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**BODY COMPOSITION, RESTING ENERGY EXPENDITURE,
AND PUBERTAL MATURATION IN AFRICAN AMERICAN AND
CAUCASIAN CHILDREN AND ADOLESCENTS**

by

MIN SUN

A DISSERTATION

**Submitted to the graduate faculty of the University of Alabama at Birmingham,
in partial fulfillment of the requirements for the degree of
Doctor of Philosophy**

BIRMINGHAM, ALABAMA

2000

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ABSTRACT OF DISSERTATION
GRADUATE SCHOOL, UNIVERSITY OF ALABAMA AT BIRMINGHAM

Degree Doctor of Philosophy Program Nutrition Sciences

Name of Candidate Min Sun

Committee Chairs Barbara A. Gower and Michael I. Goran

Title Body Composition, Resting Energy Expenditure, and Pubertal Maturation in
African American and Caucasian Children and Adolescents

Previous cross-sectional studies have shown inconsistent ethnic difference in resting energy expenditure (REE) in African Americans and Caucasians across the life span. We examined the relationship between maturation stage and REE during puberty both cross-sectionally and longitudinally. Total energy expenditure (TEE) was measured using the doubly labeled water technique, REE was measured by indirect calorimetry after an overnight fast, and activity-related energy expenditure (AEE) was estimated from the difference between TEE and REE after reducing TEE by 10% to account for the thermic effect of feeding. Fat mass (FM) and fat-free mass (FFM) were measured by dual-energy X-ray absorptiometry. Fasting serum dehydroepiandrosterone-sulfate (DHEAS), androstenedione, and estrone-sulfate were measured by radioimmunoassay. Maturation stage was determined using the criteria of Tanner by a pediatrician.

We examined TEE, REE, and AEE in prepubertal Caucasian (18 girls, 21 boys) and African American (29 girls, 30 boys) children. TEE, REE, and AEE were similar in Caucasian and African-American prepubertal children after adjusting for FFM and FM. We further examined the relationship between hormonal indices of maturation and TEE, REE, and AEE in African American and Caucasian prepubertal children. Ethnicity was not a significant determinant of any energy expenditure component after adjusting for

body composition and hormone concentrations. In a longitudinal analysis, we examined the relationship between body composition and REE and pubertal maturation among ethnic-gender subgroups. This sample included 92 Caucasian children (28 boys and 64 girls, aged 4.9 to 12.0 years) and 64 African American children (26 boys and 38 girls, aged 4.6 to 12.1 years), who had 2 to 5 years of annual measurements in body composition and REE. Tanner stage ranged from 1 to 5. REE decreased by Tanner stage after adjusting for age, ethnicity, gender, FM, and FFM. There was a significantly higher REE in Caucasians than in African Americans and in boys than in girls after adjusting for age, Tanner stage, FM and FFM. In conclusion, an ethnic difference in REE emerged after the onset of puberty in our cohort. This difference was not explained by pubertal stage or distribution of FFM.

DEDICATION

To
Simone
And
My parents
Who have shown great faith in me

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I gratefully acknowledge my graduate committee: Dr. Michael Goran, for recruiting me into this research group and providing diligent guidance and candid advice; Dr. Barbara Gower, for inspiring me with her intense enthusiasm and knowledge in science and her genuine friendship; Dr. Tim Nagy, for his brilliant insights and suggestions for my research; Dr. Gary Hunter, for his constant encouragement and great interest and support in whatever I have chosen to do; and Dr. Alfred Bartolucci, who patiently guided my learning in statistics and told me to 'keep smiling'.

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LIST OF ABBREVIATIONS

AEE	Activity-related energy expenditure
BMI	Body mass index
DHEA	Dihydroepiandrosterone
DHEAS	Dihydroepiandrosterone-sulfate
DXA	Dual-energy X-ray absorptiometry
FFM	Fat-free mass, lean mass
FM	Fat mass
LLM	Limb lean mass
REE	Resting energy expenditure
TEE	Total energy expenditure
TLM	Trunk lean mass

INTRODUCTION

The Genesis of the Research

Data from National Health Nutrition Examination Survey III have shown that the prevalence of overweight (body mass index [BMI] \geq 95th percentile) among children (aged 6 -11 yr) increased between 1976-1980 (prevalence of 7.6%) and 1988-1991 (prevalence of 10.9%) (1). In a cohort of 6,000 children in Jefferson County, Alabama, African American children had a higher prevalence of obesity (defined as $>120\%$ ideal body weight) than Caucasian children of the same age (5). At age 5, the prevalence in African American girls was 27%, compared with 10% in Caucasian girls; for boys, the prevalence was 22% in African Americans and 6% in Caucasians. By age 11, the prevalence of obesity in African Americans vs. Caucasians was 47 vs. 23% in girls and 29 vs. 13% in boys. The increasing rate of obesity in children and the underlying cause of the higher prevalence of obesity in African Americans than in Caucasians are poorly understood.

Obesity results from an imbalance between energy input and output. However, the etiology of obesity is very complex. Some evidence suggests that reduced energy expenditure may be involved in the etiology of obesity in children (10, 17); however, other studies do not support this concept (3, 4, 8, 9).

Ethnic difference in energy expenditure has been identified in African American and Caucasian children (12, 15, 21, 22). In prepubertal children, ethnic difference in

resting energy expenditure (REE) approximated 200 kcal/d (12, 15). However, evidence regarding whether REE differs by ethnicity has been conflicting (7).

One possible explanation for the inconsistent findings among previous studies is the potential difference in the maturation status of the children. Maturation may influence energy expenditure through its effects on hormones (2, 19) or related change in lean mass (FFM) (20). African American children are reported to be more mature than Caucasian children from age 3 to 12 yr (11, 15, 16). Therefore, differences in maturation rate may result in differences in energy expenditure in different populations. Before puberty, adrenal hormones dehydroepiandrosterone-sulfate (DHEAS) and androstenedione may serve as markers for maturation. These hormones have been shown to be positively correlated with REE and total energy expenditure (TEE) in adult women (2, 19).

Another explanation for the inconsistency in the relationship between energy expenditure and ethnicity is the distribution of FFM. The distribution of FFM refers to different lean components (i.e., internal organs and skeletal muscle), which have different metabolic qualities. Trunk lean mass (TLM) and limb lean mass (LLM) determined by dual-energy X-ray absorptiometry (DXA) may serve as surrogates for mass of organs and muscles that are not easily measured. Appendicular skeletal muscle mass is the sum of skeletal muscle in arms and legs as defined in DXA scan analysis. TLM, because of the organ components, may contribute more to REE than LLM, which reflects mainly skeletal muscle (6). After adjusting for height, body weight, age, and gender, African Americans had greater total appendicular muscle mass than did Caucasians ($P = 0.0001$). Thus, we hypothesized that, compared with Caucasians, African Americans have relatively lower organ mass that may contribute to a lower REE.

No previous studies have examined longitudinally the relationship between maturation, body composition, and REE during puberty in both African American and Caucasian children.

Basic Hypothesis or Problem under Investigation

We hypothesized that REE would be associated with the maturation status of the children during puberty. African American children would have a lower REE than Caucasians after adjusting for age, Tanner stage, fat mass (FM), and FFM. This ethnic difference would become more apparent as children matured.

Overall Experimental Approach

The children that participated in the ongoing longitudinal study were African American and Caucasian boys and girls. They were recruited through the use of newspaper advertisements and by word of mouth. Ethnicity was defined on the basis of the self-ascribed ethnicity status of children's parents and grandparents derived by questionnaire. Children were ineligible if they were: 1) < 4 years of age; 2) taking medications known to affect body composition or physical activity (e.g., prednisone, ritalin, growth hormone); 3) previously diagnosed with syndromes known to affect body composition and/or fat distribution (e.g., Cushing's syndrome, Down's syndrome, type I diabetes, or hypothyroidism); or 4) diagnosed with any major illnesses since birth. All measurements were performed at the General Clinical Research Center (GCRC) or in Department of Nutrition Sciences at the University of Alabama at Birmingham between 1994 and 1999.

Children were admitted to the GCRC in the late afternoon for an overnight visit. When they arrived, a baseline urine sample was collected, and subjects were dosed with the doubly labeled water. Anthropometric measurements were obtained, and dinner was served (~ 1700). An evening snack was allowed, and after 2000 only water and energy-free, noncaffeinated beverages were permitted until after the morning testing. On the following morning after the overnight fast, resting metabolic rate was assessed by indirect calorimetry. Blood samples were collected for hormone analyses. Two timed urine samples were collected for the doubly labeled water analysis. Two weeks later, the children arrived at the Department of Nutrition Sciences at 0700 in the fasted state and body composition was determined by DXA. Two additional timed urine samples were collected for the doubly labeled water analysis.

