Body Fat Mass Is Associated With Ratio of Steroid Metabolites Reflecting 17,20-Lyase Activity in Prepubertal Girls

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Context: Pediatric obesity has been related to hyperandrogenism and premature adrenarche in previous studies. However, little is known regarding the association between body fat mass and steroidogenic enzyme activities in children.

Objective: To examine whether body fat mass is associated with serum steroid profiles in girls.

Design, Participants, and Setting: We enrolled 242 girls (125 prepubertal, 117 pubertal; age, 7–13 years). Early morning blood samples were drawn at a university hospital to measure serum steroid profiles using gas chromatography-mass spectrometry, and steroidogenic enzyme activities were assessed from the ratios of steroid metabolites.

Main Outcome Measures: We evaluated serum steroid profiles and estimated steroidogenic enzyme activities and their association with anthropometric indices and body composition.

Results: Prepubertal obese girls demonstrated significantly higher progestin, androgens (dehydroepiandrosterone [DHEA], androstenedione [A-dione], T, androsterone), and ratio of steroid metabolites reflecting 17,20-lyase activity [(DHEA + A-dione)/17-hydroxypregnenolone] compared with prepubertal controls. Pubertal obese girls demonstrated significantly higher serum T and androsterone than pubertal controls; however, serum steroid metabolite ratios reflecting steroidogenic enzyme activities did not significantly differ among obese and non-obese girls. Partial correlation analysis revealed that body fat mass was positively correlated with pregnenolone, DHEA, A-dione, T, androsterone, and ratio of (DHEA + A-dione)/17-hydroxypregnenolone in prepubertal girls only. Prepubertal girls with increased body fat mass had significantly higher serum DHEA and ratio of (DHEA + A-dione)/17-hydroxypregnenolone than controls.

Conclusions: Increased androgen production in prepubertal obese girls could be at least partly due to increased body fat mass and 17,20-lyase activity. (***J Clin Endocrinol Metab*** **101: 4653–4660, 2016)**

Adrenarche is defined as the onset of increasing adrenal secretion of 19 carbon steroids—principally dehydroepiandrosterone (DHEA) and its sulfate ester, which usually begins after the age of 6 years (1). This phenomenon also corresponds to the expansion of the adrenal gland zona reticularis and changes in steroidogenic enzyme activities. For instance, the expression of CYP17, an enzyme involved in the production of DHEA, increases during adrenarche (2). In addition, decreased expression of 3β-hydroxysteroid dehydrogenase (3β-HSD) is known to facilitate DHEA synthesis by reducing conversion of DHEA precursors to mineralocorticoids or glucocorticoids (3).

ISSN Print 0021-972X ISSN Online 1945-7197
Printed in USA

Received June 29, 2016. Accepted September 16, 2016.
First Published Online September 20, 2016

* M.-J.P. and M.H.C. share the senior authorship of this work.

Abbreviations: A-dione, androstenedione; BMI, body mass index; DHEA, dehydroepiandrosterone; DHEAS, DHEA sulfate; GC-MS, gas chromatography-mass spectrometry; HSD, hydroxysteroid dehydrogenase; 17-OH-Preg, 17-hydroxypregnenolone; PA, premature adrenarche.

Premature adrenarche (PA) refers to the appearance of pubic or axillary hair in girls younger than 8 years old and boys younger than 9 years old as a result of the rise in adrenal androgen production, without other diseases of the gonads or adrenal glands (4, 5). The prevalence of precocious puberty has recently increased among Korean children (6), and the number of Korean girls visiting hospitals for PA evaluations has also been on the rise. An increasing trend of PA has also been noted in Danish children between 1993 and 2008 (7). Moreover, childhood or pediatric obesity has been associated with increased androgen production and increased risk of PA (8, 9). Several studies have demonstrated that girls with PA tend to be obese and have excess body fat (10–12). In addition, in the general population, serum concentrations of DHEA sulfate (DHEAS) were reported to be higher in obese prepubertal children (13–16) and possibly in obese female adolescents (17). However, in vivo metabolic evidence has yet to be elucidated regarding the contribution of obesity to steroidogenic enzyme activation in children. In addition, only a few studies have investigated the association between androgen production and body composition (instead of body mass index [BMI]) (14, 18).

