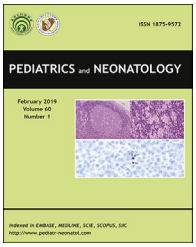
Pediatrics and Neonatology (2019) 60, 12–18

Available online at www.sciencedirect.com

ScienceDirect

journal homepage: http://www.pediatr-neonatol.com



Original Article

Vitamin D non-sufficiency is prevalent in children with chronic liver disease in a tropical country

Way Seah Lee [a,b,*], Muhammad Yazid Jalaludin [a,b], Shin Yee Wong [a], Sik Yong Ong [a], Hee Wei Foo [a], Ruey Terng Ng [a]

[a] Department of Paediatrics, University Malaya Medical Centre, 59100, Kuala Lumpur, Malaysia
[b] Paediatrics and Child Health Research Group, University Malaya, 50603, Kuala Lumpur, Malaysia

Received May 29, 2017; received in revised form Sep 9, 2017; accepted Mar 31, 2018
Available online 5 April 2018

Key Words
chronic liver disease;
vitamin D deficiency;
tropical country

Background: To determine vitamin D status in children with chronic liver disease (CLD) in a tropical country.
Methods: Cross-sectional study in Malaysian children with CLD. Factors affecting serum vitamin D level (definition: deficient $<$ 30 nmol/L; insufficient 30–50 nmol/L; sufficient $\geq$ 50 nmol/L) was analyzed.
Results: Of the 59 children studied (males 32, 54%; median age 6.8 $\pm$ 5.3 years), the three most common causes were biliary atresia (n = 25), autoimmune hepatitis (n = 16) and sclerosing cholangitis (n = 6). The overall mean daily vitamin D intake was 715 $\pm$ 562 units/day. Thirteen (22%) patients had at least one clinical signs of rickets. Seventeen (29%) had serum bilirubin level $\geq$ 34 μmol/L. Eight (14%) children were deficient in vitamin D, eight (14%) were vitamin D-insufficient and 43 (73%) were sufficient. As compared with children with serum bilirubin $<$34 μmol/L, those with serum bilirubin $\geq$34 μmol/L were more likely to have rickets (24% vs. 65%; P $<$ 0.002) and a lower serum vitamin D level (86.0 $\pm$ 54.9 nmol/L vs. 65.4 $\pm$ 48.2 nmol/L; P = 0.05) despite being given a significantly higher vitamin D dose (608 $\pm$ 571 vs. 970 $\pm$ 543 units/day; P = 0.008). The proportion of children with either deficient or insufficient vitamin D status was significantly higher in children with bilirubin level $\geq$34 μmol/L than in children $<$34 μmol/L (47% vs. 19%; P = 0.028).
Conclusion: Vitamin D deficiency and insufficiency is common in children with CLD in a tropical country. Regular monitoring of vitamin D status and screening for metabolic bone disease in all children with CLD is recommended. Higher dose of oral supplement or parenteral route should be considered, especially in those with bilirubin $\geq$34 μmol/L.

* Corresponding author. Department of Paediatrics, Faculty of Medicine, University Malaya, 50603, Kuala Lumpur, Malaysia. Fax: +60 3 7949 4704.
E-mail address: leews@um.edu.my (W.S. Lee).

https://doi.org/10.1016/j.pedneo.2018.03.011

1. Introduction

Vitamin D is crucial for skeletal development. Vitamin D deficiency can lead to poor linear growth, motor delay, bone fragility and hypocalcemic rickets.[1,2] The liver plays an important role in the metabolism of vitamin D.[3,4] Vitamin D from the skin and diet is hydroxylated in the liver into 25-hydroxyvitamin D [25(OH)D].[3] 25(OH)D is transported to the kidney where it undergoes a second hydroxylation and is then converted into 1,25(OH)D, the active form of vitamin D. 25(OH)D is the major circulating form of vitamin D and is widely used to determine the vitamin status of an individual.[3]

