BONE MINERAL DENSITY AND BODY COMPOSITION IN CHILDREN WITH CONGENITAL ADRENAL HYPERPLASIA

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ABSTRACT

Background: Children with congenital adrenal hyperplasia (CAH) are exposed to fluctuating cortisol and androgen levels. The effects these hormonal states have on bone mineral density (BMD) and body composition are not well studied.

Objective: Compare BMD and body composition in children with CAH vs. healthy age-, sex-, and BMI-matched controls.

Methods: Forty-two cases with CAH and 101 controls underwent a dual-energy X-ray absorptiometry scan. Bone age Z-scores were used as a surrogate for long-term androgen exposure in cases.

Results: Children with CAH had lower BMD Z-scores than controls. In CAH cases, BMD Z-scores were positively correlated with bone age Z-scores. Body composition markers did not differ between children with CAH and controls.

Conclusion: Lower BMD was observed in CAH cases, but no differences in body composition were identified. Among CAH cases, increased chronic androgen exposure, as measured by bone age Z-scores, was associated with higher BMD.

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List of Abbreviations:

1. congenital adrenal hyperplasia (CAH)
2. bone mineral density (BMD)
3. visceral adipose tissue (VAT)
4. bone age (BA)
5. hydrocortisone (HC)
6. Android:Gynoid (A:G)
7. body mass index (BMI)

INTRODUCTION

Congenital adrenal hyperplasia (CAH) due to 21α-hydroxylase deficiency, a form of adrenal insufficiency, is the most common cause of genital ambiguity in the newborn. Undiagnosed CAH can result in death due to salt-wasting and/or adrenal crisis. For these reasons, CAH is on the newborn screening (NBS) panel in all states in the United States. CAH is characterized by impaired cortisol synthesis and adrenal androgen over-production with a wide spectrum of clinical phenotypes based on the degree of the severity of the enzymatic defect. CAH is classified as either classic (severe phenotype) or non-classic (NC; mild phenotype). The classic phenotype is further subdivided into simple-virilizing (SV) and salt-wasting (SW) based on whether there is adequate or deficient aldosterone production, respectively.

The excessive production of adrenal androgen in patients with CAH leads to increased production of estrogen through aromatization. While estrogen is important for bone mineral density and body composition, increased estrogen levels can also result in rapid bone maturation, early epiphyseal closure, and decreased final adult height in both sexes. Treatment of CAH involves lifelong cortisol replacement is in the classic forms and in symptomatic non-classic CAH patients to replace the missing hormone and prevent the excessive production of adrenal androgen. In growing children, hydrocortisone (HC) is recommended at 10-15 mg/m^2/day in three divided doses to avoid the adverse impact on growth from long-acting steroids.[1] The challenge in treating CAH is to balance sufficient cortisol replacement to suppress overproduction of adrenal androgens (hyperandrogenemia), while avoiding the effects of cortisol excess

(hypercortisolemia), such as stunting of growth, osteopenia, hypertension, and increased weight gain. Adding to the challenge is HC's short median elimination half-life in CAH children, 58 min (range: 41-105 min), allowing most of the HC dose to be eliminated from the body within 4-5 hrs.[2] Because of HC's short half-life, the recommended 3 times a day dosing schedule exposes children to alternating periods of hypo- and hypercortisolemia with resultant hyperandrogenemia throughout each day.

Evidence for the effects of increased adrenal sex steroids in CAH and glucocorticoid therapy on bone mineral density (BMD) are mixed, with some studies finding no difference in BMD between children with CAH and controls[3-8], others finding lower BMD in children with CAH,[9-15] and one study finding increased BMD.[16] While androgens have an established positive effect on BMD through stimulation of osteoblastic activity[17], glucocorticoids can have an opposite detrimental effect. Glucocorticoids reduce BMD through several mechanisms including decreasing osteoblastic activity, decreasing calcium absorption in the gastrointestinal tract, enhancing renal losses of calcium, and increasing bone resorption.[18]