TEE was measured with the doubly labeled water technique over 14 days under free-living conditions. REE was measured using a Deltatrac Metabolic Monitor (Sensor-medics, Yorba Linda, CA) in the early morning after the children fasted overnight in the GCRC. Physical activity-related energy expenditure (AEE) was estimated from the difference between TEE and REE after reducing TEE by 10% to account for the thermal effect of feeding. Total and regional body composition was measured by DXA. Height was measured without shoes using a stadiometer. Weight was measured in light clothing on an electronic scale. Details on the measurements of energy expenditure and body composition have been discussed previously (18). Maturation stage was determined on the basis of breast stage and pubic hair development in girls (13) and genitalia development in boys (14), according to the criteria of Tanner.

Relationship among Various Parts of the Research

All components of daily energy expenditure were not previously examined in a large sample of African American and Caucasian prepubertal children. Ethnic difference in REE in prepubertal children remained inconsistent. The objective of the first study was to examine the influence of ethnicity on TEE, REE, and AEE adjusting for body composition in African American and Caucasian prepubertal children. This study is described in the paper titled “Total, resting, and activity-related energy expenditure are similar in Caucasian and African American children.”

Subtle difference in maturation rates in African American and Caucasian children may serve as an explanation for the inconsistency in ethnic difference in REE observed in previous cross-sectional studies. If so, this difference may be represented by different hormone concentrations. Therefore, the objective of the second study was to examine the relationship between hormonal indices of maturation and energy expenditure and the relationship between ethnicity and energy expenditure after adjusting for hormonal indices of maturation. This study is described in the paper titled “Do hormonal indices of maturation explain energy expenditure differences in African American and Caucasian prepubertal children?”

During puberty, body composition and REE change dramatically. The relationships between body composition or REE and pubertal maturation have not been examined longitudinally in both African American and Caucasian children and adolescents during puberty. Difference in FFM distribution may serve as another explanation for potential maturational and ethnic difference in REE. Therefore, the objectives of the third study were 1) to examine the relationship between pubertal stage and body composition or REE

longitudinally, 2) to examine the relationship between ethnicity and body composition or REE, and 3) to examine whether there was an ethnic difference in FFM distribution and whether it would contribute to any maturational and ethnic differences in REE. This study is described in the paper titled “A longitudinal study of body composition and resting energy expenditure during puberty in African American and Caucasian children and adolescents.”

TOTAL, RESTING, AND ACTIVITY-RELATED ENERGY EXPENDITURE ARE
SIMILAR IN CAUCASIAN AND AFRICAN AMERICAN CHILDREN

by

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Abstract

There is some evidence to suggest that ethnic differences in energy expenditure in adults may modulate different propensities for obesity. However, there is lack of data for the components of energy expenditure in young children of different ethnic backgrounds. In this study, we examined total energy expenditure (TEE), resting energy expenditure (REE), and physical activity-related energy expenditure (AEE) in healthy prepubertal Caucasian (18 girls, 21 boys) and African American (29 girls, 30 boys) children. TEE was measured over 14 days under free-living conditions with the doubly labeled water technique. REE was from indirect calorimetry after an overnight fast, and AEE was estimated from the difference between TEE and REE after reducing TEE by 10% to account for the thermic effect of feeding. Fat mass (FM) and fat-free mass (FFM) were measured by dual-energy X-ray absorptiometry. There were no significant effects of ethnicity or gender on TEE after adjusting for FFM or for both FFM and FM. For REE, there was no effect of ethnicity, but a significant effect of gender, with a higher REE in boys after adjusting for FFM and FM ($P < 0.001$). For AEE, there were no significant effects of ethnicity or gender after adjusting for FFM or for FFM and FM. In conclusion, ethnicity was not a significant determinant for any of the components of energy expenditure. TEE, REE, and AEE were similar in Caucasian and African American prepubertal children after adjusting for FFM or for FFM and FM.

Introduction

Data from The National Health and Nutrition Examination Survey III show that the prevalence of overweight (body mass index \geq 95th percentile) among children (age 6

- 11 yr) continued to increase between 1976 and 1980 (prevalence of 7.6%) and 1988 and 1991 (prevalence of 10.9%) (1). In a cohort of 6,200 children in Birmingham, Alabama, African American children had a higher prevalence of obesity (defined as $>120\%$ ideal body weight) than Caucasian children of the same age (8). At age five, the prevalence in African American girls was 23%, compared with 10% in Caucasian girls; for boys, the prevalence was 26% in African Americans and 13% in Caucasians. By age 10, the prevalence of obesity in African Americans vs. Caucasians was 38 vs. 21% in girls and 26 vs. 21% in boys. The underlying cause of the higher prevalence of obesity among African American children is unknown. Some evidence suggests that a reduced energy expenditure may be involved in the etiology of obesity in children (17, 22); however, other studies do not support this concept (4, 5, 11, 14).

In the prepubertal age, ethnic differences of the components in energy expenditure have been identified. We have shown that total energy expenditure was 8.5% higher because of a 37% higher physical activity-related energy expenditure in Mohawk Indian compared with Caucasian children independent of fat-free mass and gender (15). In addition, two earlier studies that compared Mohawk or Pima Indian with Caucasian children found that ethnicity was not a significant determinant of resting energy expenditure after adjusting for fat-free mass, fat mass, and gender (9, 14). In contrast, resting energy expenditure was found to be 14% lower in African American than in Caucasian children, after adjustment for age, gender, body weight, fat-free mass, and fat mass (18). A 4% lower resting energy expenditure was also found in normal weight African American compared with Caucasian girls, after adjustment for fat-free mass and body density (25).

These studies indicate that ethnicity may be an important determinant for energy expenditure in children; however, the effects of ethnicity remain uncertain.

Ethnic differences in energy expenditure have also been examined in other age groups. Among girls aged 6-16 yr, lower resting energy expenditure was found in African Americans than in Caucasians after adjusting for both body weight and lean body mass (20). Lower resting energy expenditure was also observed in African American adult women (10). Compared with Caucasians, African American women had an 8% lower resting energy expenditure independent of fat-free mass. In addition, older (>55 yr of age) African Americans had a 10% lower total energy expenditure, because of a 5% lower resting energy expenditure and a 19% lower physical activity-related energy expenditure than Caucasians, independent of fat-free mass and gender (3).

However, no previous study has examined the various components of total daily energy expenditure in African American vs. Caucasian children. We hypothesized that total daily energy expenditure was not different between Caucasian and African American children. The current study evaluated the determinants for the components of total daily energy expenditure in 98 African American and Caucasian children. The main purpose was to examine the possible influence of ethnicity on total energy expenditure, resting energy expenditure, and physical activity-related energy expenditure after controlling for fat-free mass or for both fat and fat-free mass in African American vs. Caucasian prepubertal children.

Methods

Subjects. Our sample included 39 Caucasian children (18 girls, 21 boys) aged 8.3 ± 1.4 yr (range 5.2 to 10.9 yr) and 59 African American children (29 girls, 30 boys) aged 7.5 ± 1.5 yr (range 4.7 to 10.0 yr) from Birmingham, Alabama. Children were defined as Caucasian or African American on the basis of the ethnic status of their parents and grandparents derived by questionnaire. The children were recruited with the use of newspaper advertisements, word of mouth, and a school intervention study. Children were ineligible if they were either 1) < four years of age, 2) beyond Tanner stage I, as assessed by a physical examination by a pediatrician (Tanner stage I was defined on the basis of breast stage and pubic hair development in girls and genitalia development in boys), 3) taking medications known to affect body composition or physical activity (e.g., prednisone, ritalin, growth hormone), 4) previously diagnosed with syndromes known to affect body composition and/or fat distribution (e.g., Cushing's syndrome, Down's syndrome, type I diabetes, hypothyroidism), or 5) diagnosed with any major illnesses since birth. We have previously reported body composition (16) and fitness (24) data in these children. The nature, purpose, and possible risks of the study were carefully explained to the parent before consent was obtained. Studies were performed during the school year, mainly in the fall for African Americans and mainly in the spring and fall for Caucasians. This study was approved by the Institutional Review Board at the University of Alabama at Birmingham, and parents provided informed consent before testing commenced. All measurements were performed at the General Clinical Research Center (GCRC) or in The Department of Nutrition Sciences at the University of Alabama at Birmingham between 1994 and 1997.

