Effect of long-term glucocorticoid therapy on bone mineral density of the patients with congenital adrenal hyperplasia

Sezin Ünal1,2, Ayfer Alikaşifoğlu1, Alev Özön1, Nazlı Gönç1, Nurgun Kandemir1

1Division of Pediatric Endocrinology, Department of Pediatrics, Hacettepe University Faculty of Medicine, Ankara; 2Division of Neonatology, Department of Pediatrics, University of Health Sciences, Etlik Zübeyde Hanım Women’s Health Teaching and Research Hospital, Ankara, Turkey.

ABSTRACT

Background and objectives. Congenital adrenal hyperplasia (CAH) is characterized by androgen excess which should be treated with life-long glucocorticoid therapy, thus can affect bone mineralization. We aimed to evaluate the bone mineral density (BMD) and determine the factors affecting bone mineralization in patients with CAH.

Method. This prospective case-control study was conducted in children, adolescents and young adults with classical 21-hydroxylase CAH, and age-, sex-, and pubertal stage matched healthy controls. Lumbar BMD was determined by dual-energy X-ray absorptiometry. BMD z-score was calculated using national standards with respect to height age and was referred as “low BMD” if z-score < -1 SD. Univariate analyses were performed between low BMD and normal BMD groups, and multivariate logistic regression analysis was performed to assess the independent predictors of low BMD. Correlations of Body Mass Index (BMI)-z-score, average serum 17-hydroxyprogesterone level, duration of treatment, average and cumulative glucocorticoid doses with BMD z-score were evaluated with Spearman analyses.

Results. Each group included 37 cases. BMD z-score of patients with CAH [0.47 (-0.04 – 1.56)] was higher than control group [-0.43 (-0.82 –0.05)]; p= < 0.001. Number of patients with low BMD was similar in both groups; [CAH: 6(16.2%), control: 5(13.5%); p= 0.744]. BMI- z-score was higher in patients with CAH when compared to control group; p= < 0.001. BMI z-score was lower in low BMD group as comparison to normal BMD group; p= 0.041. Each 1.0 decrease in BMI z-score, risk of having low BMD was found to increase by 1.79 (%95 CI: 1.03-3.12, p= 0.040). BMI-z-score, average serum 17-hydroxyprogesterone level, duration of treatment, average and cumulative glucocorticoid doses were not found to be correlated with BMD z-score.

Conclusion. Long-term glucocorticoid therapy did not have negative effect on BMD of patients with CAH. Higher BMI z-score in patients with CAH may have a positive effect on preserving bone health. Precautions should be taken for increased risk of obesity.

Key words: congenital adrenal hyperplasia, bone mineral density, dual-energy X-ray absorptiometry scan, body mass index.
treatment may decrease bone mineralization by reducing skeletal growth factors, intestinal and renal calcium reabsorption, and increasing bone turnover. In addition, over suppression of androgens due to excessive treatment may also affect bone mineralization negatively. Instead, insufficient treatment may result in elevation of androgens, thus, may have a protective effect against anticipated bone loss.

Most of the studies on adult patients were shown to lead to decreased bone mineral density (BMD). On the other hand, studies on children alone yielded conflicting results; some showed unchanged, others showed reduced or increased BMD using dual-energy X-ray absorptiometry (DEXA) in comparison to healthy children. As those studies on children have variable results regarding the BMD in children with CAH, there is a need for further studies.

CAH is considered to be more common in Turkey than western countries due to a high frequency of consanguinity. A comprehensive review of patients with CAH in Turkey reported that 21-OH CAH accounts for 85.7% of the patients. The aim of this study was to compare BMD in Turkish children, adolescents and young adults with classical 21-OH CAH to age, sex and pubertal stage matched healthy subjects, and determine the factors affecting the bone mineralization.