The effects of adrenal sex steroids and glucocorticoids on measures of body composition, such as visceral adipose tissue (VAT) and the Android:Gynoid (A:G) ratio (a ratio of abdominal fat to hip fat), have not been well studied in children with CAH. This area of investigation is important given that children and adults with CAH may be at an increased risk for cardiovascular disease compared to the general population.[19] VAT is associated with increased cardiometabolic risk,[20] and the A:G ratio is associated with elevated triglyceride levels and lower HDL levels and is found to be a better predictor of

cardiovascular risk than body mass index (BMI).[21] Although the A:G ratio can be calculated from dual-energy X-ray absorptiometry (DXA) measures[22], data quantifying VAT in children with CAH are limited, and no studies have examined VAT using DXA. There is only one study examining VAT in children with CAH using a computerized tomography scan[32]. Estimation of abdominal VAT using DXA has been validated in adults[23], and DXA has been shown to provide a reliable estimate in children as well[22].

The aims of this study were to: 1) compare BMD in children with CAH to healthy controls; 2) compare VAT and the A:G ratio in children with CAH to healthy controls; and 3) determine the relationship between BMD and VAT in children with CAH and in healthy controls.

BODY (The manuscript)

INTRODUCTION

Congenital adrenal hyperplasia (CAH) due to 21-hydroxylase deficiency is characterized by impaired cortisol synthesis leading to excessive production of adrenal androgen, which through aromatization, leads in turn to increased production of estrogen. Lifelong cortisol replacement is needed in the classic forms and in symptomatic non-classic CAH patients. In growing children, hydrocortisone (HC) is recommended at 10-15 mg/m^2/day in three divided doses to avoid the adverse impact on growth from long-acting steroids.[1] The challenge in treating CAH is to balance sufficient cortisol replacement to suppress overproduction of adrenal androgens (hyperandrogenemia), while avoiding the effects of cortisol excess (hypercortisolemia). Adding to the challenge

is HC's short median elimination half-life in CAH children, 58 min (range: 41-105 min), allowing most of the HC dose to be eliminated from the body within 4-5 hrs.[2] Because of HC's short half life, the recommended 3 times a day dosing schedule exposes children to alternating periods of hypo- and hypercortisolemia with resultant hyperandrogenemia throughout each day.

Evidence for the effects of increased adrenal sex steroids in CAH and glucocorticoid therapy on bone mineral density (BMD) are mixed, with some studies finding no difference in BMD between children with CAH and controls[3-8], others finding lower BMD in children with CAH,[9-15] and one study finding increased BMD.[16] While androgens have an established positive effect on BMD through stimulation of osteoblastic activity[17], glucocorticoids can have an opposite detrimental effect. Glucocorticoids reduce BMD through several mechanisms including decreasing osteoblastic activity, decreasing calcium absorption in the gastrointestinal tract, enhancing renal losses of calcium, and increasing bone resorption.[18]

The effects of adrenal sex steroids and glucocorticoids on measures of body composition, such as visceral adipose tissue (VAT) and the Android:Gynoid (A:G) ratio, have not been well studied in children with CAH. This area of investigation is important given that children and adults with CAH may be at an increased risk for cardiovascular disease compared to the general population.[19] VAT is associated with increased cardiometabolic risk,[20] while the A:G ratio is associated with elevated triglyceride levels and lower HDL levels and a better predictor of cardiovascular risk than BMI.[21] Although the A:G ratio can be calculated from dual-energy X-ray absorptiometry (DXA)

measures[22], data quantifying VAT in children with CAH are limited, and no studies have examined VAT using DXA. Estimation of abdominal VAT using DXA has been validated in adults[23], and DXA has been shown to provide a reliable estimate in children as well[22].

The aims of this study were to: 1) compare BMD in children with CAH to healthy controls; 2) compare VAT and the A:G ratio in children with CAH to healthy controls; and 3) determine the relationship between BMD and VAT in children with CAH and in healthy controls.