General outline of protocol. Children were admitted to the GCRC in the late afternoon for an overnight visit. On arrival, a baseline urine sample was collected, and subjects were dosed with the doubly labeled water. Anthropometric measurements were obtained, and dinner was served (~1700). An evening snack was allowed, and after 2000 only water and energy-free, noncaffeinated beverages were permitted until after the morning testing. On the following morning after overnight fast, resting metabolic rate was assessed by indirect calorimetry on awakening of subjects, and two timed urine samples were collected for the doubly labeled water analysis. Two weeks later, the children arrived at the Energy Metabolism Research Unit at 0700 in the fasted state, and body composition was determined by dual-energy X-ray absorptiometry (DXA). Two additional timed urine samples were collected for the doubly labeled water analysis.

Measurement of energy expenditure components. Total energy expenditure was measured over 14 days under free-living conditions with the doubly labeled water technique with the use of a protocol with a theoretical error of < 5%, as previously described (11). For deuterium relative to baseline, the average initial enrichment was $1,183.28 \pm 301.19$ ‰, and the mean final enrichment was 332.75 ± 125.63 ‰ (equivalent to just over 2 biological half-lives). For oxygen-18 relative to the baseline, the average initial enrichment was 110.55 ± 20.93 ‰, and the mean final enrichment was 19.00 ± 6.92 ‰ (equivalent to just over 2 biological half-lives). Samples were analyzed in triplicate for H_2^{18}O and $^2\text{H}_2\text{O}$ by isotope ratio mass spectrometry at the University of Alabama at Birmingham as previously described (11). The mean dilution space ratios (DSR) were not significantly different among Caucasian girls, Caucasian boys, African American girls, and African

American boys. When all samples for deuterium and oxygen-18 were reanalyzed in seven subjects, values of total energy expenditure were in close agreement ($\pm 4.3\%$), as previously described (11). CO_2 production rate was determined using *equation R2* of Speakman et al. (23), assuming a fixed DSR of 1.0427, and energy expenditure was calculated using *equation 12* of de Weir (5). Mean value for the food quotient of the children's diet (0.90 in Caucasian and 0.87 in African American) was determined by duplicate 24-h recall. Total energy expenditure data were screened for physiological outliers by regressing total energy expenditure against body weight, fat-free mass, and resting energy expenditure. Data were considered outliers if the residual from any of these regressions were greater or less than three standard deviations from the means. Seven values did not meet these criteria and were removed for the main analyses presented.

Resting energy expenditure was measured in the early morning after an overnight fast in the GCRC, using a Deltatrac Metabolic Monitor (Sensormedics, Yorba Linda, CA). During testing, all subjects were instructed to lie as still as possible and remain awake. An adult-size canopy hood was used to collect the expired air. After a 10-min equilibration period, data on oxygen consumption and carbon dioxide production were collected continuously for 20 min. Energy expenditure was calculated using the equation of de Weir (5).

Physical activity-related energy expenditure was estimated in two ways. One was from the difference between total and resting energy expenditure after reducing total energy expenditure by 10% to account for the thermic effect of feeding (19). Another was from the residual of the regression between total and resting energy expenditure.

Assessment of body composition and anthropometry. Total and regional body composition were measured by DXA using a Lunar DPX-L densitometer that we have previously validated in the pediatric body weight range (13, 21). Subjects were scanned in light clothing while lying flat on their backs with arms by the side. DXA scans were performed and analyzed using pediatric software (version 1.5e), as previously described (13, 21). On the day of each test, the DPX-L was calibrated using the procedures provided by the manufacturer. Height was measured without shoes using a stadiometer. Weight was measured in light clothing on an electronic scale.

Statistics. Differences in physical characteristics, body composition, and energy expenditure among African American and Caucasian boys and girls were examined in a two-way (gender and ethnic groups) analysis of covariance (ANCOVA). Total energy expenditure, resting energy expenditure, and activity-related energy expenditure were examined as dependent variables with fat and fat-free mass as covariates and ethnicity and gender as the main effect variables. Total energy expenditure was also examined, with resting energy expenditure as a covariate, for differences in nonresting (i.e., mainly physical activity-related) energy expenditure. Similarity of regression slopes among the subgroups (by ethnicity and gender) was examined by the significance of the interaction between the covariate and each of the two grouping variables and was a prerequisite for proceeding with each of the ANCOVA models. Data were analyzed using SAS software version 6.10 (Carey, NC), with a significance level set at $P < 0.05$ for all tests.

Results

Subject characteristics. The characteristics of the children are presented in Table 1. Caucasian children were older than the African Americans ($P = 0.006$). The girls overall were heavier ($P = 0.001$), had greater fat mass ($P = 0.0001$), and had a higher absolute physical activity-related energy expenditure than boys ($P = 0.02$). Caucasian girls had the highest weight, body mass, and absolute total energy expenditure among the four subgroups.

Analysis of ethnic differences in total energy expenditure. The relationships between total energy expenditure and fat-free mass in the four subgroups are summarized in Table 2 and Fig. 1. The regression slopes were not significantly different among the four subgroups. After adjusting for fat-free mass or for fat-free and fat mass, adjusted least square means of total energy expenditure were not significantly influenced by ethnicity or the interaction of ethnicity and gender (Table 2). When the interaction of ethnicity and gender was excluded from the ANCOVA model, the effect of ethnicity remained insignificant ($P = 0.2$). When genders were combined, there were still no significant effects of ethnicity on total energy expenditure after adjusting for fat-free mass ($P = 0.2$) or for fat-free and fat mass ($P = 0.2$) (data not shown).

Analysis of ethnic differences in resting energy expenditure. The relationships between resting energy expenditure and fat-free mass in the four subgroups are summarized in Table 3 and Fig. 1. The regression slopes were not significantly different among the four subgroups. Resting energy expenditure was not significantly influenced by ethnicity

Table 1. *Subject characteristics*

	All Children (<i>n</i> = 98)	Caucasian Girls (<i>n</i> = 18)	Caucasian Boys (<i>n</i> = 21)	African American Girls (<i>n</i> = 29)	African American Boys (<i>n</i> = 30)	2- Way ANOVA (<i>P</i> value)
Age, yr	7.8 ± 1.5 (4.7 - 10.9)	8.4 ± 1.1 (5.2 - 9.8)	8.3 ± 1.6 (5.6 - 10.9)	7.6 ± 1.6 (4.7 - 9.9)	7.3 ± 1.5 (5.1 - 10.0)	Ethnicity (<i>P</i> = 0.006)
Weight, kg	33.4 ± 11.9 (17.0 - 68.4)	41.2 ± 13.0 (18.5 - 66.0)	29.3 ± 7.2 (17.0 - 45.1)	34.5 ± 11.9 (18.8 - 62.6)	30.5 ± 11.2 (19.8 - 68.4)	Gender (<i>P</i> = 0.001)
Height, cm	129.5 ± 10.8 (107.0 - 155.0)	131.9 ± 11.1 (110.0 - 152.8)	129.8 ± 9.9 (110.0 - 148.5)	129.4 ± 11.3 (107.8 - 155.0)	127.8 ± 11.1 (107.0 - 153.0)	NS
FFM, kg	22.0 ± 5.4 (12.4 - 37.6)	23.9 ± 6.2 (13.4 - 37.6)	20.8 ± 3.8 (12.4 - 25.9)	21.7 ± 5.0 (14.0 - 31.6)	21.9 ± 5.9 (14.6 - 35.3)	NS
FM, kg	10.8 ± 8.2 (2.0 - 44.7)	16.0 ± 9.9 (4.2 - 44.7)	7.1 ± 4.7 (2.6 - 19.7)	12.7 ± 7.6 (3.8 - 29.2)	8.2 ± 7.7 (2.0 - 30.8)	Gender (<i>P</i> = 0.0001)
TEE, kcal/day	1,757 ± 403 (972 - 2,749)	1,946 ± 432 (1,246 - 2,723)	1,704 ± 330 (1,189 - 2,537)	1,717 ± 377 (1,221 - 2,538)	1,718 ± 441 (972 - 2,749)	NS
REE, kcal/day	1,247 ± 215 (919 - 2,116)	1,280 ± 221 (919 - 1,869)	1,244 ± 198 (919 - 1,765)	1,210 ± 199 (967 - 1,611)	1,265 ± 242 (930 - 2,116)	NS
AEE, kcal/day	334 ± 255 (-232 - 998)	472 ± 260 (95 - 928)	290 ± 206 (-80 - 715)	335 ± 242 (-183 - 965)	282 ± 277 (-232 - 998)	Gender (<i>P</i> = 0.02)

Data are means ± SD with range in parenthesis; *n*, no. of children/group. FFM, fat free mass; FM, fat mass; TEE, total energy expenditure; REE, resting energy expenditure; AEE, activity-related energy expenditure; ANOVA, analysis of variances; NS, not significant.