In this study, we aimed to examine whether body fat mass is associated with altered steroid profiles and estimated steroidogenic enzyme activities in prepubertal and pubertal girls using gas chromatography-mass spectrometry (GC-MS)-based steroid signatures in serum samples.

Subjects and Methods

Study subjects

A total of 101 overweight or obese girls and 141 age-matched controls aged 7 to 13 years were recruited from the Inje University Hospital (Seoul, Korea) between 2013 and 2014. For controls, non-overweight girls who visited the health clinic center for regular growth and development check-ups were enrolled. All participants were clinically evaluated through physical examination and careful evaluation of past histories to exclude possible medication use, chronic diseases, and endocrinopathies. Individuals with a history of small size for gestational age, premature pubarche, abnormal thyroid functions, or impaired glucose tolerance were also excluded. The study protocol was approved by the Institutional Review Board of Inje University Hospital (IRB no. SPIRB13–094), and written informed consent was obtained from all subjects or from their parents, in compliance with the Declaration of Helsinki principles.

Anthropometric measurements

While the subjects were wearing light clothes without shoes, their height and weight were measured by a stadiometer (Dongsahn Jenix Co) and Inbody 720 (Biospace Co. Ltd), respectively. BMI (weight in kilograms/height in meters^2) was then calculated. Body composition was measured through bioelectrical impedance analysis using the Inbody 720. Overweightness and obesity were categorized according to the age- and sex-specific percentiles for BMI of national reference standards (19). Overweight was defined as BMI $\geq$ 85th percentile and $<$ 95th percentile, and obesity was defined as BMI $\geq$ 95th percentile. The pubertal stages for breast development were determined by a pediatric endocrinologist according to the Marshall and Tanner method (20).

Sample collection and analysis

Blood samples were collected at 8 AM after a 10-hour overnight fast. All samples were stored at −80°C until required. Steroid profiling was performed to determine plasma levels of pregnenolone, 17-hydroxypregnenolone (17-OH-Preg), DHEA, androstenedione (A-dione), T, and androsterone. The quantitative profiling of serum steroids was based on a previous report (21). In brief, serum samples (0.2 mL) were spiked with 15 μL of five internal standards (d_3-testosterone and d_4-estradiol, 0.2 μg/mL; d_4-cortisol and d_8-17α-hydroxyprogesterone, 1 μg/mL; and d_9-progesterone, 2 μg/mL) and diluted with 2.8 mL of 0.2 M acetate buffer (pH 5.2) and 100 μL of aqueous 0.2% ascorbic acid. The diluted sample was loaded onto an Oasis HLB cartridge, which was then washed with 2 mL of deionized water and eluted twice with 2 mL of methanol. The methanol eluate was evaporated and then extracted twice with 1 mL of 0.2 M phosphate buffer (pH 7.2), 100 μL of aqueous 0.2% ascorbic acid, and 2.5 mL of ethyl acetate:*n*-hexane (2:3, v/v). The isolated organic solvents were evaporated by nitrogen stream at 40°C and further dried in a vacuum desiccator over P_2O_5-KOH for at least 30 minutes. Finally, the dried residue was derivatized with MSTFA/NH_4I/DTE (30 μL; 500:4:2, v/w/w) at 60°C for 20 minutes, and 2 μL of the resulting mixture was injected to GC-MS in the selected-ion monitoring mode.

Estimation of steroidogenic enzyme activities

The steroidogenic enzyme activities including 17α-hydroxylase, 17,20-lyase, 3β-HSD, and 17β-HSD) were estimated by product/precursor ratios of specific serum steroid metabolites: (17-OH-Preg + DHEA)/pregnenolone, (DHEA + A-dione)/17-OH-Preg, (T + A-dione)/DHEA, and T/A-dione, respectively (Figure 1).