METHODS

Subjects

A retrospective cohort of 42 cases with CAH (all 21-hydroxylase deficiency; age range 7.6-17.7 years; 40% male; 19 with salt-wasting, 13 with simple-virilizing, and 10 with non-classic) were identified. Each case had molecular and biochemical testing to help confirm the diagnosis of CAH. Controls (43% male) were identified from a separate cross-sectional study evaluating cardiometabolic risk factors and vascular health in children ages 8-18 years and matched to cases based on age (±1 year), sex, and BMI Z-score (± 0.5 SD). On average, 2.4 controls (range 1-4) were matched to each case. The study was approved by the University of Minnesota Institutional Review Board.

Anthropometrics

Each case and control underwent height and weight measurements and a DXA scan as part of either clinical (cases) or research (controls) protocols. For controls, height

and weight measurements were taken three times on the day of the DXA exam on a calibrated, wall-mounted stadiometer and averaged. For cases, height and weight measurements were obtained in a similar fashion from the closest clinic visit to the DXA exam (average 9 days from the DXA exam). Height and weight Z-scores were calculated using GenenCALC 3.0 Software. BMI Z-scores were generated for all cases and controls using standard growth charts from the Center for Disease Control (CDC)[24]. Tanner staging of puberty was assessed either at the closest clinic visit to the DXA exam or on the day of the DXA exam (controls). Breast development was assessed in both female cases and controls. For males, testicular volume was measured in cases only, with a volume of 4 cc or greater being considered pubertal. Pubic hair was assessed in male cases and controls and female cases.

DXA

DXA scans for both cases and controls were performed using GE Lunar iDXA system (iDXA, General Electric Medical Systems, Madison, WI, USA). Total body bone mineral density (TBMD) Z-scores were calculated using pediatric software, and TBMD Z-scores were adjusted for height-for-age Z-scores ($TBMD_{HAZ}$)[25]. Total body composition measures (percent total tissue fat, android fat mass, and gynoid fat mass) were calculated from DXA using enCoreTM software (platform version 16.3, General Electric Medical Systems, Madison, WI, USA). Estimates of abdominal visceral adipose tissue (VAT) were obtained using methods previously described for children and adolescents.[23] Android (abdominal) fat was measured using a region-of-interest automatically defined with a caudal limit placed at the top of the iliac crest and its height

set to twenty percent of the distance from the top of the iliac crest to the base of the skull.[26] The gynoid region (hip/gluteal) is located mid-pelvis to mid-thigh, with the upper limit set below the iliac crest a distance 1.5 times the height of the android region and the lower limit set a distance of 2 times the height of the android region.[26] All scans were reviewed for accurate placement of the android box. By default, a VAT value of 2 grams or less was outside the precision of the machine and considered to be inaccurate, either because the value was below the threshold to detect VAT or the DXA machine could not accurately measure the VAT. Only one case and 5 controls had a VAT value of 2 grams or less, and these values were removed from analyses.

Glucocorticoids

Most cases with CAH were being treated with HC (79%). There were 2 cases on prednisone and 3 cases on dexamethasone who had transitioned from HC after reaching their final adult heights. Five cases with non-classic CAH were not on treatment with glucocorticoids but still had evidence of hyperandrogenemia with advanced bone age and/or growth acceleration. Prednisone and dexamethasone doses were converted into equivalent HC doses in $mg/m^2/day$ using standard glucocorticoid equivalencies (20 mg of HC = 5 mg of prednisone = 0.4 mg of dexamethasone).[27] For each case, the mean HC dose was calculated from treatment doses recorded at available clinic visits performed within 3, 6, and 12 months prior the DXA exam (mean number of clinic visits was 2.8).

Bone age

A bone age radiograph was performed only in cases with CAH, and bone age Z-scores were used as a surrogate measure of androgen exposure. The bone age radiograph

closest in time to the DXA scan (median 3 months) was interpreted using the Greulich and Pyle method. Bone age Z-scores for cases were calculated using the following formula: (bone age of the case – mean skeletal age for the case's chronological age) / (standard deviation of the skeletal age for the case's chronological age)[28]. Skeletal age means and standard deviations for girls and boys were obtained from Greulich and Pyle.