**Material and Methods**

This prospective case-control study was approved by Hacettepe University Ethics Committee (LUT 06/63 – 58) and funded by Hacettepe University Scientific Research Unit Grant (06-D07-101-005). Written informed consent was obtained from the parents of patients with CAH and the healthy controls for their participation in the study.

We included children older than three years, as well as adolescents and young adults with classical 21-OH CAH, who had been on glucocorticoid replacement for more than two years. Age, sex and pubertal stage matched healthy subjects composed the control group. None of the study subjects received calcium or vitamin D supplementation at the time of enrollment.

Diagnosis was made on clinical features (vomiting, dehydration, shock, failure to thrive, ambiguous genitalia in females, and hyperandrogenism in males) and elevated serum 17-hydroxypogesterone (17-OHP) > 300 nmol/L. Patients were accepted as salt wasting 21-OH CAH if hyponatremia (serum sodium < 135 mmol/L), hyperkalemia (serum potassium > 5.5 mmol/L) and increased plasma renin activity was documented. They were considered as simple virilizing CAH if serum electrolytes and plasma renin activity were within normal limits at the time of initial diagnosis.

All patients with CAH and controls underwent thorough physical examination. We recorded chromosomal sex, pubertal stage, body weight to the nearest 0.1 kg, and height to the nearest 0.1 cm. The body mass index (BMI) was calculated using the formula; BMI=Weight(kg)/Height(m^2). Subjects were considered as overweight if a BMI percentile of 85-95 percentile with respect to national standards, and as obese if a BMI percentile greater than > 95 p with respect to national standards and BMI z-score was calculated. Overweighed and obese subjects were analyzed together.

Average glucocorticoid dose was calculated as; sum of glucocorticoid replacement dose at each assessment divided by total number of assessments and presented as mg/m^2/day, hydrocortisone equivalents. Cumulative glucocorticoid dose was calculated as; average glucocorticoid dose x 365 x duration of treatment in years and presented as gr/m^2. Glucocorticoid doses were converted to hydrocortisone equivalents as follows: 5 mg hydrocortisone=1 mg prednisolone. Average serum 17-OHP level (Radioimmune assay) was calculated as; sum of serum 17-OHP level at each assessment divided by total number of assessments and presented as mmol/L. Patients
were identified as poor control if mean serum 17-OHP > 60 nmol/L.21

The BMD of the patients and healthy subjects in the case and control groups were determined using DEXA, Hologic® QDR 4500A. The examination was performed in supine position BMD (gr/cm²) and bone mineral content (BMC; gr) were determined from lumbar vertebrae (L₁₋₄). BMD z-score was calculated using national standards with respect to height age.22 Included patients and healthy subjects were grouped according to BMD z-score, where BMD z-score higher than -1.0 was considered as “normal BMD”, and those lower than -1.0 were considered as “low BMD”.23 Bone Mineral Apparent Density (BMAD; BMC/vertebral area; gr/cm³) was calculated.24

Statistical analyses were performed using SPSS V15.0 for Windows (SPSS, Chicago, IL, USA). Categorical variables were presented as n (%), and were compared by Chi-square test, Fisher exact test or Yates correction, where appropriate. A p value of 0.05 was considered statistically significant. The distribution of numerical variables was investigated using visual and analytic methods and compared between patients and controls using Mann-Whitney U test. Results were presented as median (IQR; interquartile range). Univariate analyses were performed between patients and controls, and low BMD and normal BMD groups among both all subjects and only patients with CAH. Variables with p value < 0.10 in univariate analysis between low BMD and normal BMD groups were included in multivariate logistic regression analysis models (backward stepwise model) to assess the independent predictors of low BMD. Hosmer-Lemeshow goodness of fit statistics were used to assess the model fit. Spearman correlation analyses were performed between BMD z-score and BMI z-score, average serum 17-OHP level, average and cumulative glucocorticoid dose and duration of treatment in patients.