Statistical analysis

The data are expressed as means ± standard deviations (continuous), or number (percentage) (categorical). Differences for anthropometric characteristics and Tanner stages for breast development among obesity groups were tested using ANOVA or Fisher's exact tests, as appropriate. *P*-for-trends of serum steroid hormone levels and serum steroid metabolite ratios reflecting steroidogenic enzyme activities according to the obesity status were obtained in linear regression, modeling the respective variables as continuous variables. Adjusted correlation coefficients between steroid hormone levels and anthropometric data were calculated using partial correlation analysis. Furthermore, differences of estimated marginal means (95% confidence interval) of steroid hormone levels and ratios of serum steroid metabolites reflecting 17,20-lyase activity by body fat mass status in prepubertal girls were obtained using analysis of covariance. All statistical tests were performed using SPSS (version 22.0; SPSS Inc.), and a *P* value $<$.05 was considered significant.

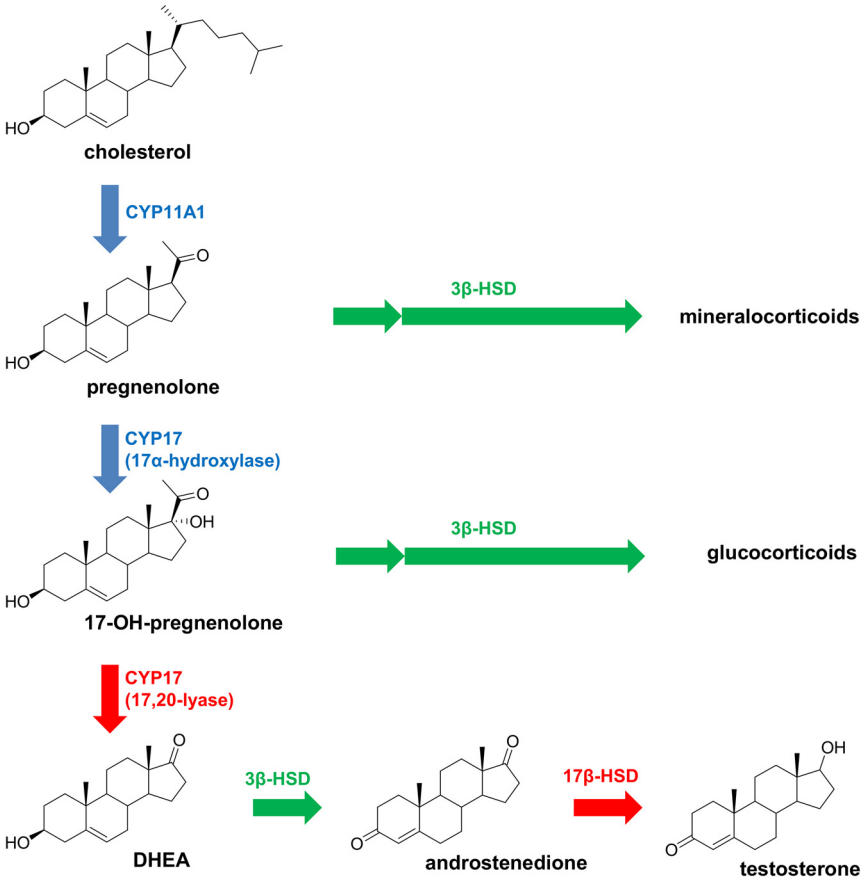

Figure 1. Steroid pathways for biosynthesis of adrenocortical steroids. CYP11A1, cytochrome P450 cholesterol side-chain cleavage; CYP17, 17α-hydroxylase/17,20-lyase; 17β-HSD, 17β-hydroxysteroid dehydrogenase; 3β-HSD, 3β-hydroxysteroid dehydrogenase.

Results

Anthropometric characteristics of the study population are summarized in Table 1. In prepubertal girls, obese girls were taller than control and overweight girls, whereas this was not the case in pubertal girls. Body fat percentage and mass were both significantly higher in proportion to the degree of obesity in prepubertal and pubertal girls.