Diseases of the liver may interfere with the production of the active metabolites of vitamin D resulting in abnormal calcium and bone metabolism.[3,4] Metabolic bone disease associated with chronic liver disease (CLD) is also called hepatic osteodystrophy.[5] In children, hepatic osteodystrophy affects both the existing bone mineral and growth plates.[5,6] As a result, children with CLD are prone to have not only low bone mass, fractures and short stature, but also rickets and spine abnormalities.[5,6]

Vitamin D deficiency is the major cause of hepatic osteodystrophy. In CLD, vitamin D deficiency is multifactorial, which includes malabsorption of fat-soluble vitamins from deficient intestinal bile acids, malnutrition, and a lack of sunlight exposure.[7] In end-stage liver failure, poor 25-hydroxylation is another contributing factor.[8]

Non-sufficient level of vitamin D (usually defined as serum 25[OH]D concentrations $<$ 30 nmol/L) is prevalent in the general population the world over,[9,10] including in tropical countries with abundant sunlight exposure such as Malaysia.[11–13] In the adult population, the prevalence of vitamin D deficiency in CLD has been reported to range from 64 to 92%.[14–16]

Little is known about the prevalence of vitamin D deficiency and metabolic bone disease in children with CLD from tropical setting with abundant sunlight. The present study aimed to address the gap in the knowledge in this area.

2. Methods

This was a cross-sectional study in children with CLD attending the Children's Liver Clinic of Department of Paediatrics, University Malaya Medical Center (UMMC), Kuala Lumpur, Malaysia. The present study was approved by the institutional review board of UMMC (MEC 1013.99).

2.1. Patients' recruitment

Consecutive patients with CLD attending Children's Liver Clinic of UMMC, from July 2014 to July 2016, were recruited. The diagnosis of CLD was based on pre-defined criteria. Informed consent was obtained from the parents of participating patients.

2.2. Exclusion criteria

The following patients were excluded from the study: (a) concurrent diagnosis of chronic renal failure or parathyroid disease, (b) previous parathyroid surgery, (c) concurrent anti-convulsant therapy metabolized through cytochrome P450 activity.

2.3. Data collection

Age, gender, underlying diagnosis, disease nature and status, and vitamin D supplement regimen were obtained. All patients had a detailed history and physical examination. History obtained included symptoms of vitamin D deficiency and previous history of bone fractures. Physical signs of vitamin D deficiency and rickets were screened systematically. Metabolic bone disease was defined as presence of fractures or osteoporosis by x-ray criteria. Growth retardation was defined as height-for-age z-score < -2 SD, according to World Health Organization (WHO) growth charts appropriate for age and sex.

The following laboratory investigations were performed: full blood count, renal function test, liver function test, C-reactive protein and erythrocyte sedimentation rate (ESR), serum calcium, phosphate, vitamin D and serum intact parathyroid hormone (iPTH).

2.4. Classification of disease status

All cases were classified according to the latest serum bilirubin level into either $<$34 μmol/L or $\geq$34 μmol/L. CLD was also classified as either stable or progressive disease based on a combination of changes in clinical status, liver functions, Child-Pugh scores,[17] and repeated histopathological examination, where applicable. Patients were considered as having a stable CLD if two consecutive Child-Pugh Score assessed six months apart remained as class A (score or 5–6), stable liver synthetic functions, and no significant clinical concerns upon repeated review. Patients were considered as having a progressive disease if there was the presence of any of the following criteria: (a) clinical deterioration, (b) worsening Child-Pugh scores, (c) deteriorating liver synthetic function, or (d) escalating medical therapy.

