Pediatric Pulmonology 38:119–122 (2004)

Serum Vitamin D Levels in Children With Cystic Fibrosis

R.J. Chavasse, FRCPCH,* J. Francis, RSCN, I. Balfour-Lynn, FRCPCH, M. Rosenthal, FRCPCH, and A. Bush, FRCPCH

Summary. Osteopenia is increasingly recognized in adults with cystic fibrosis (CF), and is potentially related to vitamin D deficiency in both adulthood and childhood. Vitamin D supplements are recommended and prescribed to all pancreatic-insufficient patients. We aimed to ascertain whether vitamin D deficiency in children with CF was prevalent. 25-hydroxyvitamin D (25-OHD) was measured in 290 children attending a specialist pediatric CF clinic for annual assessment. 25-OHD levels were compared with reference values and to other biochemical markers, lung function, and growth. Levels were also analyzed by pancreatic status and by the presence of CF-related liver disease. Median 25-OHD was 65 (range, 9–190) nmol/l. One percent had levels below 15 nmol/l, and 6% had levels less than 25 nmol/l. Levels were lower in adolescents ($P < 0.001$) and during the "winter" months ($P < 0.001$). No relationship was found with pancreatic status or liver disease. In conclusion, the majority of children had normal 25-OHD levels. Interpretation is difficult due to a lack of knowledge of optimal levels of 25-OHD required for healthy bone accretion. Lower levels in adolescents may be a precursor to low levels in adulthood, and did not seem to be simply related to poor compliance with supplementation. This may reflect normal physiology. **Pediatr Pulmonol. 2004; 38:119–122.** © 2004 Wiley-Liss, Inc.

Key words: cystic fibrosis; 25-hydroxyvitamin D; osteopenia; pancreatic insufficiency; CF-related liver disease.

INTRODUCTION

Reduced bone mineral density is a significant cause of morbidity in adults with cystic fibrosis (CF), leading to vertebral collapse, rib fractures, and a decline in pulmonary function.[1] Poor bone accretion during childhood and, in particular, puberty may be responsible.[2] Deficiency of vitamin D, secondary to either fat malabsorption or reduced sunlight exposure, is one factor that may affect bone accretion.

In cystic fibrosis, vitamin D may be deficient from early life[3] and can persist or occur at other times, despite supplementation with vitamins and pancreatic enzymes. The incidence of vitamin D deficiency in children with CF was reported to be between 4–30%, depending on the geographical location or definition of deficiency used,[4,5] and may be lower than in non-CF controls.[6] Overt bone disease, such as rickets, is rarely seen in children with CF.[5] Regular assessment of vitamin D status has been recommended.[7,8]

We hypothesized that vitamin D levels would be low in many children with cystic fibrosis, despite supplementation, predisposing them to osteoporosis in later life. We evaluated vitamin D levels in children attending our specialist pediatric CF clinic for their yearly assessment to evaluate whether vitamin D deficiency was prevalent, whether there were predictors of deficiency, and whether the current vitamin supplementation regimen was sufficient.

© 2004 Wiley-Liss, Inc.

METHODS

Vitamin D, as 25-hydroxyvitamin D (25-OHD), has been measured as part of the annual assessment protocol at our tertiary pediatric CF center since mid-1999. We identified all patients, aged 1–18 years, with confirmed CF (positive sweat ± genetic testing) under the care of our clinic and who attended annual assessments during the study period. Levels of 25-OHD measured between August 1999–April 2001 were compared to the laboratory reference range[9] and a pediatric reference range derived from healthy British children.[10] Levels were also compared to other fat-soluble vitamin levels (vitamins A and E), bone and liver biochemical parameters, pulmonary function, growth, and date of testing. Pancreatic-insufficient children in our clinic are routinely prescribed a daily vitamin D supplement of 800–1,200 IU (20–30 mcg). These

Department of Paediatrics, Royal Brompton Hospital, London, UK.

*Correspondence to: Dr. R.J. Chavasse, Queen Mary's Hospital, Wrythe Lane, Carshalton, Surrey SM5 1AA, UK.
E-mail: richard.chavasse@epsom-sthelier.nhs.uk

Received 18 August 2003; Revised 30 December 2003; Accepted 28 January 2004.