Table 2. *Linear regression between TEE and FFM, and adjusted least square means for TEE obtained from ANCOVA adjusted either for FFM or for both FFM and FM*

	Intercept kcal/day	Slope, * kcal·day ⁻¹ ·kg ⁻¹	Adjusted <i>r</i> ²	Adjusted Least Square Means kcal/day	
				TEE adjusted for FFM†	TEE adjusted for FFM and FM‡
Caucasian girls (<i>n</i> = 18)	550 ± 232	58.3 ± 9.4	0.69	1,831 ± 60	1,766 ± 57
Caucasian boys (<i>n</i> = 21)	721 ± 371	47.3 ± 17.6	0.25	1,774 ± 56	1,824 ± 53
African American girls (<i>n</i> = 29)	414 ± 191	60.1 ± 8.6	0.63	1,734 ± 47	1,686 ± 44
African American boys (<i>n</i> = 30)	385 ± 180	70.0 ± 8.0	0.67	1,725 ± 46	1,777 ± 44

Data are means ± SE. Least square means were adjusted for ethnicity, gender, and the interaction of ethnicity and gender. * Slopes are not significantly different among 4 subgroups (*P* > 0.2). †Least square means of TEE adjusted for FFM are not significantly different among subgroups (ethnicity, *P* = 0.2; gender, *P* = 0.5; interaction, *P* = 0.7). ‡Least square means of TEE adjusted for both FFM and FM are not significantly different among subgroups (ethnicity, *P* = 0.2; gender, *P* = 0.2; interaction, *P* = 0.7). ANCOVA, analysis of covariance.

or the interaction of ethnicity and gender after adjustments for fat-free mass or both fat-free mass and fat mass. When the interaction of ethnicity and gender was excluded from the ANCOVA model, the effect of ethnicity remained insignificant.

Analysis of ethnic differences in activity-related energy expenditure. The relationships between activity-related energy expenditure (expressed as the difference between total and resting energy expenditure) and fat-free mass in the four subgroups are summarized in Table 4 and Fig. 1. The regression slopes were not significantly different among the four subgroups. Activity-related energy expenditure was not significantly influenced by ethnicity

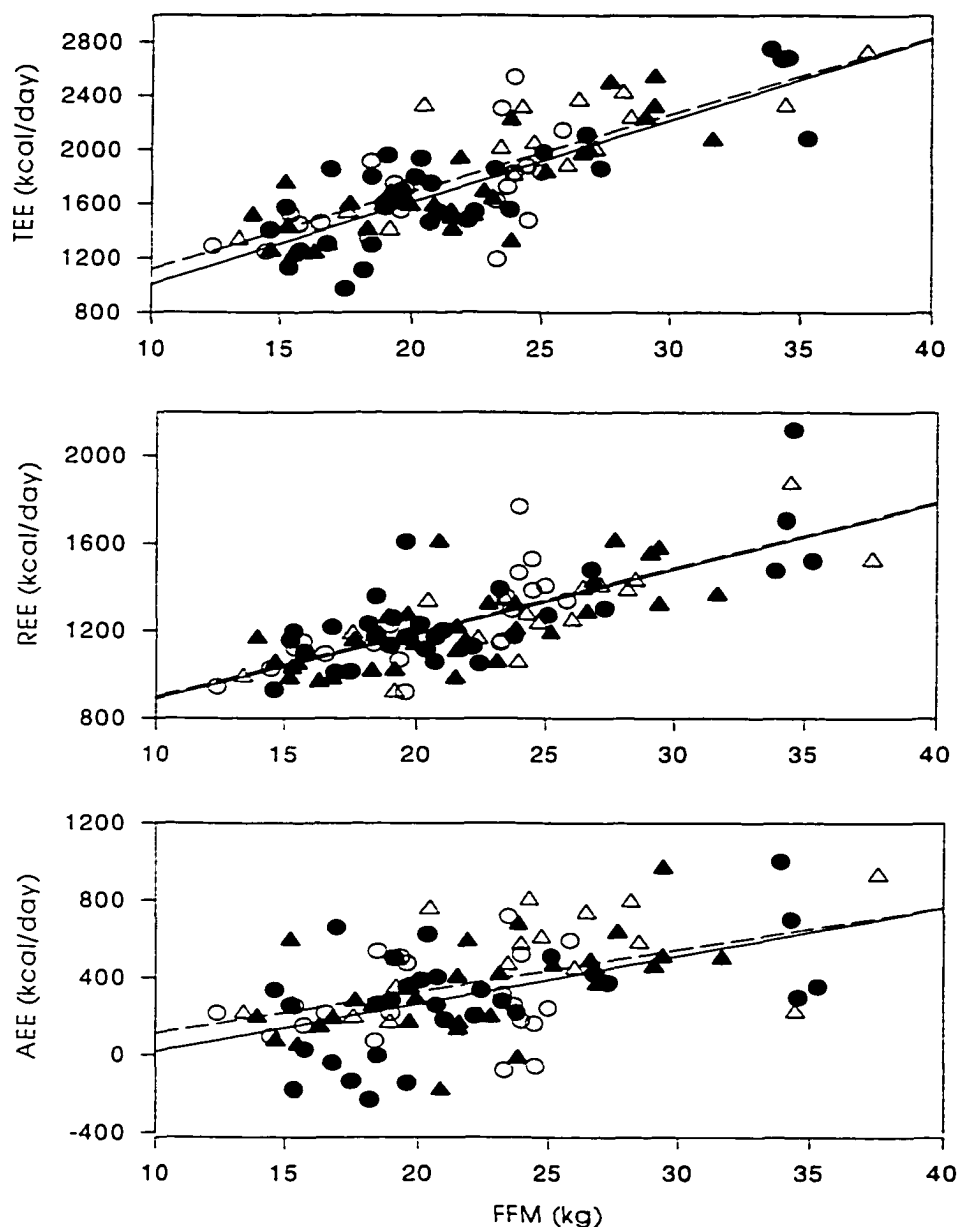


Fig. 1. Relationships between total energy expenditure (TEE) and fat-free mass (FFM), resting energy expenditure (REE) and FFM, and activity-related energy expenditure (AEE) and FFM in 39 Caucasian (dotted line, open symbols) and 59 African American (solid line, filled symbols) children. Triangles, girls; circles, boys. Regression slopes are not significantly different for both ethnic groups for all components of energy expenditure. Regression equations for Caucasian children: TEE (kcal/day) = 57.0 FFM + 548.5; REE (kcal/day) = 29.6 FFM + 599.8; AEE (kcal/day) = 21.7 FFM - 106.2. Regression equations for African American children: TEE (kcal/day) = 60.6 FFM + 397.9; REE (kcal/day) = 29.7 FFM + 590.1; AEE (kcal/day) = 24.8 FFM - 232.0.

Table 3. Linear regression between REE and FFM and adjusted least square means for REE obtained from ANCOVA adjusted either for FFM or for both FFM and FM

	Intercept, kcal/day	Slope, * Kcal·day ⁻¹ · kg ⁻¹	Adjusted <i>r</i> ²	Adjusted Least Square Means, kcal/day	
				REE adjusted for FFM†	REE adjusted for FFM and FM‡
Caucasian girls (<i>n</i> = 18)	589 ± 127	28.8 ± 5.1	0.64	1220 ± 35	1183 ± 33
Caucasian boys (<i>n</i> = 21)	462 ± 186	37.5 ± 8.8	0.47	1278 ± 33	1306 ± 31
African American girls (<i>n</i> = 29)	591 ± 117	28.5 ± 5.3	0.50	1219 ± 27	1192 ± 26
African- American boys (<i>n</i> = 30)	599 ± 115	30.4 ± 5.1	0.55	1267 ± 27	1297 ± 26

Data are means ± SE. Least square means were adjusted for ethnicity, gender, and interaction of ethnicity and gender. *Slopes are not significantly different among four subgroups ($P > 0.2$). †Least square means of REE adjusted for FFM are not different among subgroups (ethnicity, $P = 0.8$; gender, $P = 0.08$; interaction, $P = 0.9$). ‡Least square means of REE adjusted for both FFM and FM are significantly influenced by gender ($P = 0.0005$), but not by ethnicity ($P = 0.9$) and interaction ($P = 0.8$).

or the interaction of ethnicity and gender. When the interaction of ethnicity and gender was excluded from the ANCOVA model, the effect of ethnicity remained insignificant ($P = 0.2$). When genders were combined, there was still no significant effect of ethnicity on activity-related energy expenditure after adjusting for fat-free mass ($P = 0.2$), or for fat-free and fat mass ($P = 0.2$) (data not shown). The relationships between total energy expenditure and resting energy expenditure are summarized in Table 5. The regression slopes were not significantly different among the four subgroups. When resting energy expenditure was the only covariate in the model, there was not a significant effect of eth-

nicity on total energy expenditure. When fat-free mass or both fat-free and fat mass were added to the model, the effects of ethnicity remained insignificant.