Steroid hormone serum concentrations and estimated steroidogenic enzyme activity relative to obesity and pubertal status are also summarized in Table 2. In prepubertal girls, concentrations of progestins (progesterone, 17-OH-Preg) and androgens (DHEA, A-dione, T, androsterone) significantly increased with obesity (*P*-for-trend <.05). In pubertal girls, T and androsterone levels also significantly increased with obesity (*P*-for-trend <.05), but no significant differences were detected in concentrations of progestins and other androgens. The ratio of (DHEA + A-dione)/17-OH-Preg reflecting 17,20-lyase activity was significantly higher in prepubertal obese girls than control or overweight girls, whereas this difference relative to obesity was not detected in pubertal girls. No significant differences were detected in serum steroid metabolite ratios reflecting activities of 17α-hydroxylase, 3β-HSD, and 17β-HSD relative to obesity status.

Age and pubertal stage-adjusted correlation coefficients between steroid hormone levels and anthropometric data are summarized in Supplemental Table 1. DHEA, A-dione, and T concentrations were positively correlated

Table 1. General Characteristics of Subjects by Obesity Status

	Prepubertal Girls (n = 125)				Pubertal Girls (n = 117)			
	Control	Overweight	Obese	*P* Value	Control	Overweight	Obese	*P* Value
n	68	24	33		73	23	21	
Age, y	8.5 ± 0.8	8.3 ± 0.8	8.4 ± 0.9	.377	9.5 ± 1.4	9.3 ± 1.5	9.3 ± 1.8	.725
Height, cm	131.4 ± 5.8	130.0 ± 6.9	134.5 ± 6.5	.011	138.6 ± 7.7	139.1 ± 6.9	139.5 ± 11.5	.914
Weight, kg	28.9 ± 4.1	33.0 ± 4.9	41.3 ± 5.9	<.0001	33.2 ± 5.8	40.1 ± 9.4	45.9 ± 12.3	<.0001
BMI, kg/m^2	16.8 ± 1.5	19.4 ± 1.3	22.5 ± 2.1	<.0001	17.1 ± 1.6	20.5 ± 1.8	23.1 ± 2.5	<.0001
BMI percentile	52.0 ± 23.5	87.4 ± 12.0	97.8 ± 1.5	<.0001	48.4 ± 23.4	87.7 ± 10.5	97.7 ± 1.6	<.0001
Body fat %	22.5 ± 6.6	29.0 ± 4.7	35.7 ± 6.7	<.0001	22.3 ± 5.4	29.3 ± 8.7	35.0 ± 4.9	<.0001
Body fat mass, kg	6.8 ± 2.7	9.7 ± 2.5	14.9 ± 4.3	<.0001	7.7 ± 2.8	12.1 ± 5.8	16.3 ± 5.6	<.0001
Tanner stage[a]				.999				.711
1	68 (100)	24 (100)	33 (100)					
2					36 (49.3)	11 (47.8)	10 (47.6)	
3					24 (32.9)	7 (30.4)	8 (38.1)	
4					11 (15.1)	3 (13.0)	1 (4.8)	
5					2 (2.7)	2 (8.7)	2 (9.5)	

Data are expressed as means ± SD or number (percentage). *P* values were derived by ANOVA or Fisher's exact test.

[a] Tanner stage of breast development.

Table 2. Serum Steroid Hormones and Activities of Steroidogenic Enzymes by Obesity and Puberty Status