DOI 10.1002/ppul.20047
Published online 14 May 2004 in Wiley InterScience (www.interscience.wiley.com).

TABLE 1— Vitamin Preparations Used in Our Clinic[1]

	Preparation	Vitamin D	Dose
Abidec (Pfizer UK)	Liquid	10 mcg/0.6 ml	<1 yr, 0.6 ml; 1–4 yrs, 1.2 ml; 4–8 yrs, 1.8 ml
Dalivit (Eastern UK)*	Liquid	10 mcg/0.6 ml	<1 yr, 0.6 ml; 1–4 yrs, 1.2 ml; 4–8 yrs, 1.8 ml
Multivitamin BPC (np)	Tablet	7.5 mcg/tab	>8 yrs, 2–3 capsules
Vitamin A & D (np)*	Capsule	10 mcg/cap	>8 yrs, 2–3 capsules

[1]10 mcg = 400 i.u.
yr, year; yrs, years; np, non proprietary.
*Current recommended formulations based on similarity of vitamin constituents.
All formulations used during the study period.

supplements are available free of charge to children by the National Health Service (see Table 1 for formulations).

Vitamin D status was also analyzed for subgroups of patients with pancreatic sufficiency and those with liver disease. 25-OHD levels of pancreatic-sufficient children (n = 13) were compared with levels from the two pancreatic-insufficient subjects best matched by age, gender, and season of testing. Pancreatic-sufficient subjects are not routinely offered vitamin supplementation. Similarly, 25-OHD levels were compared between children with CF-related liver disease with two age-, sex-, and season-matched subjects with no documented liver involvement. For this purpose, children with liver disease were defined as those receiving ursodeoxycholic acid, as an easily identifiable surrogate marker; by protocol, we prescribe ursodeoxycholic acid for children with abnormal liver ultrasound findings or persistently raised liver enzymes. No subject had liver failure.

Change in 25-OHD level was assessed in all children who had two measurements performed 12 months apart during the study period.

25-OHD was measured by an in-house, competitive protein-binding assay following extraction and chromatography of 25-OHD on silicic acid,[9] performed at Charing Cross Hospital. This assay provides comparable results to radioimmunoassay techniques[4,11] reported elsewhere (personal communication, Dr. G Carter, Clinical Biochemistry, Charing Cross Hospital). The laboratory reference range (15–100 nmol/l) was developed with 40 healthy adults,[9] and compares favorably with a pediatric reference range described in a population of 72 children in London, using a similar assay.[12] Results were also compared with national reference data acquired from 1,150 healthy children (56% of a nationally representative sample of children aged 4–18 years) measured by radioimmunoassay (Incstar, Minnisota) (males, mean 62 nmol/l; 2.5th and 97.5th percentiles, 15.3–127.7 nmol/l; females, 60.6, 15.8–127.1)[10] 25 nmol/l is generally regarded as the lower limit of normal.[1,10]

Spirometry was assessed in all children over 6 years of age, using standard methodology to measure forced expiratory volume in 1 sec (FEV_1) and forced vital capacity (FVC).

Data analysis was performed on SPSS version 9. The relationship between 25-OHD and other parameters was assessed by Spearman correlation coefficient. Comparisons between groups were made by the Mann-Whitney U-statistic. Change in 25-OHD between repeat measurements was made by Wilcoxon sign-rank test.

RESULTS

Three hundred and twenty subjects had an annual assessment during the study period. Two hundred and ninety (91%) had 25-OHD measured on at least one occasion. The median age (range) was 9.0 years (0.9–18.5 years) (Table 2). Fifty-three percent were girls. The median serum 25-OHD was 65 nmol/l (range, 9–190 nmol/l), following a normal distribution. Four subjects (1%) had 25-OHD levels below the laboratory reference range of 15 nmol/l,[9] and 17 (6%) less than 25 nmol/l. There was no correlation with serum vitamin A or E, calcium, phosphate, alkaline phosphatase, pulmonary function (percent predicted FEV_1 or FVC), or weight or height Z-scores. There were significantly lower levels in adolescents (age >13, n = 76, median 53 nmol/l, range 9–130) compared to children under 5 years (n = 56, median 75 nmol/l, range 32–190) or children between 5–12 years (n = 158, median 70 nmol/l, range 20–183; see Fig. 1).