Table 4. Linear regression between AEE and FFM and adjusted least square means obtained from ANCOVA, adjusted either for FFM or for FFM and FM

	Intercept kcal/day	Slope, * kcal·day ⁻¹ ·kg ⁻¹	Adjusted <i>r</i> ²	Adjusted Least Square Means, kcal/day	
				AEE adjusted for FFM†	AEE adjusted for FFM and FM‡
Caucasian girls (<i>n</i> = 18)	-93.7 ± 212.1	23.6 ± 8.6	0.28	428 ± 53	406 ± 54
Caucasian boys (<i>n</i> = 21)	186.5 ± 273.1	5.2 ± 12.9	-0.05	318 ± 50	335 ± 51
African American girls (<i>n</i> = 29)	-218.9 ± 174.3	25.5 ± 7.8	0.26	342 ± 41	326 ± 42
African American boys (<i>n</i> = 30)	-252.4 ± 169.4	24.4 ± 7.5	0.25	284 ± 40	302 ± 42

Data are means ± SE. All least square means were adjusted for ethnicity, gender, and interaction of ethnicity and gender. *Slopes are not significantly different among 4 subgroups (*P* > 0.2). †Least square means of AEE adjusted for FFM are not significantly different among subgroups (ethnicity, *P* = 0.2; gender, *P* = 0.08; interaction, *P* = 0.6). ‡Least square means of AEE adjusted for both FFM and FM are not significantly different among subgroups (ethnicity, *P* = 0.2; gender, *P* = 0.4; interaction, *P* = 0.6).

Discussion

Our study is the first attempt to examine ethnic differences in all components of daily energy expenditure in a heterogeneous group of African American and Caucasian prepubertal children. The primary finding is that ethnicity (African American vs. Caucasian Children) was not a significant independent determinant of total energy expenditure, resting energy expenditure, or physical activity-related energy expenditure after adjusting for fat-free mass or for both fat-free and fat mass.

Table 5. Linear regression between TEE and REE and adjusted least square means obtained from ANCOVA, adjusted either for REE and FFM, or FFM and FM

	Intercept kcal/day	Slope, * kcal·day ⁻¹ ·kg ⁻¹	Adjusted <i>r</i> ²	Adjusted Least Square Means (kcal/day)		
				Adjusted for REE†	Adjusted for FFM‡	Adjusted for FFM and FM ^^
Caucasian girls (<i>n</i> = 18)	15.7 ± 404	1.5 ± 0.3	0.57	1902 ± 65	1865 ± 56	1790 ± 57
Caucasian boys (<i>n</i> = 21)	212 ± 333	1.2 ± 0.3	0.49	1707 ± 60	1755 ± 53	1801 ± 53
African American girls (<i>n</i> = 29)	83 ± 314	1.4 ± 0.3	0.49	1766 ± 51	1750 ± 44	1706 ± 44
African American boys (<i>n</i> = 30)	45 ± 305	1.3 ± 0.2	0.51	1695 ± 50	1712 ± 43	1758 ± 44

Data are means ± SE. All least square means were adjusted for ethnicity, gender, and interaction of ethnicity and gender. *Slopes are not significantly different among 4 subgroups (*P* > 0.2). †Least square means of TEE adjusted for REE are significantly influenced by gender (*P* = 0.02), but not by ethnicity (*P* = 0.2) and interaction (*P* = 0.3). ‡Least square means of TEE adjusted for REE and FFM are not significantly different among subgroups (ethnicity, *P* = 0.1; gender, *P* = 0.2; interaction, *P* = 0.6). ^^Least square means of TEE adjusted for REE, FFM, and FM are not significantly different among subgroups (ethnicity, *P* = 0.2; gender, *P* = 0.6; interaction, *P* = 0.7).

We did not observe an ethnic difference in total energy expenditure when we compared African American with Caucasian children. No previous study has examined the ethnic influence (African American vs. Caucasian) on total energy expenditure in prepubertal children. The effect of ethnicity (African American vs. Caucasian) on total energy expenditure has been described in only one previous study in older (> 55 yr of age) adults ($n = 164$) (3). In that study, total energy expenditure was 10% lower in African American than in Caucasian subjects after adjusting for fat-free mass. The mechanism is unclear for the inconsistency in the ethnic effect on total energy expenditure in different age groups.

Resting energy expenditure was not significantly influenced by ethnicity when African American and Caucasian children were compared. This result does not support previous data, which showed a significantly lower resting energy expenditure in African American compared with Caucasian children (18, 20, 25). Despite similar methodology compared with the current study, two of the prior studies (18, 20) have found a strikingly lower resting energy expenditure (over 200 kcal/day) in African American than in Caucasian children. One explanation could be the small sample sizes in those two studies (<10 in some subgroups) that confounded the results. However, the lower resting energy expenditure in African Americans is supported by data in adult women (10) and in older adults (3).

Ethnicity did not have a significant influence on physical activity-related energy expenditure in this study. We have previously shown that a subgroup of African American children ($n = 44$) had a 15% lower maximal oxygen consumption (VO_{2max}) compared with a subgroup ($n = 31$) of Caucasian children from the same population, independent of

differences in soft lean tissue, leg lean tissue, fat mass, total energy expenditure, and physical activity-related energy expenditure ($P < 0.001$) (24). Because activity-related energy expenditure was similar and $\text{VO}_{2\text{max}}$ was lower, African American children may either have a decreased capacity to sustain exercise aerobically for extended periods of time or participated in less intense activities for longer periods of time compared with Caucasian children. Therefore, there may be ethnicity-related differences in qualitative aspects of physical activity. However, in this study, ethnicity was not found to affect activity-related energy expenditure. Thus additional studies that focus on potential ethnic differences in qualitative aspects of physical activity and activity pattern are needed.

In addition, our study did not find a significant effect of gender on total energy expenditure (Table 2) and activity-related energy expenditure (Tables 4 and 5). Our previous study in 30 Caucasian children (4-6 yr of age) also did not find a significant gender effect on total energy expenditure after adjusting for body composition (12). Conversely, in older adults (3), total energy expenditure was 16% higher in males than in females ($P < 0.01$) after controlling for fat-free mass. In our study, gender was a significant determinant of resting energy expenditure, which was higher in boys than in girls (Table 3), after adjustment for fat-free and fat mass. This result is consistent with other findings that observed higher resting energy expenditure in boys than in girls of different ethnic groups (9, 14, 15). The higher resting energy expenditure in males was suggested to be a hormonal influence (7). However, because all the children in our study were at prepubertal age and the same Tanner stage, a hormonal effect would not be expected at this young age (2). Thus the independent effect of gender may not be attributed entirely to differences in sex hormone levels.

A possible explanation for the inconsistent findings relating to ethnic differences in energy expenditure may be the tremendous variation in total energy expenditure, especially activity-related energy expenditure. Total and activity-related energy expenditure were measured for only 14 days, which may not reflect variation over time. Moreover, changes in energy expenditure and energy intake may occur in some critical periods of time in development (such as in early infancy or adolescence). In addition, differences in energy expenditure among different ethnic and gender groups may appear at different stages of development.

In our study, total energy expenditure may also have been influenced by other factors such as age, body mass, and the study design. There was a small age difference between African American (7.4 yr) and Caucasian (8.3 yr) children. Because all the children were at the same Tanner stage, we do not believe this small age difference had a major impact on the primary findings. This fact was confirmed by including age in the ANCOVA models as a covariate. When that was done, we still did not see a significant effect of ethnicity on energy expenditure components. The observation of a larger body mass in Caucasian girls may have been due to selection bias. Approximately one-half of our cohort was obese (defined by a weight-for-height >90th percentile) and one-half was nonobese. Therefore, the difference in body mass among subgroups raises the question of whether this particular group represents the general population. The limitation of this study was the use of cross-sectional study design. Although we did not observe an ethnic difference in any component of daily energy expenditure, we cannot rule out the possible influence of energy expenditure on the development of obesity on a long-term basis. Longitudinal studies may better describe the influences of different components of energy

expenditure on the changes of body composition in the development of obesity, especially in African American children. Further longitudinal studies are needed to examine the underlying etiology of obesity.