Results

A total number of 37 patients with classical 21-OH CAH (salt wasting; n= 26, simple virilizing; n= 11) were included in the study. Thirty-seven age, sex, and pubertal stage matched healthy subjects formed the control group. The age at the time of enrollment was 10.0 (7.5 – 15.5) years in patients with CAH and 10.5 (6.1 – 14.5) years in control group; p= 0.546. Each group included 15 (40.6%) male and 19 (51.4%) prepubertal subjects.

Median duration of follow-up in patients with CAH was 9.2 (IQR: 6.6 – 12.5) years. Twenty-two patients were diagnosed in the first year of life, of which 10 were diagnosed during the neonatal period. The median age of diagnosis of the remaining 15 patients diagnosed after the first year of life was 3.0 (IQR: 2.1 – 6.3) years. Patients were on steroid treatment for a median of 9.2 (6.4 – 12.5) years, and 11 out of 37 were on prednisolone treatment for 2 (1.0 - 3.6) years. With regards to gender assignment 20 patients with CAH (46,XX) were assigned female while, two patients with 46,XX were raised as male due to development of male gender identity. Clinical characteristics of patients with 21-OH CAH are shown in Table I. Fourteen patients were poor controlled.

BMD z-score of patients with CAH [0.47 (-0.04 – 1.56)] was higher than the control group [-0.43 (-0.82 – 0.05)]; p= < 0.001. There were similar number of patients with low BMD; [n (%); CAH: 6 (16.2%) vs control group:5 (13.5%); p= 0.744]. BMD, BMC, and BMAD of patients did not differ significantly from healthy controls (0.254, 0.701, 0.534 respectively). Height SDS of patients and controls were statistically similar; p= 0.234. However, BMI and BMI z-score were significantly higher in patients with CAH in comparison to controls. Likewise, there were more overweight/obese subjects in patients with CAH than in the control group; 54.1% vs 18.9%, p= 0.002. The data are summarized in Table II.
Low BMD and normal BMD groups were compared according to age, BMI z-score, being CAH, male gender, prepubertal status and overweight/obesity as detailed in Table III. Of all comparisons, BMI z-score was found to be significantly lower in low BMD group as comparison to normal BMD group; \( p= 0.041 \). Multivariate logistic regression model including BMI z-score and male gender revealed that each 1.0 decrease in BMI z-score, risk of having low BMD increases by 1.79 (%95 CI: 1.03-3.12, \( p= 0.040 \)).

Twenty-three patients with well controlled CAH [9.5 (6.8 – 14.9) years] and 23 control subjects [10.4 (6.0 – 12.3) years] were compared for BMI z-score, BMI z-score, and ratio of cases with low BMD. Each group included 8 (34.8%) male and 15 (65.2%) prepubertal subjects. Patients with well controlled CAH had higher BMI z-score [0.42 (-0.44 – 1.33) vs -0.60 (-0.89 – -0.08); \( p= 0.005 \)], and BMI z-score [1.37 (1.05 – 2.16) vs -0.20 (-0.50 – 0.98); \( p= 0.003 \)] as compared to control group. Four patients (17.4%) in CAH group and three subjects (13%) in control group

| Table I. Clinical characteristics of patients with 21-hydroxylase congenital adrenal hyperplasia. |
|-----------------------------------------------|
| **n= 37**                                    |
| Age; years                                   |
| Male; n (%)                                  |
| Prepubertal status; n (%)                    |
| Salt wasting / simple virilizing n (%)       |
| Diagnosis within first year; n (%)           |
| Bone age / chronological age at the time of diagnosis |
| Bone age / chronological age at the time of evaluation |
| Duration of treatment; years                 |
| Prednisolone treatment, n (%)                |
| Average glucocorticoids dose, hydrocortisone equivalent; mg/m²/day |
| Cumulative glucocorticoid dose, hydrocortisone equivalent; g/m² |
| Average serum 17-hydroxyprogesterone level; nmol/L |
| Values are represented as n (%) or median (interquartile range) where appropriate. |