25-OHD levels were significantly lower in the ''winter'' months, i.e., October–March (59 nmol/l, SEM 5.2) compared to April–September (77 nmol/l, SEM 6.1) ($P < 0.001$). Seventy-five percent of low levels, irrespective of definition, were measured in the ''winter'' months.

There was no difference between pancreatic-sufficient children (n = 13, median 60 nmol/l, range 25–135) and matched pancreatic-insufficient subjects (n = 26, median 72 nmol/l, range 9–162). Nor was there any difference between children with CF-related liver disease (n = 21, median 55 nmol/l, range 14–102) and matched non-liver-disease subjects (n = 40, median 60 nmol/l, range 16–83).

One hundred and ten children had two measurements of 25-OHD within the study period. The median change over 12 months was −2 nmol/l (range, −52 to 72 nmol/l). There

TABLE 2— Characteristics of Subjects by Age Range, Including Biochemical, Anthropological, and Pulmonary Markers (Mean ± 2 SD)

	Age 1–4	Age 5–12	Age >13	Total
Number	56	158	76	290
Age (range)	2.3 (0.9–4.8)	8.3 (5.0–12.8)	15.1 (13.0–19.0)	9 (0.9–19.0)
Male (%)	27 (48)	66 (42)	38 (50)	131 (45)
Winter:summer	24:32	79:81	39:37	142:150
Pancreatic-sufficient	4	5	4	13
Liver disease	2	8	11	21
Calcium (mmol/l) (2.2–2.62)	2.44 (±0.14)	2.42 (±0.52)	2.37 (±0.16)	2.4 (±0.4)
Inorganic phosphate (mmol/l) (0.8–1.4)	1.74 (±0.4)	1.6 (±0.4)	1.4 (±0.46)	1.6 (±0.5)
Alkaline phosphate (U/l)	737 (±355)	657 (±367)	637 (±551)	667 (±425)
Height Z-score	−0.29 (±2.7)	−0.49 (±2.2)	−0.54 (±2.1)	−0.4 (2.3)
Weight Z-score	−0.34 (±2.7)	−0.24 (±2.0)	−0.51 (±2.4)	−0.3 (±2.3)
FEV_1 % predicted	n/a	86 (±37.1)	84 (±43)	85.6 (±41)
FVC % predicted	n/a	93.5 (±32.5)	95 (±31)	93.4 (±35.2)

was no association with age or level of 25-OHD in the first year.

DISCUSSION

The majority of children seemed to have normal values of 25 OHD. Only 1% of children had levels below the reference range (15 nmol/l).[9,10,12] Only three patients had values which fell below the 2.5th percentile of their respective age- and sex-specific ranges.[10] Other series used different definitions of low 25-OHD (range, 25–40 nmol/l),[3,4,11] and using these definitions, up to 14% of this clinic could be classed as deficient: 25 nmol/l is usually accepted as the lower limit of normal,[1,3,10] and 6% of the clinic would be classified as deficient by this criterion. A similar proportion of healthy children also had 25-OHD levels below this threshold.[10]

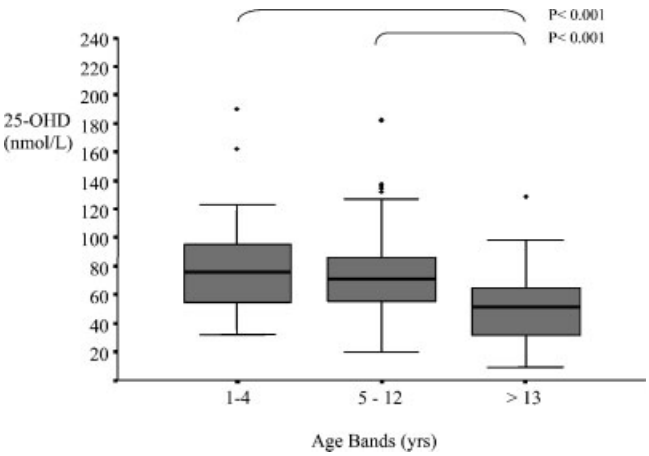

Fig. 1. 25-OHD levels in children with cystic fibrosis by age group: ages 1–4 (n = 56), ages 5–12 (n = 158), and age greater than 13 (n = 76). Boxplot shows median, quartiles, and range with outliers.

