stage proposed by Tanner[17] and this evaluation was performed by pediatricians.

The bone mineral density (BMD) of the whole body, except the head (TBLH), was determined by DXA (GE Lunar Prodigy DPX-NT, GE software) and performed in a specialized clinic, by a qualified professional. The position adopted for the examination were those recommended by the International Society for Clinical Densitometry (ISCD)[7]. BMD data were presented in Z scores, using the database provided by the manufacturer, according to sex, age and adjusted for patients' weight and height. Low BMD was determined when the Z score was ≤ -2 standard deviations (SD)[10]. The dosage of 25 (OH) D was performed using the chemiluminescence method, adopting the cutoff points suggested by the American Academy of Pediatrics (AAP) [12] and by the European Society of Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN)[13], who recommend: sufficiency ≥ 30 to 100 ng / ml; insufficiency between 20.0 to 29.9 ng / ml and deficiency <20.0 ng / ml. In the present study, hypovitaminosis D was considered when the values of 25 (OH) D were deficient[11,13]. For the analysis and tabulation of the data, the statistical package SPSS, version 22 (Statistical Package for Social Science) was used. The variables were described by absolute and relative frequency, mean and standard deviation (SD) or median and interquartile range (IIQ). Student's t-tests, Mann-Whitney and Pearson's chi-square tests or Fisher's exact test were used to compare the groups. After a bivariate analysis of the groups with and without low BMD performed on patients under conservative treatment, variables with a p-value <0.20 (low vitamin D and disease severity) were adjusted in a logistic regression model. Values of p <0.05 were considered significant.

RESULTS

Were studied 132 patients from pediatric chronic liver disease outpatients were studied. Of these, 98 (74.2%) were under conservative treatment and 34 (25.8%) had undergone liver transplantation. The most frequent etiology of DHC was biliary atresia 26.5%, followed by autoimmune hepatitis 22.7%. The duration of transplantation varied from one to 14 years (median = 8; IIQ = 5 - 11 years). Table 1 shows the clinical and sociodemographic characteristics of these patients.

Table 1. Demographic, anthropometric and clinical characteristics of patients followed up at the pediatric outpatient clinic for chronic liver diseases. Salvador, Bahia, Brazil, 2016 – 2017

Characteristics	Transplanted patients		p
	Yes	No	
	34 (25.8%)	98 (74.2%)	
Age (in years) *	10.5 (3.1)	12.4 (3.5)	0.007
Women	20 (58,8%)	59 (60.2%)	0.887
Sexual Maturation Stage			0.093
Prepubertal	22 (64,7%)	46 (46.9%)	
Pubescent	4 (11,8%)	29 (29.6%)	
Postpubertal	8 (23,5%)	23 (23.5%)	
Anthropometric State			0.310
Thinness / Eutrophic	27 (79,4%)	69 (70.4%)	
Overweight / obesity	7 (20,6%)	29 (29.6%)	
Severity of the disease (MELD/PELD)			0.009
Bigger severity	2 (5.9%)	27 (27.6%)	
Less severity	32 (94.1%)	71 (72.4%)	
Use of corticosteroids	10 (29.4%)	-	<0.001
Serum albumin g/dL*	4.1 (0.4)	4.1 (0.6)	0.744
RNI*	1.1 (0.1)	1.2 (0.2)	0.043
ALT U/L†	22.6 (20.0 – 35.7)	39.5 (22.0 – 86.5)	0.014
AST U/L†	31.5 (24.2 – 41.5)	44.0 (27.8 – 85.0)	0.009
Alkaline phosphatase U/L†	275,5 (196,0 – 397,0)	239.5 (149.0 – 369.8)	0.232
GGT U/L†	25.5 (19.8 – 78.0)	38.5 (23.8 – 137.5)	0.070
Total bilirubin mg/dL*	0.5 (0.4 – 0.6)	0.7 (0.5 – 1.1)	<0.001
Indirect bilirubin mg/dL *	0.2 (0.1 – 0.3)	0.3 (0.2 – 0.5)	0.004
Hematocrit %*	38.3 (5.8)	36.9 (4.2)	0.139
Hemoglobin g/dL*	13.2 (1.3)	12.3 (1.5)	0.003
Urea mg/dL*	22.4 (6.2)	23.8 (8.8)	0.397
Creatinine mg/dL *	0.6 (0.2)	0.6 (0.2)	0.330
Low 25- hydroxyvitamin D	15 (44.1%)	35 (36,1%)	0.407
Bone mineral density			0.007
Normal	33 (97.1%)	75 (76.5%)	
Low	1 (2.9%)	23 (23.5%)	