| Table II. Bone mineral densitometry and body mass index results in patients with congenital adrenal hyperplasia and control group. |
|----------------------------------------------------------------------------------------------------------------------------------|
| Patients with CAH (n= 37)                                                                                                            |
| Control group (n= 37)                                                                                                                |
| **p-value** |
| BMD z-score                                                                                                                           |
| Low BMD, n (%)                                                                                                                        |
| BMD, gr/cm²                                                                                                                            |
| BMC, gr                                                                                                                                |
| BMAD, gr/cm²                                                                                                                           |
| Height SDS                                                                                                                               |
| BMI, kg/m²                                                                                                                              |
| BMI z-score                                                                                                                              |
| Overweight/Obesity, n (%)                                                                                                               |
| Values are represented as n (%) or median (interquartile range) where appropriate. |

BMD: Bone mineral density, BMC: Bone mineral content, BMAD: Bone mineral apparent density, BMI: Body mass index, CAH: Congenital adrenal hyperplasia, SDS: Standard deviation score.
had low BMD; p= 0.500. Fourteen patients with poor controlled CAH [12.8 (8.0 – 16.0) years] and 14 control subjects [12.6 (6.4 – 16.0) years] were analyzed for same parameters. Each group included 7 male (50%) and 4 (28.6%) prepubertal subjects. BMD z-score [CAH: 0.64 (-0.04 – 1.94) vs Control: -0.26 (-0.71 – 0.72); p= 0.077] and BMI z-score [CAH: 0.50 (-1.32 – 1.25) vs Control: 0.95 (-0.13 – 1.72); p= 0.041] were higher in patients with poor controlled CAH than control group. There were 2 subjects with low BMD in each group. BMD z-score and BMI z-score of patients with well controlled CAH and poor controlled CAH were similar; p= 0.546 and p= 0.841 respectively.

Among 37 patients with CAH, six patients had low BMD and 31 patients had normal BMD. Low BMD group included fewer male patients [n (%): 2 (33.3%) vs 16 (51.6%); p= 0.414], less overweight/obese patients [n (%): 2 (33.3%) vs 18 (58.1%); p= 0.266] as compared to normal BMD group, although not statistically significant. BMD z-score of CAH patients with low BMD was lower than control group but not statistically significant [Low BMD: 0.10 (-1.42 – 1.44) and Normal BMD: 1.44 (0.95 – 2.16); p= 0.113]. Ratio of prepubertal [n (%): 3 (50.0%) vs 16 (51.6%); p= 0.942] and poor controlled [n (%): 2 (33.3%) vs 12 (38.7%); p= 1.000] patients were similar in low and normal BMD groups. Salt wasting 21-OH CAH accounted for all patients in low BMD group and 65% of patients in normal BMD group; p= 0.080.

BMI z-score, average serum 17-OHP level, average glucocorticoid dose, cumulative glucocorticoid dose and duration of treatment were not found to be correlated with BMD z-score in patients with CAH as shown in Table IV.

**Discussion**

In this prospective study, we demonstrated that BMD z-score in patients with 21-OH CAH was increased in comparison to age, sex and puberty matched healthy controls. Previous studies
found that bone mineralization of children with CAH was either unchanged, reduced or increased when compared to healthy children.3,6-17