2.5. Definition of vitamin D status

Vitamin D status was defined according to the definition by the Institute of Medicine and European Society of Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) in this study: deficient (serum 25[OH]D level $\leq$ 30 nmol/L), insufficient (25[OH]D level between 30 and 50 nmol/L), and sufficient (25[OH]D level $>$ 50 nmol/L).[18,19]

2.6. Vitamin D supplementation

In the current study, vitamin D supplements were given in the form of cholecalciferol, calcitriol or multivitamin supplements containing vitamin D. The total daily vitamin D intake was estimated from the total amounts of vitamin D and vitamin D-containing multivitamins. For the purpose of the present study, the total daily dose of vitamin D supplement was divided into: (a) $<$ 1000 units/day; (b) $\geq$ 1000 unit/day.[18] Generally, it was the policy of the unit to provide higher dose of vitamin D supplements ($\geq$1000 of

vitamin D) in older children, children with impaired excretory hepatic function, or with decompensated liver status.

2.7. Biochemical analysis

Vitamin D and iPTH levels were measured using an automated assay (Elecsys Vitamin D and PTH electrochemilunescence technology, Roche Diagnostics, Mannheim, Germany). Serum vitamin D levels and iPTH were reported in nmol/L and pmol/L, respectively. The reference range (2.5th and 97.5th) for iPTH level in healthy children was 0.10–4.15 pmol/L (1.0–39.1 ng/L).[20] In the clinical laboratory of UMMC, an iPTH level of ≥6.0 pmol/L was diagnostic of hyperparathyroidism.

2.8. Statistical analysis

Data were managed with SPSS 22.0. Descriptive statistics were used for demographic status, serum levels of vitamin D and iPTH. Independent t-test and Mann–Whitney test were used for analysis of continuous variables. Chi-square and Fisher's exact tests were used for categorical data. A P-value of less than 0.05 was considered as statically significant.

3. Results

During the study period, 59 consecutive patients with CLD attending the Children's Liver Clinic of UMMC who fulfilled the study criteria were recruited (Table 1). There were 32 (54%) males. The median (±S.D.) age was 6.8 (±5.3) years. Five (8.5%) patients were infants aged younger than one year while 60% (n = 34) were aged five years or older.

Table 1 Demographic and clinical profiles of 59 children with chronic liver disease.

	n	%
Gender		
Male	32	54
Female	27	46
Age (years ±S.D.)	6.8 ± 5.3	
Age group		
< 1 year	5	8.5
1–4 year	20	34
5–9 year	15	25
10–14 year	14	24
15–18 year	5	8.5
Underlying cause of liver disease		
Biliary atresia	25	42
Autoimmune hepatitis	16	27
Primary sclerosing cholangitis	6	10
Glycogen storage disease	4	7
Progressive familial intrahepatic cholestasis	3	5.1
Alagille's syndrome	2	3.3
Cryptogenic liver cirrhosis	2	3.3
Congenital hepatic fibrosis	1	2
Status of disease		
Inactive/Stable	34	58
Active/Progressive	25	42

3.1. Causes of CLD

The three most common causes of CLD were biliary atresia after Kasai's surgery (n = 25, 42%), autoimmune hepatitis (n = 16, 27%), and primary sclerosing cholangitis (10%; Table 1).

3.2. Rickets and metabolic bone disease

Overall, 22% of all children had at least one physical signs of rickets (Table 2). Except for rachitic rosary (n = 6), other signs of rickets were uncommon (Table 2). Short stature was seen in 29% (n = 17). A 3-year-old child with PFIC had a history of pathological bone fractures. Thus, metabolic bone disease was present in 2% of children. As compared to children with serum bilirubin level <34 μmol/L, children with serum bilirubin level ≥34 μmol/L were more likely to show at least one clinical signs of rickets (24% vs. 65%; P = 0.002).

3.3. Liver biochemistry and laboratory bone profiles

Liver biochemistry and laboratory bone profiles are shown in Table 2. With the exception of serum aspartate aminotransferase (AST) level, there were no significant differences between children with serum bilirubin level below or above 34 μmol/L in terms of serum calcium and phosphate, and liver enzymes. The serum AST level in children with serum bilirubin level <34 μmol/L was significantly lower than children with serum bilirubin level ≥34 μmol/L (mean ± S.D. = 78 ± 60 IU/L vs. 167 ± 148 IU/L, P = 0.015).