In conclusion, ethnicity was not a significant determinant for any of the components of energy expenditure in this cross-sectional analysis. Total energy expenditure, resting energy expenditure, and physical activity-related energy expenditure were similar in Caucasian and African American prepubertal children after adjustment for body composition.

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DO HORMONAL INDICES OF MATURATION EXPLAIN ENERGY
EXPENDITURE DIFFERENCES IN AFRICAN AMERICAN AND CAUCASIAN
PREPUBERTAL CHILDREN?

by

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Abstract

The purpose of this study was to examine the relationships between hormonal indices of maturation and total, resting, and physical activity-related energy expenditure (TEE, REE, and AEE) in African American and Caucasian prepubertal children. Sixty-four African American and 48 Caucasian prepubertal children were measured for TEE (by doubly labeled water), REE (by indirect calorimetry), fat mass and fat-free mass (by dual-energy X-ray absorptiometry), and fasting serum dehydroepiandrosterone-sulfate (DHEAS), androstenedione, and estrone-sulfate (by radioimmunoassay). Serum concentrations of hormones correlated significantly with REE and TEE (r values range from 0.33 to 0.76, $P < 0.001$). Only androstenedione correlated significantly with AEE ($r = 0.23$, $P < 0.05$). However, these correlations were no longer significant after adjusting energy expenditure components for fat-free mass. In multiple regression models, ethnicity was not a significant determinant of any energy expenditure component after adjusting for body composition and hormone concentrations. In conclusion, hormonal indices of maturation do not influence energy expenditure in this group of African American and Caucasian prepubertal children.

Introduction

Previous studies have shown a lower resting energy expenditure in African Americans compared with Caucasians. These findings have been shown in children (13, 19, 31, 32), adult women (6, 12), and the elderly (3). However, a previous study from our laboratory in prepubertal children did not observe differences in total, resting, or physical

activity-related energy expenditure between African Americans and Caucasians, after adjusting for gender and body composition (26).

One possible explanation for inconsistent findings among studies is potential differences in the maturation status of children. Maturation may influence energy expenditure through its related changes in fat-free mass (30) or effects on hormones (1, 27). At age 3, 7, 8, 9, and 10, African American children are more mature (11, 20). Therefore, differences between studies regarding ethnic difference in energy expenditure could be due to population differences in maturation, which may obscure the actual relationship between ethnicity and energy expenditure previously observed. However, after adjusting for pubertal stage (defined by physical examination) and lean body mass, Morrison et al (19) still observed a lower resting energy expenditure in African American than in Caucasian girls (6-16 yr). Therefore, we hypothesized that there could be an ethnic difference in energy expenditure after adjusting for differences in maturation in our prepubertal cohort.

Because the rate of maturation differs between African American and Caucasian children, differences between studies in the presence or absence of an ethnic difference in resting energy expenditure may be due to differences in the concentrations of circulating hormones that increase with maturation. Our previous study (26) was conducted in prepubertal children, as defined by physical examination (Tanner stage I). Even within stages of maturation defined by physical examination, there may be more subtle differences in maturation, which could be reflected by differences in hormones including dehydroepiandrosterone-sulfate (DHEAS) and androstenedione (21, 23).

The androgens DHEAS and androstenedione have been shown to be positively correlated with total energy expenditure in adult women (1, 27). The relationships between androgen concentrations and components of energy expenditure have never been examined in children. Estradiol has been shown to increase energy expenditure in rats (10). However, the relationship of estradiol and energy expenditure has never been examined in humans. Estrone-sulfate, the form of estrogen that circulates in the highest concentration, could be locally converted to estradiol, which is undetectable in the systemic circulation in prepubertal children. Therefore, estrone-sulfate may serve as a marker for estrogen status in prepubertal children. In addition, adrenal androgens could be converted to estrogens. Because of wide ranges of hormone concentrations in prepubertal children (2), the influence of hormones on energy expenditure needs to be further examined when considering ethnic differences in energy expenditure. We hypothesized that adrenal androgens and estrogen are positively associated with energy expenditure in prepubertal children and could possibly explain reported ethnic differences in energy expenditure. The main purpose of this study is to examine whether concentrations of hormones are related to total, resting, and activity-related energy expenditure after adjustment for body composition in African American and Caucasian prepubertal children.

Methods

Subjects. Our study included 48 Caucasian children (30 boys and 18 girls) and 64 African American children (34 boys and 30 girls) from Birmingham, Alabama. The children were recruited using newspaper advertisements and by word of mouth. Children included in this study were at the prepubertal stage based on breast stage and pubic hair de-

velopment in girls (15) and genitalia development in boys (16), according to the criteria of Tanner. We further confirmed prepubertal status by absence of detectable concentrations of testosterone and estradiol. Two subjects had marginally detectable estradiol concentrations of 4.90 and 4.96 pg/ml and six subjects had low testosterone concentrations ranging from 12.0 to 21.9 ng/dl. These hormone values are below those associated with pubertal maturation (4, 14). Ethnicity was defined based on the self-ascribed ethnicity status of children's parents and grandparents derived by questionnaire. The eligibility criteria were discussed in detail in a previous paper (26). We have also previously reported energy expenditure (26), body composition (9), and aerobic fitness (28) data in these children. The nature, purpose, and possible risks of the study were carefully explained to the parent before obtaining consent. This study was approved by the Institutional Review Board at the University of Alabama at Birmingham, and parents provided informed consent before testing commenced. All measurements were performed at the General Clinical Research Center (GCRC) or in The Department of Nutrition Sciences at the University of Alabama at Birmingham between 1994 and 1997.

General outline of protocol. Children were admitted to the GCRC in the late afternoon for an overnight visit. Upon arrival, a baseline urine sample was collected, and subjects were dosed with the doubly labeled water. Anthropometric measurements were obtained, and dinner was served (~ 1700). An evening snack was allowed, and after 2000 only water and energy-free, noncaffeinated beverages were permitted until after the morning testing. On the following morning after an overnight fast, resting metabolic rate was assessed by indirect calorimetry upon awakening at 0500. The subjects were allowed

to urinate if necessary. If the subjects urinated, they were asked to rest for 15 min before resting metabolic rate measurement was taken. At 0700, blood samples were collected for hormone analyses. Two timed urine samples were collected for the doubly labeled water analysis. Two weeks later, the children arrived at the Department of Nutrition Sciences at 0700 in the fasted state and body composition was determined by dual-energy X-ray absorptiometry (DXA). Two additional timed urine samples were collected for the doubly labeled water analysis.

Measurement of energy expenditure components. Total energy expenditure was measured over 14 days under free-living conditions with the doubly labeled water technique, using a protocol with a theoretical error of less than 5%, as previously described (7). Samples were analyzed in triplicate for H_2^{18}O and $^2\text{H}_2\text{O}$ by isotope ratio mass spectrometry at the University of Alabama at Birmingham as previously described (7). The mean dilution space ratios (DSR) were not significantly different between gender or ethnic groups. CO_2 production rate was determined using *equation R2* of Speakman et al (24) and assuming a fixed DSR of 1.0427; energy expenditure was calculated using *equation 12* of De V. Weir (5); and the mean value for the food quotient of the children's diet (0.90 in Caucasian and 0.87 in African American) was determined by 24 h recall performed in duplicate.

Resting energy expenditure was measured in the early morning after an overnight fast in the GCRC using a Deltatrac Metabolic Monitor (Sensormedics, Yorba Linda, CA). During testing, all subjects were instructed to lie as still as possible and remain awake. An adult-size canopy hood was used to collect the expired air. After a 10-min equilibra-

tion period, data on oxygen consumption and carbon dioxide production were collected continuously for 20 min. Energy expenditure was calculated using *equation 12* of De V. Weir (5).

Physical activity-related energy expenditure was estimated from the difference between total and resting energy expenditure after reducing total energy expenditure by 10% to account for the thermic effect of feeding. Seventy-six subjects had complete analyses for total and activity-related energy expenditure.