As reviewed in the clinical report of American Academy of Pediatrics about assessment of bone densitometry in children and adolescents, evaluation of results of DEXA is a debate and may require more than the calculation of z-score. Chronic illness may result in either delayed or advanced growth and pubertal development, which are factors that contribute to a low bone mass for age or gender. BMD, calculated by DEXA as BMC/cm², adjusts bone mineral content for the area, but not for the volume of bone. Bearing this in mind, if two people of different heights but similar ages have comparable volumetric bone density, the shorter person will be reported to have a lower BMD and BMD z-score than the taller one. Similarly, tall people have big and large vertebrae, thus BMD z-score will be higher than actual. Likewise, pubertal problems will cause alterations in bone size, geometry, and density that occur with sex-steroid exposure. Although controversy exists concerning the optimal method for adjusting variations in bone size, body composition, and maturity, BMD results are recommended to be adjusted for height or height age over age-, gender- specific z-score.25 Besides, children with CAH often have accelerated puberty, whose outcome on bone mineralization cannot not be denied. Moreover, it is known that patients with CAH could be either taller or shorter than chronological age, while the bone age being either advanced or delayed. Ganesh et al.15 reported that BMD and BMC results were well correlated with height for age of children with CAH. Bearing in mind that the presence of variations in the anthropometric measurements we evaluated BMD z-scores for height age as some studies in the literature.13,15,16

Increased risk of obesity in patients with CAH was emphasized in many studies and attributed to the glucocorticoid treatment to increase the fat mass rather than the increased serum androgens to increase the lean mass, the advanced bone age maturation and parental obesity.9,19,26 As in line with those studies, we also found that there were approximately three times more obese subjects among patients with CAH than in control group. Moreover, our results showed that BMD was higher in patients with CAH when compared to the control group. This finding can be attributed to higher BMI z-score in patients as; each 1 decrease in standard deviation score of BMI was found to increase the risk of having low BMD by 1.79 supporting the previous studies.9,13 The protection of higher BMI on bone density loss in adult patients with CAH was also emphasized in adult patients. The excess adipose tissue leading to increased conversion of estrogen from adrenal precursors and positive effect of a better nutritional condition on bone mineralization were said to be probable reasons to explain positive effect of increased BMI on BMD.13,27 Moreover, mechanical loading on weight bearing bones may contribute to the higher BMD z-scores in patients with higher BMI z-score.28

Children with CAH require lifelong glucocorticoid treatment. Glucocorticoids give rise to deficient bone mineralization by direct suppression of osteoblastic activity, inhibiting calcium absorption from the gut, thereby leading to secondary hyperparathyroidism and increasing bone resorption by the osteoclasts, and inhibiting renal tubular calcium resorption.2

In our study, BMD z-score were found to be related neither with dose (mean or cumulative) nor the duration of the treatment as in line with the literature.14 Similarly, currently used replacement doses of glucocorticoids was found not to have a major impact on bone in patients with CAH when bone mineralization was assessed by quantitative ultrasonometry.29 Nevertheless, data about the effect of glucocorticoid treatment varies and many studies that relate osteopenia in patients with CAH to glucocorticoid use exist in the literature. Not only the dose but the duration of treatment was evaluated thoroughly and found to be
negatively correlated with bone mineralization in some studies.\textsuperscript{9,13,15,17} Prednisolone treatment instead of hydrocortisone was shown to decrease bone mineralization.\textsuperscript{5} Eleven patients in our study were on prednisolone treatment for 2.0 (1.0 - 3.6) years, which is quite a small percentage of total duration of treatment 9.2 (6.4 – 12.5) years. Thus, no detailed analyses were performed regarding steroid type. Previous studies showed increased, decreased or unchanged bone mineralization in patients with poor controlled CAH as compared to patients with well controlled CAH.\textsuperscript{6,8,21} In cases with poor controlled CAH, elevated sex steroids and decreased exposure to glucocorticoids may have positive effect on BMD. However, the ratio of patients with poor controlled CAH in low BMD and normal BMD groups were similar. In addition to this, patients with well controlled CAH and with poor controlled CAH had similar BMD z-scores, and serum 17-OHP levels were not correlated with BMD z-score.

Puberty is a significant stage of development which is known to affect bone mineralization. While Gussinye et al.\textsuperscript{7} stated that prepubertal children with CAH had higher BMD z-score, Yilmaz et al.\textsuperscript{30} showed that bone mineralization is affected positively during puberty. No difference in number of prepubertal subjects was observed between low BMD and normal BMD groups.