Statistical Analysis

Preliminary analyses showed that the distribution of VAT Mass was skewed to the right (i.e. had a long upper tail) so that it was more appropriate to the use the common log (log to base 10) of VAT Mass (log_{10} VAT Mass) as a dependent variable. Log_{10} VAT Mass was also used as a predictor in analyses of BMD measures.

All analyses that included both cases and controls used a mixed linear model. The random effect was a cluster consisting of a case and its 1 to 4 matched controls. Fixed effects were case versus control and other covariates of interest (e.g. VAT Mass) or adjusters. The analysis used the restricted likelihood method and adjusted averages are SAS's least-squares means.

Analyses that included cases only used one-way ANOVA or multiple linear regression and the closely associated Pearson's correlation (r).

All analyses were done using JMP (v. 12.0 Pro, SAS Institute Inc, CaryNC).

RESULTS

Table 1 shows characteristics of CAH cases and controls. The groups did not differ significantly in ethnicity, height standard deviation, weight standard deviation, or

Tanner Stage. The salt-wasting subtype accounted for 45% of cases, followed by simple-virilizing (31%), and non-classic (24%). The mean HC (or its equivalent) dose for the CAH cases receiving glucocorticoids was 11.3 mg/m2/day.

BMD

Cases had lower TBMD Z-scores compared to controls (0.81 vs. 1.27, p=0.003) (Table 2). This difference remained significant after adjusting for height-for-age Z-scores (-0.51 vs. -0.01, p=0.001). TBMD Z-scores were positively correlated with bone age Z-scores in cases with CAH (r=0.63, p<0.0001), even after adjusting for height-for-age Z-scores (r=0.51, p=0.001) (Figure 1). TBMD and $TBMD_{HAZ}$ Z-scores were not associated with average HC dose (p=0.68 and p=0.67 respectively), CAH subtype (p=0.22 and p=0.50 respectively), or age at diagnosis (p=0.18 and p=0.23 respectively).

Body Composition

VAT did not differ significantly between cases and controls (2.00 vs. 2.11, p=0.11) (Table 2). Among CAH cases, the association between VAT and bone age Z-scores trended in the negative direction (r=-0.26), but the results did not reach significance (p=0.11). VAT was not significantly associated with HC dose (p=0.98), CAH subtype (p=0.36), or age at diagnosis (p=0.15). Although cases with CAH had slightly lower percent total tissue fat (30.6% in cases vs. 32.4% in controls, p=0.052) and A:G ratios (0.405 in cases vs. 0.411 in controls, p=0.33) than controls, the differences did not reach significance. The A:G ratio was not associated with average HC dose (p=0.08), CAH subtype (p=0.29), or age at diagnosis (p=0.33).

BMD vs. VAT

In CAH cases, TBMD and $TBMD_{HAZ}$ Z-scores and VAT were negatively correlated after adjusting for BMI Z-score (for TBMD, slope= -0.66, p=0.024; for $TBMD_{HAZ,}$ slope= -0.96, p=<0.001). The correlation between $TBMD_{HAZ}$ Z-scores and VAT remained upon further adjustment for bone age Z-score (for TBMD, p=0.30; for $TBMD_{HAZ,}$ p<0.001). TBMD and $TBMD_{HAZ}$ Z-scores and VAT were not associated in controls after adjustment for BMI Z-score (for TBMD, slope= -0.005, p=0.98; for $TBMD_{HAZ,}$ slope= -0.17, p=0.38).

DISCUSSION

We found that children with CAH had lower TBMD Z-scores compared to controls. TBMD and $TBMD_{HAZ}$ Z-scores were positively correlated with bone age Z-scores but were not correlated with HC dose, CAH subtype, or age at diagnosis. While studies in the literature have assessed BMD in children with CAH, comparison between the studies is difficult due to varying age ranges (and pubertal stage), dosage and type of glucocorticoid used; inconsistently reported subtypes; and variable measures of control assessment of androgen exposure. Therefore, it is not surprising that studies have reported BMD to be reduced[9, 10, 12-15], increased[16] and or no different[3-8] in children with CAH.