	Prepubertal Girls (n = 125)				Pubertal Girls (n = 117)			
	Control	Overweight	Obese	*P*-for-trend	Control	Overweight	Obese	*P*-for-trend
n	68	24	33		73	23	21	
Pregnenolone, ng/mL	1.35 ± 0.81	1.35 ± 0.87	2.04 ± 0.97	<.0001	1.65 ± 0.95	1.55 ± 0.70	2.07 ± 0.73	.102
17-OH-Preg, ng/mL	1.69 ± 1.06	2.03 ± 1.60	2.44 ± 1.52	.029	2.10 ± 1.67	2.15 ± 1.71	2.46 ± 1.87	.691
DHEA, ng/mL	1.80 ± 0.96	1.83 ± 1.11	3.15 ± 1.59	<.0001	2.70 ± 1.42	2.67 ± 1.31	3.23 ± 1.24	.263
A-dione, ng/mL	0.98 ± 0.60	1.00 ± 0.72	1.62 ± 0.72	<.0001	1.52 ± 0.80	1.43 ± 0.83	1.90 ± 0.73	.103
T, ng/mL	0.83 ± 0.45	0.79 ± 0.51	1.31 ± 0.52	<.0001	1.08 ± 0.52	0.99 ± 0.51	1.43 ± 0.47	.011
Androsterone, ng/mL	0.74 ± 0.34	0.90 ± 0.47	1.02 ± 0.42	.004	0.95 ± 0.33	1.13 ± 0.44	1.15 ± 0.37	.011
(17-OH-Preg + DHEA)/ Pregnenolone[a]	2.9 ± 1.5	3.1 ± 1.4	3.0 ± 1.3	.759	3.4 ± 2.0	3.4 ± 1.9	3.0 ± 1.5	.496
(DHEA + A-dione)/ 17-OH-Preg[a]	1.8 ± 0.6	1.7 ± 0.9	2.4 ± 1.4	.004	2.4 ± 0.9	2.3 ± 0.9	2.5 ± 1.0	.729
(T + A-dione)/DHEA[a]	1.1 ± 0.4	1.0 ± 0.3	1.0 ± 0.3	.361	1.0 ± 0.4	1.0 ± 0.4	1.1 ± 0.3	.753
T/A-dione[a]	0.9 ± 0.2	0.9 ± 0.3	0.9 ± 0.4	.992	0.8 ± 0.2	0.8 ± 0.4	0.8 ± 0.3	.466

P-for-trend was obtained in linear regression, modeling the respective variables as continuous variables.

[a] Product-to-precursor ratios of serum steroid metabolites were used to estimate the activities of steroidogenic enzymes including 17α-hydroxylase [(17-OH-Preg + DHEA)/pregnenolone], 17,20-lyase [(DHEA + A-dione)/17-OH-Preg], 3β-HSD [(T + A-dione)/DHEA], and 17β-HSD [T/A-dione].

with height percentile, body fat mass, and body muscle mass. Androsterone and pregnenolone levels demonstrated positive correlations with height percentile and body fat mass, respectively. After the adjustment for height percentile, pregnenolone, DHEA, A-dione, T, and androsterone levels all maintained significant correlations with body fat mass alone. The correlations between body fat mass and steroid hormone levels/serum steroid metabolite ratios by puberty status are summarized in Table 3. In prepubertal girls, body fat mass was positively correlated with serum concentrations of pregnenolone, DHEA, A-dione, T, and androsterone. There was a positive association between the ratio of (DHEA + A-dione)/17-OH-Preg and body fat mass in prepubertal girls (Figure 2); however, there was no significant difference in pubertal girls.

Adjusted mean values of serum DHEA and the ratio of (DHEA+A-dione)/17-OH-Preg by body fat mass status in prepubertal girls are summarized in Supplemental Figure 1. After the adjustment for age, height percentile, BMI percentile, and all other steroid metabolite concentrations, prepubertal girls with body fat mass > the 90th percentile value had significantly higher serum DHEA concentrations and ratios of (DHEA + A-dione)/17-OH-Preg than prepubertal girls with body fat mass ≤ the 90th percentile value. Other steroid hormones including pregnenolone, A-dione, T, and androsterone concentrations were comparable between these two groups (Table 4).

Discussion

In this study, we demonstrated that body fat mass in prepubertal girls was positively correlated with major

Table 3. Correlations Between Body Fat Mass and Steroid Levels (ng/mL)/Steroidogenic Enzyme Activities

	Prepubertal Girls		Pubertal Girls	
	β	*P* Value	β	*P* Value
Pregnenolone, ng/mL	**0.279**	**.003**	0.090	.240
17-OH-Preg, ng/mL	0.154	.098	−0.037	.800
DHEA, ng/mL	**0.322**	**<.0001**	0.016	.714
A-dione, ng/mL	**0.306**	**.001**	0.053	.468
T, ng/mL	**0.273**	**.003**	0.111	.158
Androsterone, ng/mL	**0.232**	**.012**	0.069	.216
(17-OH-Preg + DHEA)/pregnenolone[a]	−0.098	.297	−0.073	.467
(DHEA + A-dione)/17-OH-Preg[a]	**0.206**	**.027**	0.030	.762
(T + A-dione)/DHEA[a]	−0.049	.600	0.056	.578
T/A-dione[a]	−0.029	.756	0.105	.296

Correlation coefficients (β), adjusted for age, Tanner stage of breast development, and height percentile were calculated using partial correlation analysis. Values that were statistically significant at $P < .05$ are indicated in bold.