* Average (standard deviation); † Median (Interquartile range).

Table 2. Comparison of patients under conservative treatment according to bone mineral density. Salvador, Bahia, Brazil, 2016 – 2017

Characteristics		Bone mineral density		Bivariate analysis (p)	
		Low	Normal		
		23 (23.5%)	75 (76.5%)		
		12.6 (4,.)	12.3 (3.4)	0.747	
				0.368	
		12 (52.2%)	47 (62.7%)		
		11 (47.8%)	28 (37.3%)		
				0.346	
		18 (78.3%)	51 (68.0%)		
		5 (21.7%)	24 (32.0%)		
Sexual Maturation Stage			0.687		
	11 (47.8%)	35 (46.7%)			
	8 (34.8%)	21 (28.0%)			
	4 (17.4%)	19 (25.3%)			
		13 (56.5%)	22 (29.7%)	0.019	0.038
		8 (34.8%)	25 (33.3%)	0.898	
				0.155	0.256
		9 (39.1%)	18 (24.0%)		
		14 (60.9%)	57 (76.0%)		

*Average (standard deviation). † The regression model included the variables "low vitamin D" and disease severity ".

Among patients under conservative treatment, 23 (23.5%) had low BMD while only one (2.9%) in the group of transplant recipients had this condition. This was a male, 15 years old, postpubertal, eutrophic, submitted to liver transplantation four years ago due to Budd-Chiari Syndrome, MELD score = 11 (greater severity), normal serum calcium and vitamin D levels, did not use corticosteroids, vitamins or minerals and had a BMD z score of -2.3. Table 2 compares the profile of patients undergoing conservative treatment according to BMD.

DISCUSSION

This study showed a higher frequency of low BMD in patients with DHC undergoing conservative treatment when compared to those who underwent liver transplantation.

Regarding hypovitaminosis D, there was no statistically significant difference between the groups evaluated. However, in the conservative treatment group, patients with low BMD had a higher percentage of hypovitaminosis D. Bone mineralization impairment is one of the known complications of CHD, especially in adult patients in advanced stages of the disease.18 Although the available data on pediatric patients are scarce and controversial,[5,19] the evidence accumulated over the past few years, shows high frequency of deficient bone mineralization in children and adolescents with chronic liver disease[2,20,21]. A Brazilian study carried out by Taveira et al.,[5] in the city of Ribeirão Preto, with 35 children and adolescents (13 with chronic cholestatic liver disease and 22 controls), found reduced bone mineral density in 61.5% of the patients, however the authors did not find 25 (OH) D deficiency, but