Our results differ from one study that demonstrated significantly lower 25-OHD levels in CF subjects compared to non-CF subjects, despite excess dietary vitamin D intake in the CF population.[6] However, levels in the 11 CF subjects (55 nmol/l, range 19–102) were comparable to the normal reference values,[9,10,12] whereas the control population was significantly higher (98 nmol/l, range 38–245). The reasons for this were not discussed in the original paper, but one could hypothesize that exaggerated sunlight exposure in the healthy population was associated with local climactic conditions or skewed data compounded by the small sample size.

25-OHD was significantly lower during the adolescent years, confirming a previous report.[11] This also mirrors the normal population,[10] but there is no clear explanation for this phenomenon either in health or disease. Low vitamin levels in CF are frequently ascribed to poor compliance with supplementation, a problem more manifest in adolescence. This is unlikely to be the sole factor, as no correlation was found with vitamin A and E levels (particularly vitamin A, which is taken as a joint supplement with vitamin D), and no similar age-related decline was seen in these other vitamins. Another explanation may be general decline in health, although there was no correlation to predicted pulmonary function or anthropometry. Hypovitaminosis D is more common in adults with CF (36% <25 nmol/l),[1] and therefore it is possible that there is a progressive decline from adolescence.

Seasonal variation in 25-OHD is not reflected in the published reference range utilized by our laboratory,[9] but was widely reported in temperate climates.[10,11,13] The majority of low levels were recorded in the "winter" months, irrespective of definition of deficiency. Although the sample was cross-sectional, we can assume that many low levels in "winter" normalize in "summer" as a consequence of increased sunlight exposure. The significance of a temporary low level is unclear but potentially

important.[13] Although not practical for our clinic, it may be more useful to measure 25-OHD levels during the same season to eliminate this variation.

Normal ranges provide only limited information, and as yet there is no consensus of the optimal range of 25-OHD for healthy bone formation. The optimal level may vary at different ages and disease states, and may potentially exceed 80 nmol/l at times.[14] At this level, the majority of the clinic (64%) could be classed as deficient. Monitoring changes in parathyroid hormone (PTH) may help indicate suboptimal levels of 25-OHD, as parathyroid hormone secretion rises in deficiency states (secondary hyperparathyroidism), but this is not without difficulty.[14] Mortensen et al.[6] reported no significant difference in PTH between CF and non-CF subjects, despite significantly different 25-OHD levels. Some patients in both groups had raised PTH levels, potentially indicating suboptimal 25-OHD levels in these individuals.[6]

Predictors of low bone mineral density in adults included poor lung function, frequent courses of intravenous antibiotics, and increased energy requirements.[1] Pancreatic sufficiency was not protective. Persistent lung inflammation, reduced physical activity, corticosteroid therapy, delayed puberty, poor nutrition,[1] and vitamin K deficiency (effect on osteocalcin and matrix gamma-carboxyglutamic acid)[15] may also contribute to decreased bone accretion in childhood. Although vitamin D has a well-recognized role in bone metabolism, the importance in CF-related bone disease is unknown and may be superseded by other factors. Potentially, children with CF may require increased 25-OHD levels, in excess of healthy children, to optimize bone accretion, and current levels of supplementation may be too low. In one study, increasing supplementation up to 2,000 IU in children with CF did not result in significant changes in bone mineralization.[16]

Routine assessment of vitamin D has been suggested for all patients with CF.[5] Restricting measurement to high-risk groups[11] would seem inappropriate, as 25-OHD levels may be low at diagnosis even in asymptomatic infants detected by screening.[3] Care must be taken in interpretation of results with particular reference to age, sex, and season of testing. Optimal, rather than normal, levels for healthy bone accretion need to be established for children with CF by evaluating 25-OHD in association with parathyroid hormone and bone mineral status in a large, longitudinal population study. Promotion of good bone health is imperative, and advice should include exercise, diet, and sunlight exposure as well as compliance with vitamin supplementation.