[a] Product-to-precursor ratios of serum steroid metabolites were used to estimate the activities of steroidogenic enzymes including 17α-hydroxylase [(17-OH-Preg + DHEA)/pregnenolone], 17,20-lyase [(DHEA + A-dione)/17-OH-Preg], 3β-HSD [(T + A-dione)/DHEA], and 17β-HSD [T/A-dione].

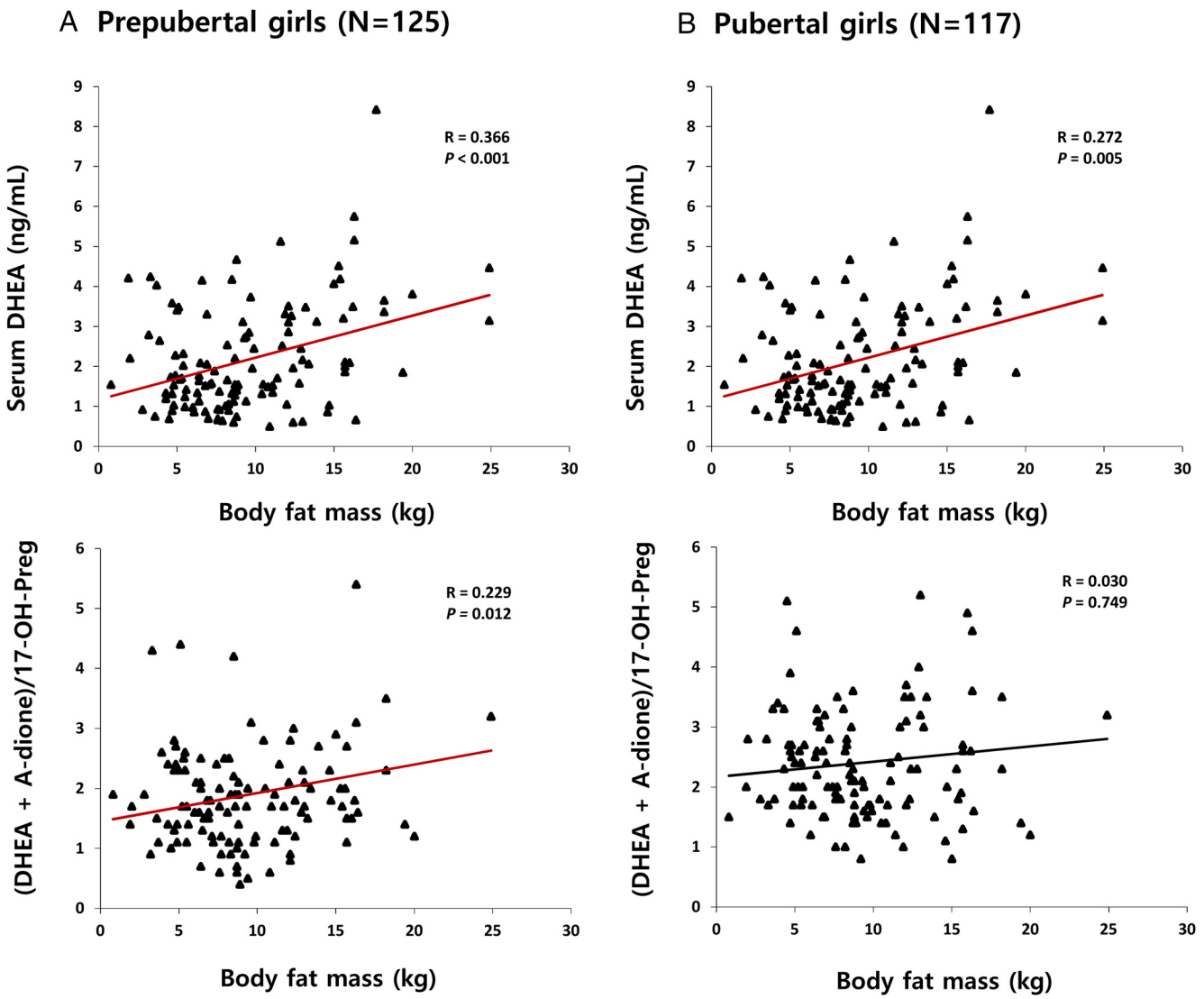

Figure 2. The correlation between body fat mass (kilograms) and serum DHEA levels (nanograms/milliliter)/ratios of (DHEA + A-dione)/17-OH-Preg reflecting 17,20-lyase activity among prepubertal (A) and pubertal (B) girls. A-dione, androstenedione; DHEA, dehydroepiandrosterone; 17-OH-Preg, 17-hydroxypregnenolone.

serum androgen levels and the ratio of serum steroid metabolites reflecting activity of 17,20-lyase, a key regulatory steroidogenic enzyme in producing DHEA from its precursor, 17-OH-Preg. Our present study is also the first in vivo study evaluating the correlation between estimated 17,20-lyase activity and increased androgen production in prepubertal obese girls, to the best of our knowledge.

Recent studies have demonstrated a significant association between concurrent obesity indicators including body fat index with serum DHEAS in prepubertal children (13). However, it is also true that no significant correlation was reported between DHEAS excretion and concurrent BMI/body fat mass (12), which is probably due to the fact that they did not adjust the findings for height and pubertal stage. Results of several studies reported that androgen levels were significantly associated with height and pubertal stage in children (14, 20, 22). In addition, body fat mass itself was also markedly associated with height and/or pubertal stage. Therefore, when determining the correlation between body fat mass and androgen production, it is considered pivotal to exclude the effects of height and pubertal stage on the aforementioned covariates.