Assessment of body composition and anthropometry. Total and regional body compositions were measured by DXA using a Lunar DPX-L densitometer that we have previously validated in the pediatric body weight range (8, 22). Subjects were scanned in light clothing while lying flat on their backs with arms by the side. DXA scans were performed and analyzed using pediatric software (version 1.5e), as previously described (8, 22). On the day of each test, the DPX-L was calibrated using the procedures provided by the manufacturer. Height was measured without shoes using a stadiometer. Weight was measured in light clothing on an electronic scale.

Hormone assays. After overnight fasting, three samples of blood were collected over a period of 40 min. The blood samples were centrifuged at 1800 g for 10 min at 4°C. Sera were separated, pooled, and placed in cryovials, and all samples were stored at -85° until assayed for hormones. DHEAS, androstenedione, and estrone-sulfate were analyzed in duplicate by radioimmunoassay (Diagnostic System Laboratories, Webster, TX) and estradiol was analyzed in duplicate by double antibody radioimmunoassay (Diagnos-

tic Products Corporation, Los Angeles, CA). Testosterone was analyzed in duplicate by immunoassay (Coat-A-Count Total Testosterone, Diagnostic Products Corporation, Los Angeles, CA). In our laboratory, the limit of detectability and intra-assay coefficients of variation (CV) are as follows: DHEAS, 2.3 $\mu\text{g/dl}$ and 11.7% at 63% bound (23.5 $\mu\text{g/dl}$); androstenedione, 0.03 ng/ml and 11.0% at 55% bound (1.14 ng/ml); estrone-sulfate, 0.05 ng/ml and 6.2% at 62% bound (0.46 ng/ml); estradiol, 4.2 pg/ml and 3.6% at 51% bound (52.0 pg/ml); and testosterone, 11.8 ng/dl and 2.7% at 62% bound (119.3 ng/dl). The mean interassay coefficients of variation are as follows: DHEAS, 10% at 54% bound (36.0 $\mu\text{g/dl}$), 9.1% at 26% bound (191.1 $\mu\text{g/dl}$), 2.5% at 13% bound (630.0 $\mu\text{g/dl}$); androstenedione, 7.1% at 53% bound (1.05 ng/ml) and 5.1% at 19% bound (5.98 ng/ml); estrone-sulfate, 2.7% at 58% bound (0.59 ng/ml); estradiol, 7.2% at 48% bound (60.3 pg/ml) and 3.1% at 31% bound (159.1 pg/ml); and testosterone, 11.4% at 70% bound (65.4 ng/dl), 6.8% at 40% bound (344.4 ng/dl), and 7.5% at 30% bound (634.5 ng/dl).

Statistics. Differences in physical characteristics, body composition, energy expenditure, and serum concentrations of hormones among African American and Caucasian boys and girls were examined using a two-way (gender and ethnic groups) analysis of variance (ANOVA). Student *t*-tests were used to explore possible interaction effects between gender and ethnicity. Fat mass and hormone variables were log transformed in order to achieve normality of distribution. The correlations between energy expenditure variables (total, resting, and activity-related energy expenditure) and hormone concentrations were examined with Pearson correlation analysis. A first-order partial correlation was conducted for energy expenditure and hormone concentrations after controlling for

fat-free mass. Pearson correlations among hormone variables were also examined. Multicollinearity was tested by Pearson correlation analysis for all the independent variables ($r < 0.95$) before proceeding with multiple regression analyses. In multiple regression models, each component of energy expenditure was first examined as a dependent variable with fat-free mass, ethnicity, and gender as independent variables. Serum concentrations of DHEAS, androstenedione, and estrone-sulfate were then included in each model. Data were analyzed using SAS software version 6.10 (Carey, NC), with a significance level set at $P < 0.05$ for all tests.

Results

Subject characteristics. The characteristics of the children, including energy expenditure components and hormone concentrations, are presented in Table 1. Caucasian children were older than African American children ($P = 0.01$). The girls had greater fat mass ($P = 0.03$). There were significant interaction effects between gender and ethnicity for fat-free mass ($P = 0.01$) and resting energy expenditure ($P = 0.03$). This indicates that the gender differences for fat-free mass and resting energy expenditure were not consistent for Caucasians and African Americans, and ethnic differences for fat-free mass and resting energy expenditure were not consistent for boys and girls. Student *t*-tests (data not shown) showed that gender difference in fat-free mass was significant for African Americans ($P = 0.003$), but not for Caucasians ($P = 0.5$). Ethnic difference in fat-free mass was significant for girls ($P = 0.04$), but not for boys ($P = 0.4$). The same pattern was observed for resting energy expenditure (data not shown). There was no significant effect of gender

Table 1. Subject characteristics, energy expenditure, and hormone levels

	Caucasian boys (<i>n</i> = 30)	Caucasian girls (<i>n</i> = 18)	African American boys (<i>n</i> = 34)	African American girls (<i>n</i> = 30)	Two - Way ANOVA (<i>P</i> value)
Age (y)	8.5 ± 1.4 (5.6, 11.0)	8.7 ± 1.4 (5.9, 11.0)	8.0 ± 1.6 (5.1, 10.9)	7.7 ± 1.6 (4.7, 10.2)	Ethnicity (0.01), gender (NS)
Weight (kg)	34.4 ± 10.6 (17.0, 63.9)	40.5 ± 17.4 (22.2, 84.4)	34.9 ± 12.4 (19.8, 68.4)	31.6 ± 11.2 (14.1, 58.0)	NS
Height (cm)	132.1 ± 10.8 (110, 156)	133.7 ± 12.7 (117.0, 162.5)	133.0 ± 10.6 (113.0, 153.0)	128.4 ± 12.1 (104.8, 155.0)	NS
Body mass index (kg/cm ²)	19.3 ± 3.7 (13.9, 26.2)	19.2 ± 4.2 (11.8, 29.2)	21.8 ± 5.2 (15.1, 32.0)	18.7 ± 4.3 (12.8, 28.3)	NS
Fat-free mass (kg)	22.4 ± 4.7 (12.4, 35.6)	23.6 ± 6.9 (14.5, 41.3)	23.5 ± 5.3 (14.6, 35.5)	19.6 ± 4.6 (10.6, 29.1)	Ethnicity, gender (NS), interaction (0.01)
Fat mass (kg)	9.4 ± 6.2 (2.6, 24.0)	14.8 ± 10.7 (4.2, 44.7)	9.6 ± 7.3 (1.8, 30.8)	10.5 ± 7.0 (1.9, 26.3)	Ethnicity, interaction (NS), gender (0.03)
Total energy Expenditure (kcal/d)	1697 ± 408 (1178, 2975)	1853 ± 418 (1235, 2393)	1,741 ± 385 (990, 2721)	1680 ± 445 (940, 2585)	NS
Resting energy expenditure (kcal/d)	1,335 ± 243 (919, 1915)	1,351 ± 252 (1,027, 2088)	1,343 ± 282 (930, 2116)	1,156 ± 191 (869, 1610)	Ethnicity (0.05), gender (NS), interaction (0.03)
Activity-related energy expenditure (kcal/d)	289 ± 324 (-304, 1216)	330 ± 283 (55, 784)	312 ± 238 (-213, 744)	353 ± 247 (-23, 1007)	NS
DHEAS (μg/dl)	44.9 ± 40.0 (5.6, 178.5)	45.9 ± 31.7 (10.0, 110.8)	53.5 ± 39.3 (5.9, 178.4)	37.7 ± 28.5 (5.6, 110.2)	NS
Androstenedione (ng/ml)	0.5 ± 0.4 (0.04, 1.5)	0.6 ± 0.3 (0.1, 1.2)	0.6 ± 0.3 (0.1, 1.5)	0.6 ± 0.4 (0.1, 1.5)	NS
Estrone-sulfate (ng/ml)	0.5 ± 0.3 (0.1, 1.7)	0.6 ± 0.4 (0.1, 1.5)	0.5 ± 0.3 (0.1, 1.1)	0.5 ± 0.3 (0.2, 1.3)	NS

Data are means ± SD with range in parenthesis. NS, not significant.

or ethnicity in serum concentrations of DHEAS, androstenedione, and estrone-sulfate.

These effects remained insignificant after adjusting for age (data not shown).

Correlations. Pearson correlation and first-order partial correlation coefficients for energy expenditure and hormones with and without adjustment for fat-free mass are presented in Table 2.