There was no statistical significant difference in the iPTH level between children with serum bilirubin level below or above 34 μmol/L. A total of five children (8.5%) had an iPTH level of ≥6.0 pmol/L.

3.4. Vitamin D supplementation

The total daily vitamin D supplementation is shown in Table 2. Nineteen (32%) children did not have any vitamin D or multivitamin supplements. Children with serum bilirubin level ≥34 μmol/L were given a significantly higher daily vitamin D doses than children with serum bilirubin level <34 μmol/L (mean ± S.D.; 970 ± 543 units/day vs. 608 ± 571 units/day; P = 0.008).

3.5. Serum vitamin D level

The serum vitamin D level for children with serum bilirubin level <34 μmol/L CLD was significantly higher than those with serum bilirubin level ≥34 μmol/L (mean ± S.D. = 86.0 ± 54.9 nmol/L vs. 65.4 ± 48.2 nmol/L; P = 0.05; Table 2). Similarly, children who were given daily vitamin D dose ≥1000 units/day had a significantly higher

Table 2 Clinical signs of vitamin D deficiency and bone profiles in 59 children with chronic liver disease.

	Serum bilirubin <34 μmol/L (n = 42)	Serum bilirubin 34 ≥ μmol/L (n = 17)	All (n = 59)	P-value
Any clinical indicator of rickets	10 (24%)	11 (65%)	21 (22%)	0.002
Bone tenderness	0	2	2	
Muscle weakness	0	0	0	
Skeletal deformity	0	1	1	
Rachitic rosary	4	2	6	
Double maleoli sign	0	1	1	
Widening of wrists	2	1	3	
Metabolic bone disease (history of fractures)	1	0	1	
Short statute (height-for-age < 3rd centile)	9	8	17 (29%)	
Liver biochemistry and bone profiles				
Total bilirubin (mean ± S.D.; μmol/L)	12 ± 8.4	257 ± 228	83 ± 164	<0.001
AST (mean ± S.D.; IU/L)	78 ± 60	167 ± 148	104 ± 101	0.015
ALT (mean ± S.D.; IU/L)	75 ± 65	118 ± 85	87 ± 73	0.716
Alkaline phosphatase (mean ± S.D.; IU/L)	355 ± 194	548 ± 236	411 ± 223	0.127
γGT (mean ± S.D.; IU/L)	157 ± 209	136 ± 145	151 ± 192	0.207
Serum calcium (mean ± S.D.; mmol/L)	2.41 ± 0.16	2.32 ± 0.16	2.38 ± 0.16	0.453
Serum phosphate (mean ± S.D.; mmol/L)	1.50 ± 0.31	1.35 ± 0.23	1.46 ± 0.29	0.021
Serum iPTH level (mean ± S.D.; pmol/L)	3.59 ± 2.97	3.65 ± 3.57	3.60 ± 3.12	0.182
Serum vitamin D level (mean ± S.D.; nmol/L)	86.0 ± 54.9	65.4 ± 48.2	80.1 ± 53.5	0.05
No. of patient with iPTH level ≥6.0 pmol/L	3	2	5	
Daily total vitamin D supplementation				
0–1000 units/day	28	8	36	
≥1000 units/day	14	9	23	
Mean (±S.D.; units/day)	608 ± 571	970 ± 543	715 ± 562	0.008
Serum vitamin D level				
< 30 nmol/L	3	5	8 (14%)	
30–50 nmol/L	5	3	8 (14%)	
> 50 nmol/L	34	9	43 (73%)	

Note: AST – aspartate aminotransferase; ALT – alanine aminotransferase; γGT – gamma-glutamyl transpeptidase; iPTH – intact parathyroid hormone.

serum vitamin D level than those given <1000 units/day (mean ± S.D. = 99.0 ± 77.1 nmol/L vs. 67.9 ± 34.2 nmol/L; P = 0.017). However, there was no significant difference in the serum vitamin D between children with stable disease and progressive disease (P = 0.89; Table 3).