The evidence regarding the association between adrenal steroidogenic enzyme activity and childhood obesity is very limited at this juncture. Steroidogenic enzyme activities could be accurately determined by measuring mRNA expression of corresponding enzymes in specimens of the steroidogenic organs. However, in clinical settings in which invasive biopsy from these organs is not always feasible, steroidogenic enzyme activities could be estimated from the ratios of circulating or urinary steroid metabolites (23, 24). In particular, mass spectrometry-based steroid profiling, which could measure multiple

Table 4. Estimated Marginal Means (95% Confidence Interval) of Steroids and Ratio of (DHEA + A-dione)/17-OH-Preg by Body Fat Mass Status Among Prepubertal Girls

	Body Fat Mass ≤90th Percentile	Body Fat Mass >90th Percentile	*P* Value
n	113	12	
Pregnenolone, ng/mL[a]	1.55 (1.48–1.62)	1.36 (1.11–1.61)	.163
DHEA, ng/mL[a]	**2.09 (1.95–2.23)**	**2.96 (2.45–3.42)**	**.001**
A-dione, ng/mL[a]	1.15 (1.08–1.21)	1.10 (0.87–1.33)	.716
T, ng/mL[a]	0.94 (0.91–0.98)	1.00 (0.87–1.12)	.398
Androsterone, ng/mL[a]	0.86 (0.80–0.92)	0.66 (0.44–0.87)	.081
(DHEA + A-dione)/17-OH-Preg[b]	**1.79 (1.62–1.97)**	**2.87 (2.31–3.43)**	**.001**

P values were derived by analysis of covariance. Ratio of (DHEA + A-dione)/17-OH-Preg reflects 17,20 lyase activity. Values that were statistically significant at $P < .05$ are indicated in bold.

[a] Adjusted for age, height percentile, BMI percentile, and all other steroid hormone levels in the table.