Among studies that reported decreased BMD in CAH children compared to controls, two studies found that higher HC dose (or its equivalents) was associated with lower BMD Z-scores,[12, 14] whereas Demirel et al[15] and our study found no association. The rest of the studies did not examine this relationship.[9, 10, 13] Most children in the two

studies that found a negative relationship were on long-acting glucocorticoids. The finding of no relationship between total daily HC dose and decreased BMD suggests that other factors such as dose distribution, individual cortisol pharmacokinetics, glucocorticoid receptor sensitivity and type of glucocorticoid, all of which determine cortisol and androgen exposures over the course of the day, may play a role in the impact of glucocorticoids on BMD. For example, in our study, patients were treated with HC within the recommended range, but this did not prevent the presumed adverse effects of glucocorticoids on BMD as potentially some patients were more glucocorticoid sensitive or had decreased cortisol clearance and/or increased half-life, and therefore had higher biologically active glucocorticoid available at the tissue level.

Among studies that reported decreased BMD, the relationship between advanced bone age and BMD Z-scores was examined only in de Almeida Freire et al, who found, as we did, that in CAH cases advanced bone age was associated with higher BMD Z-scores.[10] This finding is not surprising as higher bone age Z-scores indicate greater androgen exposure which can lead to an increase in BMD through androgen stimulation of osteoblastic activity.[17] Aromatization of elevated adrenal androgen to estrogen may also have some protective effects on bone. Estrogen reduces the release of inflammatory cytokines such as IL-6, IL-1, and TNF-alpha from osteoblasts and decreases osteoclastic activity by suppressing RANK-L-induced osteoclast differentiation.[29] In addition, estrogen can increase osteoprotegerin production[30] and prevent glucocorticoid-induced apoptosis in osteoblasts.[31]

Several studies have found no difference in BMD Z-scores between children with

CAH and controls.[3-8, 11] In some of these studies, national reference data were used instead of age-, sex-, and BMI-matched controls;[3, 4, 7, 11] BMD was not adjusted for height-for-age Z-score,[4-8] which could lead to under- or overestimation of BMD Z-score[25]; the CAH subtype[3, 4] or the type of glucocorticoid was not always specified[4, 7]; and in one study, only females with CAH were evaluated.[8] Three of these studies found no association between HC dose and BMD Z-scores[4, 8, 11], and the rest did not examine the relationship.[3, 5-7]

Only one study found increased BMD in children with CAH[16], but that study differed from ours in that it included not only children with 21-hydroxylase deficiency but also 11β-hydroxylase deficiency; did not use age-, sex-, and BMI-matched controls; did not adjust BMD Z-scores for height-for-age Z-scores; did not examine measures of androgen control; did not specify the type of glucocorticoid used; and did not specify the pubertal status of the patients.

Our study was the first to use DXA to calculate VAT in children with CAH and found no difference in VAT between cases and controls. Only one previous study, by Kim et al, quantified VAT in children with CAH using a computed tomography scan and found increased VAT in 28 children with CAH.[32] Several factors other than the methodology may account for this difference. One of them is the dose of HC used, which was higher in the study by Kim et al., 19.5 (SD 5.4) mg/m^2/day vs. 11.3 (SD 2.9) mg/m2/day in our study. In addition, CAH cases in Kim et al. were shorter (height Z-score: -0.96 vs. 0.67) and more overweight/obese (BMI Z-score: 1.31 vs 0.95), indicating higher glucocorticoid exposure. High doses of glucocorticoids increase VAT by

promoting the differentiation of pre-adipocytes into central fat[33] and by decreasing adrenal androgen production though suppression of the hypothalamic-pituitary adrenal axis. Androgen, mostly through aromatization to estrogen, increases lipolysis, helps prevent the differentiation of pre-adipocytes into adipocytes, and, in VAT specifically, inhibits lipoprotein lipase (which promotes fat accumulation).[34] While we did not find a significant negative association between bone age Z-scores and VAT, the association trended in this direction, which may imply the role of increased adrenal sex steroids in decreasing VAT in children with CAH.