3.6. Vitamin D status

The overall prevalence of vitamin D deficiency (serum vitamin D level ≤ 30 nmol/L) was 14% (n = 8). Another 14% (n = 8) had an insufficient serum level of vitamin D (30–50 nmol/L).

3.7. Factors affecting vitamin D status (Table 3)

There was no significant difference between the prevalence of vitamin D sufficiency and gender, age, progression of liver disease (progressive vs. stable), and daily vitamin D intake (<1000 units/day vs. ≥ 1000 units/day). However, children with a higher level of serum bilirubin level (≥34 μmol/L) were more likely to have a serum vitamin D level ≤ 50 nmol/L (either deficient or insufficient) as compared to children with serum bilirubin level < 34 μmol/L (47% vs. 19%; P = 0.028).

4. Discussion

Vitamin D deficiency is a common problem worldwide, including a tropical country like Malaysia where sunshine is abundant throughout the year. Chin et al. found that while the prevalence of vitamin D deficiency (defined as serum 25 [OH]D level < 30 nmol/L) was 0.5%, the prevalence of insufficiency (30–50 nmol/L) was 22.7%.[21] However, Al-Sadat et al. observed that the prevalence of vitamin D deficiency (defined as vitamin V < 37.5 nmol/L) in Malaysian adolescents was as high as 78.9%.[13]

The present study shows that vitamin D deficiency was prevalent in children with CLD, despite vitamin D supplementation. Overall, 14% of the patients had a deficient serum vitamin D level (<30 nmol/L) while another 14% were insufficient in serum vitamin D (30–50 nmol/L). Thus, a total of 28% were either deficient or insufficient. In addition, more than one in five children (22%) with CLD had at least one physical sign of vitamin D deficiency. Biochemical evidence of hyperparathyroidism, characterized by an elevated level of iPTH, was seen in 8.5% of the patients.

It is difficult to compare the findings of the present study with those reported in the literature, mainly because of differences in the definition of vitamin status as well as the

Table 3 Factors affecting serum vitamin D level in 59 children with chronic liver disease.

	Mean (±S.D.) Serum Vitamin D Level (nmol/L)	P-value [a]	Vitamin D Sufficient (>50 nmol/L) n = 43 (%)	Vitamin D Deficient (<30 nmol/L) and Insufficient (30–50 nmol/L) n = 16 (%)	P-value [b]
Gender					
Male (n = 32)	80.1 ± 41.1	0.41	23	9	0.85
Female (n = 27)	80.0 ± 65.9		20	7	
Age group					
<1 year	91.8 ± 54.8		3	2	0.82
1–4 year	77.8 ± 72.4		14	6	
5–9 year	92.7 ± 48.		12	3	
10–14 year	67.9 ± 26.4		11	3	
15–18 year	73.6 ± 40.9		3	2	
Serum bilirubin level					
<34 μmol/L (n = 42)	86.0 ± 54.9	0.05	34 (81%)	8 (19%)	0.028
≥34 μmol/L (n = 17)	65.4 ± 48.2		9 (53%)	8 (47%)	
Progression of disease					
Progressive disease (n = 34)	65.7 ± 43.7	0.84	28 (82%)	6 (18%)	0.056
Stable disease (n = 25)	90.6 ± 58.0		15 (60%)	10 (40%)	
Daily vitamin D supplementation					
<1000 units/day	67.9 ± 34.3	0.017	25 (69%)	11 (31%)	0.55
≥1000 units/day	99.0 ± 71.1		18 (78%)	5 (22%)	

Note.
[a] Student t-test.
[b] Chi-square test between vitamin D sufficiency and non-sufficiency.