Patients with salt wasting 21-OH CAH were shown to have lower BMD when compared to late onset patients, but no variation between the forms of classical CAH was shown to exist with regards to bone mineralization.\textsuperscript{12} We also noted similar percentage of patients with salt wasting 21-OH CAH in low BMD and normal BMD groups.

Small sample size, wide range in the age of subjects, inclusion patients on prednisolone treatment limit the results of our study to be generalized to all patients with CAH. Also current literature suggest the evidence of vitamin D deficiency and genetic factors in the bone mineralization in the patients, which we did not evaluated.\textsuperscript{10,14,31} Despite the limitations of the current study, BMD z-score was not lower in patients with CAH, and this supports the idea that CAH itself does not affect bone mineralization negatively.

In conclusion, the results of our study revealed that an explicit negativity was not observed in bone mineralization of patients with 21-OH CAH patients followed up with appropriate treatment protocols. Moreover, higher BMI z-score may contribute to preservation of bone health in patients with CAH. In addition, it should be considered that the patients with CAH are at risk for obesity, and precautions are needed to be taken to this end.

REFERENCES

1. Speiser PW, White PC. Congenital adrenal hyperplasia. N Engl J Med 2003; 349: 776-788.
2. Chakhtoura Z, Bachelot A, Samara-Boustani D, et al; Centre des Maladies Endocriniennes Rares de la Croissance and Association Surrénales. Impact of total cumulative glucocorticoid dose on bone mineral density in patients with 21-hydroxylase deficiency. Eur J Endocrinol 2008; 158: 879-887.
3. Arisaka O, Hoshi M, Kanazawa S, et al. Preliminary report: effect of adrenal androgen and estrogen on bone maturation and bone mineral density. Metabolism 2001; 50: 377-379.
4. Ceccato F, Barbot M, Albiger N, et al. Long-term glucocorticoid effect on bone mineral density in patients with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. Eur J Endocrinol 2016; 175: 101-106.
5. Koetz KR, Venta M, Diederich S, Quinkler M. Bone mineral density is not significantly reduced in adult patients on low-dose glucocorticoid replacement therapy. J Clin Endocrinol Metab 2012; 97: 85-92.
6. Girgis R, Winter JS. The effects of glucocorticoid replacement therapy on growth, bone mineral density, and bone turnover markers in children with congenital adrenal hyperplasia. J Clin Endocrinol Metab 1997; 82: 3926-3929.
7. Gussinye M, Carrascosa A, Potau N, et al. Bone mineral density in prepubertal and in adolescent and young adult patients with the salt-wasting form of congenital adrenal hyperplasia. Pediatrics 1997; 100: 671-674.
8. Elnecave RH, Kopacek C, Rigatto M, Keller Brenner J, Sisson de Castro JA. Bone mineral density in girls with classical congenital adrenal hyperplasia due to CYP21 deficiency. J Pediatr Endocrinol Metab 2008; 21: 1155-1162.

9. Cetinkaya S, Kara C. The effect of glucocorticoid replacement therapy on bone mineral density in children with congenital adrenal hyperplasia. J Pediatr Endocrinol Metab 2011; 24: 265-269.

10. Okten A, Cakir M, Makuloglu M. Bone mineral status, bone turnover markers and vitamin D status in children with congenital adrenal hyperplasia. Minerva Endocrinol 2012; 37: 275-282.

11. Ganesh R, Suresh N, Janakiraman L. Bone mineral content and density in Indian children with congenital adrenal hyperplasia. Indian Pediatr 2018; 55: 880-882.

12. Paganini C, Radetti G, Livieri C, Braga V, Migliavacca D, Adami S. Height, bone mineral density and bone markers in congenital adrenal hyperplasia. Horm Res 2000; 54: 164-168.