[b] Adjusted for age, height percentile, and BMI percentile.

analytes at the sample time with high sensitivity and specificity, has been widely used to determine enzyme activities estimated by the product-to-precursor ratios (23, 25). Recently, steroidogenic enzyme activities were evaluated by the ratios of urinary steroids in prepubertal children (18). Results demonstrated that increased androgen production was inversely associated with 3β-HSD and 21-hydroxylase activities independent of body fat (18). However, that particular study by no means evaluated the impact of body fat on specific steroidogenic enzyme activities. Therefore, in this study, we first demonstrated that height percentile, body fat mass, and body muscle mass were all associated with serum androgen levels independent of age and pubertal stage, and only body fat mass was statistically significant after adjusting for height percentile. Prepubertal girls with increased body fat mass (>90th percentile) had 1.4 times higher serum DHEAS concentrations and 1.6 times higher estimated 17,20-lyase activity than prepubertal girls with body fat mass ≤ 90th percentile, even after adjusting for BMI percentile.

Increased serum insulin and IGF levels have generally been considered pivotal features of obese children with premature adrenarche, with these hormonal factors being implicated in regulating androgen production. Previous in vitro studies demonstrated that treatment with IGF-1, IGF-2, and insulin resulted in higher expression of mRNA for CYP17 (26, 27) and/or 3β-HSD (27) in human adrenocortical cells. However, results of in vivo studies were not necessarily consistent. A few small sample-sized studies reported that androgen levels were significantly associated with insulin and IGF-1 levels (28, 29). However, two other studies have failed to demonstrate the possible association of insulin and IGF-1 with adrenarche in prepubertal children (30) and with androgen production in pubertal boys (29). Therefore, the significance of insulin and IGF-1 in the regulation of adrenal androgen production remains unclear, and further investigations are required for clarification.

This correlation of increased androgen production with body fat mass in children did indicate the possible roles of adipokines produced by adipocyte tissue. Previous studies on this topic have been generally focused on the effect of leptin, one of the major adipokines. Serum leptin levels were reported to be correlated with androgens in children with premature adrenarche (31, 32). This possible correlation between leptin and androgen was also supported by results of an in vitro study, which demonstrated that leptin treatment stimulated the 17,20-lyase activity of the CYP17 enzyme in one human adrenal cell line without significant concomitant effects upon 17-α hydroxylase activity (33). Of particular interest in our present study is the finding that body fat mass was significantly correlated with estimated 17,20-lyase activity and subsequent DHEA increment in prepubertal girls. Body fat mass has been reported to be closely related to systemic leptin levels in prepubertal children (34), and therefore we speculate that leptin produced by adipose tissue might mediate increased steroidogenic enzyme activities in obese children. However, in vivo evidence regarding metabolic signals from adipose tissue and steroidogenic enzyme activities should be explored for clarification in further studies.

In this study, the correlation between body fat and estimated 17,20-lyase activity was detected only in prepubertal girls, whose steroidogenic enzyme activities were considered to be derived mainly from adrenal glands. In pubertal girls, a considerable proportion of androgens are produced from ovarian cells under the stimulation of gonadotropin (35). Insulin (28, 36) and IGF-1 (28, 32) have both been reported to be involved in ovarian hyperandrogenism in pubertal girls. Therefore, it is reasonable to postulate that fat mass and adipokines could reflect early adrenarche in prepubertal girls, whereas other hormonal factors could be related to ovarian hyperandrogenism in pubertal girls.

It is important to note some limitations in this study. First, this was a cross-sectional study, and temporal se-

quence could not necessarily be inferred. Second, we did not validate adipokines released from body fat tissue. However, this is the first study that attempted to correlate body composition with overall body steroidogenic enzyme activity using GC-MS-based steroid signatures in prepubertal and pubertal girls.

In conclusion, body fat could increase androgen production and 17,20-lyase activity of adrenal glands in obese prepubertal girls. Further studies are required to elucidate possible molecular mechanisms involved in 17,20-lyase activation relating to body fat mass.

Acknowledgments

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This work was supported by the 2013 Korean Society of Pediatric Endocrinology research grant. It was partially supported by the Bio & Medical Technology Development Programs (NRF-2013M3A9B6046413) through the Ministry of Science, ICT & Future Planning.

Disclosure Summary: All of the authors have nothing to disclose.

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