underlying cause of chronic liver disease.[16,22–25] Arteh et al., who defined vitamin D deficiency as serum 25OH-D < 32 ng/ml (~80 nmol/L), found that the prevalence of vitamin D deficiency was as high as 92.4% in 118 adult patients (85% who had chronic hepatitis C) with chronic liver disease.[16] Of these, 30% were severely deficient (<30 nmol/L).[16]

In a pediatric study involving 102 children with non-alcoholic liver disease, Hourigan et al. (insufficient < 29 ng/ml, ~72 nmol/L) found that the prevalence of vitamin insufficiency was 78%.[25] Shneider et al. studied 92 infants with biliary atresia and found that biochemical evidence of vitamin D deficiency was present in 21–37%.[22] Shen et al. studied 23 children with various cholestatic disorders who received fat-soluble vitamin supplementations.[23] The authors found that 82% of children were deficient in vitamin D.[23] The prevalence was 100% in children with serum bilirubin level ≥ 3.0 mg (51 μmol/L).[23]

The present study confirms the prevalent nature of vitamin D deficiency in children with CLD in a tropical setting with plenty of sunlight throughout the year, irrespective of gender, age group or whether the disease was progressive or stable in nature.

We observed that children with a higher serum bilirubin level (≥34 μmol/L) were more likely to have either deficient (<30 nmol/L) or insufficient (30–50 nmol/L) vitamin D level despite being prescribed a much higher dose of vitamin D supplement. The most likely explanation for this observation was a higher serum bilirubin level was indicative of impaired hepatic excretory function. Impaired excretion of bile acid into the intestine may impair the absorption of vitamin D.[6]

The definition of vitamin D deficiency used for the current study followed the recommendation by the Institute of Medicine, National Institutes of Health and the European Society of Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN),[18,19] which considers a serum 25(OH)D level < 30 nmol/L (10 ng/mL) as deficient and a level of 30–50 nmol/L as insufficient.[18,19]

In the present study, signs of rickets were screened for systematically. Rickets happen during the period of rapid growth, usually before 3 years old and during puberty. Signs of rickets are not usually seen during other time periods unless there is severely depleted bone mass. However, while the signs of rickets were common in the present study, pathological fracture was seen only in one patient: a 3-year old child with PIFC (2%; 1/59). The prevalence of fractures in children with end-stage liver disease was reported to be as high as 38% in some studies.[26,27] Most of the fractures were non-vertebral and half were asymptomatic, with puberty a particularly vulnerable phase.[5] Close surveillance for fractures, especially in children with end-stage liver disease, is therefore recommended.[5]

At present, several guidelines are available recommending prevention of vitamin D deficiency in various at risk groups. ESPGHAN currently recommends the daily intake of at least 400 IU/day for healthy European children.[19] Similarly, the Pediatric Endocrine Society also recommends a daily supplement of 400 IU/day, starting in early infancy, especially in infants who are predominantly breastfed.[28]

For children who are vitamin D deficient, the Pediatric Endocrine Society recommends an initial daily dose of

1000—10,000 IU for eight to twelve weeks, followed by a daily maintenance dose of 400—1000 IU.[28]

The ideal dose of vitamin D and minerals in children with CLD to prevent and treat hepatic osteodystrophy is not well defined.[5,6] The present study shows that even at an average daily intake of 715 IU/day, many children with CLD still has a deficient level of serum vitamin D. Thus, a dose finding study in children with CLD has been recommended.[5] In children with severe jaundice with impaired bile acid secretion into the intestine, an alternative route of administering vitamin D, i.e. parenteral route should also be considered since oral supplement may not be effective to maintain an adequate serum vitamin D level.[6,29]

In addition to addressing the ideal dose and route of vitamin D supplement together with mineral in children with CLD to prevent metabolic bone disease, future research should adopt a uniform definition of vitamin D status, identifying risk factors for significant deficiency as well as long term outcome of those who were deficient during early life.[6] A detailed study on the incidence of fractures, especially vertebral fractures, is also necessary.