REFERENCES

1. Elkin SL, Fairney A, Burnett S, Kemp M, Kyd P, Burgess J, Compston JE, Hodson ME. Vertebral deformities and low bone mineral density in adults with cystic fibrosis: a cross sectional study. Osteoporosis Int 2001;12:366–372.
2. Laursen EM, Mölgaard C, Michaelsen KF, Koch C, Müller J. Bone mineral status in 134 patients with cystic fibrosis. Arch Dis Child 1999;81:235–240.
3. Feranchak AP, Sontag MK, Wagener JS, Hammond KB, Accurso FJ, Sokol RJ. Prospective long term study of fat soluble vitamin status in children with cystic fibrosis identified by newborn screen. J Pediatr 1999;135:601–610.
4. Henderson RC, Lester G. Vitamin D levels in children with cystic fibrosis. South Med J 1997;90:378–383.
5. Hubbard VS, Farrell PM, di Sant'Agnese PA. 25-hydroxycholecalciferol levels in patients with cystic fibrosis. J Pediatr 1979;94:84–86.
6. Mortensen LA, Chan GM, Alder SC, Marshall BC. Bone mineral status in prepubertal children with cystic fibrosis. J Pediatr 2000;136:648–652.
7. Cystic Fibrosis Trust. Standards for the clinical care of children and adults with cystic fibrosis. London: CF Trust; 2001.
8. Ramsey BW, Farrell PW, Pencharz P. Nutritional assessment and management in cystic fibrosis: a consensus report. Am J Clin Nutr 1992;55:108–116.
9. Eastwood JB, Daly A, Carter GD, Alaghband-Zadeh J, De Wardener HE. Plasma hydroxyvitamin D in normal subjects and patients with terminal renal failure on maintenance haemodialysis and after transplantation. Clin Sci 1979;57:473–476.
10. Gregory J, Lowe S, for Great Britain Office for National Statistics Social Survey Division. National diet and nutritional survey: young people aged 4 to 18 years. Volume 1. London: Stationary Office; 2000.
11. Grey V, Lands L, Pall H, Drury D. Monitoring of 25-OH vitamin D in children with cystic fibrosis. J Pediatr Gastroenterol Nutr 2000;3:314–319.
12. Chong SKF, Moniz C, Turner C. The role of vitamin D metabolites in the management of rickets complicating liver disease. In: Third International Congress of Paediatric Laboratory Medicine, Bristol. 1986.
13. Haddad J. Seasonal diminuition of vitamin D stores in the United States. Can darker winters lead to lighter bones? Trends Endocrinol Metab 1996;7:209–212.
14. Dawson-Hughes B, Harris SS. Definition of the optimal 25-OHD status for bone. In: Norman A, Bouillon R, Thomasset M, editors. Vitamin D endocrine system: Structural, biological, genetic and clinical aspects. Berlin: W. de Gruyter. 2000. p 909–915.
15. Beker LT, Ahrens RA, Fink RJ, O'Brien ME, Davidson KW, Sokoll LJ, Sadowski JA. Effect of vitamin K1 supplementation on vitamin K status in cystic fibrosis patients. J Paediatr Gastroenterol Nutr 1997;24:512–517.
16. Hillman L, Barbero G, Popescu M, Chanesta F, Bergfield M, Lee D, Cassidy J. Supplementation with 2000 IU vitamin D with or without 1 g calcium in children with cystic fibrosis or juvenile chronic arthritis. In: Norman A, Bouillon R, Thomasset M, editors. Vitamin D endocrine system: Structural, biological, genetic and clinical aspects. Berlin: W. de Gruyter. 2000. p 897–900.