We found a negative correlation between VAT and TBMD and $TBMD_{HAZ}$ Z-scores in the CAH cases, but did not find this association in controls. Adipose tissue secretes various inflammatory cytokines, such as TNF-a and IL-6, which activate osteoclast differentiation and activation and inhibit osteoclast apoptosis.[35] Altered hypothalamic-pituitary adrenal axis and chronic fluctuations of cortisol and adrenal sex steroids may affect fat distribution and may increase secretion of inflammatory cytokines from VAT, leading to the inverse relationship between VAT and bone mineral density.[35]

The strengths of our study were the use of age-, sex-, and BMI-matched controls; adjustment of BMD Z-scores for height-for-age Z-scores given that CAH patients tend to be taller than their peers during childhood; having most of our subjects on HC; and use of bone age Z-scores to assess chronic androgen exposure rather than single measures of steroid concentrations which reflect control only at the time of measurement. Limitations of our study include its retrospective design as well as not having the testicular exam or bone age performed in controls.

In conclusion, we found that CAH cases had lower TBMD than controls and that VAT and BMD were negatively associated in CAH cases. Bone age Z-scores and BMD were positively correlated, displaying the positive effects of androgen exposure in increasing BMD. Future longitudinal studies in children with CAH to examine the impact of chronic hormonal imbalances on BMD and VAT are needed, including duration of over- and under-cortisol exposure as well as androgen exposure over the course of the day.

FUTURE DIRECTIONS

Our study provided preliminary evidence that children with CAH have decreased BMD even when the total daily dose of hydrocortisone is within the recommended range. Future longitudinal studies in children with CAH are needed to examine the impact of chronic hormonal imbalances on BMD and VAT and how the duration of over- and under-cortisol exposure as well as androgen exposure over the course of the day and over several years impacts BMD and body composition in children with CAH.

Since there is an increased risk for fractures in adults and children with CAH[36] and the majority of bone mineral density is accrued during childhood, it is important to examine how bone mineral density changes in these children overtime. Similarly, VAT, as a marker of cardiovascular disease[20] is important to monitor overtime, since children and adults with CAH are at an increased risk for cardiovascular disease[19]. Our longitudinal study will address existing gaps in knowledge and has the potential to alter our fundamental approach to managing chronic glucocorticoid treatment and

establish a new standard of care in children with CAH. We will examine yearly BMD via dual-energy X-ray absorptiometry (DXA) scan and determine whether BMD and VAT changes overtime in both children with CAH and sibling controls. By having sibling controls, we can account for genetic differences in bone mineral density and potential genetic causes of increased VAT accumulation between patients with CAH. Bone age Z-score will be used as a surrogate marker of overall androgen exposure and bone age will be measured in both cases with CAH and sibling controls. In addition, measuring pubertal status in both cases with CAH and sibling controls will allow us to account for pubertal changes affecting BMD and VAT. Our models will also adjust for age and sex to account for factors which affect BMD and VAT. By monitoring these measures longitudinally, we may be able to intervene earlier in life, thus reducing the negative sequelae of impaired bone mineral density and cardiovascular disease in a population that is already at an increased risk.

Table 1. Characteristics of CAH Cases and Controls

Characteristic	**Cases**	**Controls**	**SE difference**	**P-value**
N	42	101		
Sex, Male, n (%)	17 (40%)	43 (43%)		
Age, mean (SE)	12.34 (0.45)	12.48 (0.44)	0.08	0.09
Ethnicity, n (%)				0.71
White Non-Hispanic	36 (86%)	81 (80%)		
Other	6 (14%)	20 (20%)		
Height SD, mean (SE)	0.67 (0.18)	0.38 (0.12)	0.21	0.16
Weight SD, mean (SE)	1.07 (0.13)	1.08 (0.12)	0.09	0.91
BMI Z-score, mean (SE)	0.95 (0.13)	1.02 (0.13)	0.04	0.09
Tanner stage, n (%)				0.76
Testicles (male)				
Tanner 1	10 (59%)	NA		
Tanner 2-3	3 (18%)	NA		
Tanner 4-5	4 (24%)	NA		
Pubic hair (male)				
Tanner 1	4 (24%)	13 (31%)		
Tanner 2-3	7 (41%)	13 (31%)		
Tanner 4-5	6 (35%)	16 (38%)		
Breast (female)				
Tanner 1	7 (28%)	10 (18%)		
Tanner 2-3	9 (36%)	23 (40%)		
Tanner 4-5	9 (36%)	24 (42%)		
Pubic hair (female)				
Tanner 1	4 (17%)	NA		
Tanner 2-3	8 (33%)	NA		
Tanner 4-5	12 (50%)	NA		
CAH subtype, n (%)		N/A		
Salt-wasting	19 (45%)			
Simple-virilizing	13 (31%)			
Non-classic	10 (24%)			
Hydrocortisone, average dose, mg/m^2/day	11.3 (2.9)	N/A		
Bone age Z-score	1.34 (2.02)	NA		