One of the most important weakness of the present study was the duration of exposure to sun was not assessed. Thus, we were not able to correlate the duration of sunlight exposure with serum vitamin D status. Other studies conducted in tropical countries among healthy children with abundant sunlight year round have shown a high prevalence of vitamin D deficiency in healthy children,[11,12] including children in Malaysia.[13,30] In addition, dietary intake of vitamin D was also not available.

In conclusion, the current study shows that vitamin D deficiency is a common problem in children with CLD despite use of vitamin D supplements. We recommend close monitoring of serum vitamin D level in all children with CLD. Children who had a higher serum bilirubin level were more likely to be vitamin D-deficient and may benefit from a much higher vitamin D supplement or parenteral route.

Conflict of interest

None of the authors has any potential conflict of interest to declare.

Acknowledgement

The authors gratefully acknowledge the willingness of children with chronic liver disease to take part in the present research. The present study was partly funded by a research grant from University Malaya High Impact Research (UM.C/625/HIR/MOHE/CHAN/13/1).

References

1. Holick MF. Vitamin D deficiency. *N Engl J Med* 2007;**357**:266—81.
2. Holick MF. Resurrection of vitamin D deficiency and rickets. *J Clin Invest* 2006;**116**:2062—72.
3. DeLuca HF. Overview of general physiologic features and functions of vitamin D. *Am J Clin Nutr* 2004;**80**:1689S—96S.
4. Imawari M, Akanuma Y, Itakura H, Muto Y, Kosaka K, Goodman DS. The effects of diseases of the liver on serum 25-hydroxyvitamin D and on the serum binding protein for vitamin D and its metabolites. *J Lab Clin Med* 1979;**93**:171—80.
5. Högler W, Baumann U, Kelly D. Endocrine and bone metabolic complications in chronic liver disease and after liver transplantation in children. *J Pediatr Gastroenterol Nutr* 2012;**54**:313—21.
6. Högler W, Baumann U, Kelly D. Growth and bone health in chronic liver disease and following liver transplantation in children. *Pediatr Endocrinol Rev* 2010;**7**:266—74.
7. Crawford BA, Kam C, Donaghy AJ, McCaughan GW. The heterogeneity of bone disease in cirrhosis: a multivariate analysis. *Osteoporos Int* 2003;**14**:987—94.
8. Stokes CS, Volmer DA, Grünhage F, Lammert F. Vitamin D in chronic liver disease. *Liver Int* 2013;**33**:338—52.
9. Thacher TD, Fischer PR, Strand MA, Pettifor JM. Nutritional rickets around the world: causes and future directions. *Ann Trop Paediatr* 2006;**26**:1—16.
10. Fraser DR. Vitamin D-deficiency in Asia. *J Steroid Biochem Mol Biol* 2004;**89-90**:491—5.
11. Bener A, Al-Ali M, Hoffmann GF. Vitamin D deficiency in healthy children in a sunny country: associated factors. *Int J Food Sci Nutr* 2009;**60**:60—70.
12. Munns CF, Simm PJ, Rodda CP, Garnett SP, Zacharin MR, Ward LM, et al. Incidence of vitamin D deficiency rickets among Australian children: an Australian paediatric surveillance unit study. *Med J Aust* 2012;**196**:466—8.
13. Al-Sadat N, Majid HA, Sim PY, Su TT, Dahlui M, Abu Bakar MF, et al. Vitamin D deficiency in Malaysian adolescents aged 13 years: findings from the Malaysian health and adolescents longitudinal research team study (MyHeARTs). *BMJ Open* 2016;**6**:e010689.
14. Chen CC, Wang SS, Jeng FS, Lee SD. Metabolic bone disease of liver cirrhosis: is it parallel to the clinical severity of cirrhosis? *J Gastroenterol Hepatol* 1996;**11**:417—21.
15. Fisher L, Fisher A. Vitamin D and parathyroid hormone in outpatients with noncholestatic chronic liver disease. *Clin Gastroenterol Hepatol* 2007;**5**:513—20.
16. Arteh J, Narra S, Nair S. Prevalence of vitamin D deficiency in chronic liver disease. *Dig Dis Sci* 2010;**55**:2624—8.
17. Pugh RN, Murray-Lyon IM, Dawson JL, Pietroni MC, Williams R. Transection of the oesophagus for bleeding oesophageal varices. *Br J Surg* 1973;**60**:646—9.
18. *Institute of Medicine (US) Committee. Dietary reference intakes for calcium and vitamin D.* Washington DC: National Academies Press (US); 2011.
19. Braegger C, Campoy C, Colomb V, Decsi T, Domellof M, Fewtrell M, et al. Vitamin D in the healthy European paediatric population. *J Pediatr Gastroenterol Nutr* 2013;**56**:692—701.
20. Cioffi M, Corradino M, Gazzerro P, Vietri MT, Di Macchia C, Contursi A, et al. Serum concentrations of intact parathyroid hormone in healthy children. *Clin Chem* 2000;**46**:863—4.
21. Chin KY, Ima-Nirwana S, Ibrahim S, Mohamwd IN, Wan Ngah WZ. Vitamin D status in Malaysian men and its associated factors. *Nutrients* 2014;**6**:5419—33.
22. Shneider BL, Magree JC, Bezerra JA, Haber B, Karpen SJ, Raghunathan T, et al. Efficacy of fat-soluble vitamin supplementation in infants with biliary atresia. *Pediatrics* 2012;**130**:e607—14.
23. Shen YM, Wu JF, Hsu HY, Ni YH, Chang MH, Liu YW, et al. Oral absorbable fat-soluble vitamin formulation in pediatric patients with cholestasis. *J Pediatr Gastroenterol Nutr* 2012;**55**:587—91.
24. Venkat VL, Shneider BL, Magee JC, Turmelle Y, Arnon R, Bezerra JA, et al. Total serum bilirubin predicts fat-soluble vitamin deficiency better than serum bile acids in infants with biliary atresia. *J Pediatr Gastroenterol Nutri* 2014;**59**:702—7.