significant drop in serum levels of insulin-like growth factor (IGF -1). Although the patients evaluated in the study by Taveira et al.[5] were chronic liver disease patients, this sample consisted only of patients with cholestatic disease, whereas in the present study, patients with various etiologies were evaluated, which may justify the higher frequency of low BMD described by the authors. compared to our results. The literature demonstrates that impaired bone mineralization in patients with CHD results in a combination of different mechanisms depending on the underlying disease.[19] A study by Mora et al.,[22] conducted in the city of Milan, Italy, involving 32 pediatric patients with chronic hepatitis B or hepatitis C infection, did not report low BMD in patients with liver disease, disagreeing with the findings of the present study. It is believed that the difference in results can probably be explained by the severity of the disease presented by patients. It is noteworthy that in the study by Mora et al.,[22] patients did not have end-stage liver disease, unlike patients in this study. From the pathophysiological point of view, hepatic impairment would affect BMD due to decreased IGF-1 production and vitamin D deficiency, however, not all studies[5,23] show an association between vitamin D and bone BMD. It is known that IGF-1 is an important stimulator of osteoblastic activity, that although it is synthesized in several tissues, most of it circulates from the liver. In this sense, some studies that indicate IGF-1 stand out as an anabolic polypeptide important in the stimulation of bone mass[5,24].

The vitamin D, in turn, also plays a fundamental role in the formation of bone mass[11], however, in chronic liver diseases there is impairment in one of the stages that involve the activation of the vitamin D metabolite that leads to impaired bone mineralization[11,12,13]. Another factor that can also compromise bone mass gain and accelerate bone loss in patients with DHF, is the deficit in nutritional status, since chronic liver diseases can induce changes in the intermediate metabolism of carbohydrates, lipids, proteins, vitamins and minerals, related to the degree of hepatic impairment.[5,19] In the present study, most patients with low BMD had anthropometric eutrophic status. It is worth mentioning that the anthropometric status was assessed by BMI / I and that this index is not a good parameter for assessing nutritional status, since it evaluates the total body mass, without considering the distribution of body compartments, and thus may not reflect the real nutritional status of patients. Thus, it is recommended to use nutritional assessment methods that best reflect the markers of bone formation and reabsorption, such as serum calcium, inorganic phosphorus and serum magnesium and the assessment of food consumption, in order to better estimate the bone health of patients with liver disease.[5,21,25,26] It was found that patients undergoing liver transplantation showed better results for bone mineralization (BMD) than those undergoing clinical treatment. Although this study has not followed the evolution of BMD before and after liver transplantation, this finding suggests that this form of treatment for DHC may be effective in controlling BMD, allowing healthy skeletal growth in these patients[27]. In a cohort study carried out in Germany with 86 patients between 1981 and 1988, the authors demonstrated that patients undergoing liver transplantation had an 88% survival in five years with monitoring of the evolution of BMD after liver transplantation[28].

Among the limitations of this study, we mention: the non-assessment of food intake of macro and micronutrients, as well as calcium, phosphorus and magnesium dosages. In addition, despite the use of DXA, the most commonly recommended method for assessing BMD in children and adolescents, there are several peculiarities in the assessment of bone mass in this population that need to be considered. Among the factors related to DXA, mention is made of the absence of reference data, especially for pediatric patients with chronic diseases; the lack of significant clinical results, concerning densitometric measurements and changes in body size and composition, regarding growth. It is noteworthy, although the density obtained by DXA is sandy (BMD) and non-volumetric (BMC) and as the area does not increase in the same proportion as the volume during growth, pediatric patients with large bones may overestimate BMD, while those with small bones can underestimate it.[4,8,29,30] It is worth mentioning that the definition of bone health is very comprehensive, not only including the amount of bone mineral mass (BMD) and levels of 25 (OH) D, but also other intrinsic factors (genetics, hormones, puberty, chronic inflammatory morbid states) and extrinsic (adequate nutrition in calcium, protein, physical activity, medication use, among others).[4,29] However, our results call attention to low BMD and hypovitaminosis D as conditions present in pediatric patients with DHC and suggest that liver transplantation may improve this condition. Thus, it is recommended that pediatric liver disease patients be evaluated and monitored with regard to bone mass and serum vitamin D levels, since it is in childhood and early adulthood that the bone mass pool of the individual and case is formed if the adequate amount of bone mass is not reached at this age, this may lead to future problems such as osteopenia, osteoporosis and bone fractures.

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