13. de Almeida Freire PO, de Lemos-Marini SH, Maciel-Guerra AT, et al. Classical congenital adrenal hyperplasia due to 21-hydroxylase deficiency: a cross-sectional study of factors involved in bone mineral density. J Bone Miner Metab 2003; 21: 396-401.

14. Demirel F, Kara O, Tepe D, Esen I. Bone mineral density and vitamin D status in children and adolescents with congenital adrenal hyperplasia. Turk J Med Sci 2014; 44: 109-114.

15. Ganesh R, Suresh N, Janakiraman L, Ravikumar K. Correlation of bone mineral parameters with anthropometric measurements and the effect of glucocorticoids on bone mineral parameters in congenital adrenal hyperplasia. Indian J Pediatr 2016; 83: 126-130.

16. Halper A, Sanchez B, Hodges JS, et al. Bone mineral density and body composition in children with congenital adrenal hyperplasia. Clin Endocrinol (Oxf) 2018; 88: 813-819.

17. Lin-Su K, New MI. Effects of adrenal steroids on the bone metabolism of children with congenital adrenal hyperplasia. Ann N Y Acad Sci 2007; 1117: 345-351.

18. Kandemir N, Yordam N. Congenital adrenal hyperplasia in Turkey: a review of 273 patients. Acta Paediatr 1997; 86: 22-25.

19. Volkl TM, Simm D, Beier C, Dorr HG. Obesity among children and adolescents with classic congenital adrenal hyperplasia due to 21-hydroxylase deficiency. Pediatrics 2006; 117: e98-e105.

20. Neyzi O, Bundak R, Gokcay G, et al. Reference values for weight, height, head circumference, and body mass index in Turkish children. J Clin Res Pediatr Endocrinol 2015; 7: 280-293.

21. Zimmermann A, Sido PG, Schulze E, et al. Bone mineral density and bone turnover in Romanian children and young adults with classical 21-hydroxylase deficiency are influenced by glucocorticoid replacement therapy. Clin Endocrinol (Oxf) 2009; 71: 477-484.

22. Goks D, Darcan S, Kose T. Bone mineral density of healthy Turkish children and adolescents. J Clin Densitom 2006; 9: 84-90.

23. Bianchi ML. Osteoporosis in children and adolescents. Bone 2007; 41: 486-495.

24. Carter DR, Bouxsein ML, Marcus R. New approaches for interpreting projected bone densitometry data. J Bone Miner Res 1992; 7: 137-145.

25. Bachrach LK, Gordon CM; Section on Endocrinology. Bone densitometry in children and adolescents. Pediatrics 2016; 138: e20162398.

26. Christiansen P, Molgaard C, Muller J. Normal bone mineral content in young adults with congenital adrenal hyperplasia due to 21-hydroxylase deficiency. Horm Res 2004; 61: 133-136.

27. Sahakitrungruang T, Wacharasindhu S, Supornsilchai V, Srivuthana S, Kingpetch K. Bone mineral density and body composition in prepubertal and adolescent patients with the classical form of 21-hydroxylase deficiency. J Med Assoc Thai 2008; 91: 705-710.

28. Vandewalle S, Taes Y, Van Helvoirt M, et al. Bone size and bone strength are increased in obese male adolescents. J Clin Endocrinol Metab 2013; 98: 3019-3028.

29. Delvecchio M, Soldano L, Lonero A, et al. Evaluation of impact of steroid replacement treatment on bone health in children with 21-hydroxylase deficiency. Endocrine 2015; 48: 995-1000.

30. Yilmaz D, Ersoy B, Bilgin E, Gumuser G, Onur E, Pinar ED. Bone mineral density in girls and boys at different pubertal stages: relation with gonadal steroids, bone formation markers, and growth parameters. J Bone Miner Metab 2005; 23: 476-482.

31. Martin S, Munoz L, Perez A, et al. Clinical and molecular studies related to bone metabolism in patients with congenital adrenal hyperplasia. J Pediatr Endocrinol Metab 2014; 27: 1161-1166.