25. Hourigan SK, Abrams S, Yates K, Pfeifer K, Torbenson M, Murray K, et al. Relation between vitamin D status and nonalcoholic fatty liver disease in children. *J Pediatr Gastroenterol Nutr* 2015;**60**:396—404.
26. Okajima H, Shigeno C, Inomata Y, Egawa H, Uemoto S, Asonuma K, et al. Long-term effects of liver transplantation on bone mineral density in children with end-stage liver disease: a 2-year prospective study. *Liver Transpl* 2003;**9**:360—4.
27. Ulivieri FM, Lisciandrano D, Gridelli B, Lucianetti A, Roggero P, Nebbia G, et al. Bone mass and body composition in children with chronic cholestasis before and after liver transplantation. *Transplant Proc* 1999;**31**:2131—4.
28. Misra M, Pacaud D, Petryk A, Collett-Solberg PF, Kappy M, Drug and Therapeutics Committee of the Lawson Wilkins Pediatric Endocrine Society. Vitamin D deficiency in children and its management: review of current knowledge and recommendations. *Pediatrics* 2008;**122**:398—417.
29. Jensen M, Abu-El-Haija M, Bishop W, Rahhal RM. Difficulty achieving vitamin D sufficiency with high-dose oral repletion therapy in infants with cholestasis. *J Pediatr Gastroenterol Nutr* 2015;**61**:187—9.
30. Poh BK, Ng BK, Siti Haslinda MD, Nik Shanita S, Wong JE, Budin SB, et al. Nutritional status and dietary intakes of children aged 6 months to 12 years: findings of the Nutrition Survey of Malaysian Children (SEANUTS Malaysia). *Br J Nutr* 2013;**110**: S21—35.