BMI, body mass index; NA, not available; N/A, not applicable; CAH, congenital adrenal hyperplasia.

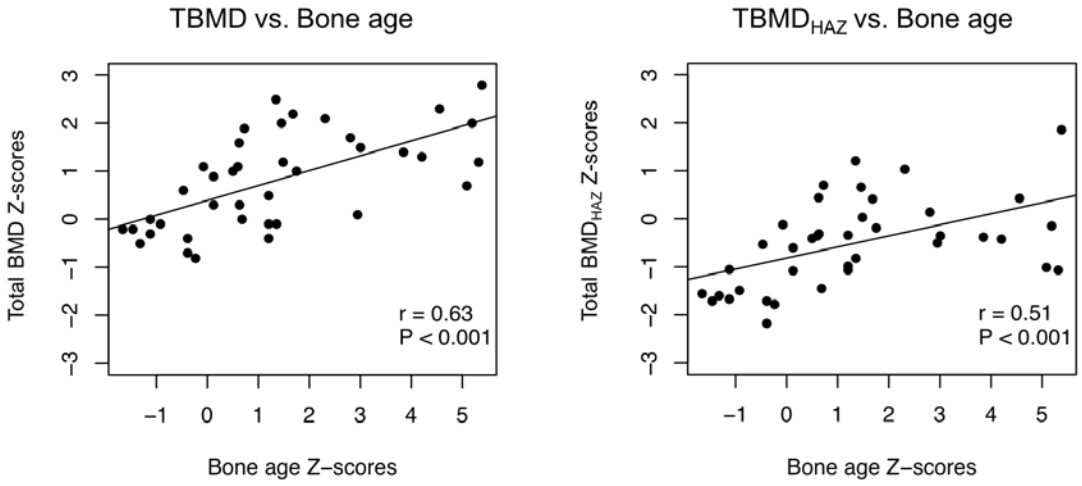
Table 2. CAH Cases and Controls compared according to BMD and Body Composition Measures

BMD or Body Composition Measures	**Cases**	**Controls**	**Control - Case**	**P-value**
TBMD Z-scores	0.81 (0.14)	1.27 (0.11)	0.47 (0.15)	0.003*
$TBMD_{HAZ}$ Z-scores	-0.51 (0.14)	-0.01 (0.10)	0.49 (0.15)	0.001*
logVAT Mass	2.00 (0.08)	2.11 (0.07)	0.11 (0.07)	0.11
A:G ratio	0.405 (0.01)	0.411 (0.01)	0.006	0.33
Percent Total Tissue Fat	30.6 (1.5)	32.4 (1.4)	0.94	0.052

Written as adjusted average (standard error); CAH, congenital adrenal hyperplasia; BMD, bone mineral density; TBMD, total body bone mineral density; $TBMD_{HAZ}$, total body bone mineral density Z-scores adjusted for height-for-age Z-scores; VAT, visceral adipose tissue; A:G, Android:Gynoid ratio

*Indicates a significant value

Figure 1.


TBMD vs. Bone age
Total BMD Z-scores
Bone age Z−scores
r = 0.63
P < 0.001
TBMD$_{HAZ}$ vs. Bone age
Total BMD$_{HAZ}$ Z-scores
Bone age Z−scores
r = 0.51
P < 0.001

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