Table 2. *Pearson and first-order correlation coefficients for energy expenditure components and hormones^a with and without adjustment for fat-free mass*

	Resting energy expenditure (<i>n</i> = 112)		Total energy Expenditure (<i>n</i> = 76)		Activity-related energy expenditure (<i>n</i> = 76)	
	<i>r</i> ^b	Partial <i>r</i> ^c	<i>r</i>	Partial <i>r</i>	<i>r</i>	Partial <i>r</i>
Fat-free mass	0.76***		0.72***		0.33**	
DHEAS	0.50***	-0.10	0.45***	-0.06	0.19	-0.05
Androstenedione	0.33***	-0.06	0.41***	0.06	0.23*	0.07
Estrone-sulfate	0.56***	-0.02	0.50***	0.05	0.21	-0.02

P* < 0.05; *P* < 0.01; ****P* < 0.001. ^aAll hormone variables are log transformed. ^b*r* is the correlation coefficient. ^cPartial *r* is the correlation coefficient after adjusting for fat-free mass.

Resting energy expenditure was significantly correlated with serum concentrations of DHEAS (*r* = 0.50, *P* = 0.0001), androstenedione (*r* = 0.33, *P* = 0.0005), and estrone-sulfate (*r* = 0.50, *P* = 0.0001). Total energy expenditure was also significantly correlated with serum concentrations of DHEAS (*r* = 0.45, *P* = 0.0001), androstenedione (*r* = 0.41, *P* = 0.0001), and estrone-sulfate (*r* = 0.50, *P* = 0.0001). Only androstenedione

was significantly correlated with activity-related energy expenditure ($r = 0.23$, $P = 0.04$). After controlling for fat-free mass, all these correlations became non-significant (Table 2). DHEAS, androstenedione, and estrone-sulfate were significantly correlated with each other ($P = 0.0001$, not shown).

Associations between hormones and energy expenditure. The relationships between hormones and components of energy expenditure are presented in Table 3.

Table 3. *Multiple regression models of the relationship between components of energy expenditure and ethnicity, gender, fat-free mass, fat mass, and various hormones*

	Independent variables		
	REE	TEE	AEE
Intercept	941.7 \pm 217.7***	1031.7 \pm 570.3	-105.8 \pm 529.4
Ethnicity ^a	-27.7 \pm 24.4	33.3 \pm 65.4	54.9 \pm 60.7
Gender ^b	-100.9 \pm 27.5***	-124.2 \pm 82.6	-40.3 \pm 76.7
Fat-free mass (kg)	26.4 \pm 3.8***	37.0 \pm 3.8*	8.1 \pm 10.9
Log (fat mass) (kg)	292.0 \pm 57.5***	619.5 \pm 196.2**	340.0 \pm 182.1
Log (DHEAS) (μ g/dl)	-151.2 \pm 93.6	-254.3 \pm 242.6	-53.1 \pm 225.2
Log (androstenedione) (ng/ml)	32.9 \pm 55.5	183.5 \pm 144.5	133.4 \pm 134.1
Log (estrone-sulfate) (ng/ml)	197.3 \pm 118.7	175.7 \pm 320.9	-122.4 \pm 297.9
Model R^2 (P value)	0.76 (0.0001)	0.55 (0.0001)	0.10 (0.05)

Data are expressed as parameter estimate \pm SE. TEE, total energy expenditure; REE, resting energy expenditure; AEE, activity-related energy expenditure. * $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$. Fat mass and hormone variables are log transformed. ^aEthnicity is coded as 1 = Caucasian, 2 = African American. ^bGender is coded as 1 = boys, 2 = girls.

There was no ethnic difference in any component of energy expenditure, after adjusting for hormone concentrations and body composition. Serum concentrations of DHEAS, androstenedione, and estrone-sulfate were not independently related to any component of energy expenditure after adjusting for ethnicity, gender, fat-free mass, and fat mass.

Discussion

In our current study, we did not observe ethnic differences in any component of energy expenditure in African American and Caucasian prepubertal children, before or after adjusting for concentrations of hormones and body composition. Therefore, unlike in other published studies, energy expenditure does not differ with ethnicity in our group of children.

Maturation may be related to changes of energy expenditure through changes in the quality and composition of fat-free mass (30). Therefore, it is conceivable that subtle and continuous changes in maturation, not detected by physical examination, may influence the quality of fat-free mass and thereby energy expenditure. In this study, fat-free mass explained the largest portion of the variance in energy expenditure. Hormones had strong correlations with energy expenditure ($P < 0.001$); however, after adjusting for fat-free mass, all significant associations disappeared. These findings indicate that the possible association between energy expenditure and ethnicity, if influenced by hormonal indices, could be potentially masked by fat-free mass. In contrast to the present results, previous studies in adult women have shown positive associations between DHEAS and androstenedione and energy expenditure, after adjusting for fat-free mass (1, 27). In the present study, even after obese children (defined as $\geq 30\%$ body fat) were excluded from

the analyses, there was no significant ethnic difference in any component of energy expenditure after adjusting for body composition and hormone concentrations (data not shown). The reason we did not detect an ethnic difference in any component of energy expenditure or an independent association between hormone concentration and energy expenditure might have been due to the low concentrations of hormones in prepubertal children. Therefore, future longitudinal studies are warranted to examine the associations between changes in energy expenditure, hormone concentrations, and body composition.

One of the reasons that hormones need to be considered when examining energy expenditure in children is the timing difference of maturation between African American and Caucasian children. Both African American boys and girls were reported to enter puberty earlier than their Caucasian counterparts (23). In a pediatric office setting (11), 27.2% African American and 6.7% Caucasian girls had evidence of pubertal development at age 7; 48.3% African Americans and 14.7% Caucasians had breast and pubic hair development at age 8. On average, African American girls began puberty at 8 to 9 yr of age, about 1 or 2 yr earlier than did Caucasian girls.

In the prepubertal stage, the ethnic difference in maturation may be apparent in different adrenal hormone concentrations in African American compared with Caucasian children. If African American children are more mature at any given age, higher concentrations of hormones may be expected in African American than Caucasian prepubertal children. A previous study of children through puberty found estradiol was significantly higher in African American than Caucasian boys after adjusting for pubertal stage, age, height, weight, and ponderosity index (weight/height^3) (23). However, androstenedione concentrations were found to be 29.4% and 20% lower in African American boys and

girls than Caucasian counterparts, respectively (23). Because changes in fat-free mass and the association between changes of maturation stage and body composition were not considered in this earlier study (23), we cannot rule out the possibility of higher hormone concentrations in African American children. No differences in concentration of DHEAS, androstenedione, or estrone-sulfate between African American and Caucasian boys and girls were found, even after adjusting for age (data not shown). Therefore, present results suggest that more advanced maturation may not necessarily be reflected in different or higher hormone concentrations in African American prepubertal children.

DHEAS and androstenedione were examined because they are strong hormone correlates of pubertal development (21, 23). The relationship between these hormones and energy expenditure, particularly with respect to potential ethnic differences, has not been previously examined in children. DHEAS was positively correlated with resting energy expenditure ($P < 0.01$) in women (1). Androstenedione explained 4% of the variance in sleeping energy expenditure (1), and 3% of the variance in resting energy expenditure after adjusting for body composition in women (27). These results suggest that androgens may affect processes that ultimately increase energy expenditure. However, this effect is not shown in this group of prepubertal children. The absence of association between total or resting energy expenditure and hormone concentration cannot be explained by an insufficient sample size. Power analyses (SPSS Sample Power 1.0, New York, 1997) show that our multiple regression models had sufficient power to detect 99% of the variance, which is more than the observed variance in total (55%) and resting energy expenditure (76%) given the sample size and number of variables. However, the absence of associa-

tion between activity-related energy expenditure and hormone concentrations may have suffered from an insufficient sample size ($n = 76$, $r^2 = 0.49$).

The mechanism of the influence of adrenal hormone action on energy expenditure observed previously is not well understood. Previous studies have shown that testosterone has a protein anabolic effect (17, 18). Testosterone deficiency is related to decreased whole body protein anabolism and reduced resting energy expenditure, which results from decreased lipid oxidation and fat-free mass in males (17). On the other hand, testosterone administration has been shown to increase muscle mass and muscle protein synthesis in prepubertal boys (18). These data suggest that adrenal androgens, such as DHEAS and androstenedione, may be related to growth and deposition of lean mass, which may in turn influence energy expenditure.

Another possible explanation for ethnic differences in energy expenditure is socioeconomic status (SES). Lower SES has been hypothesized to be associated with psychosocial stress (29), which is associated with elevated cortisol concentrations (25). In our study, African American children had significantly lower SES status than Caucasian children (unpublished data). However, in multivariate analyses, SES was not related to any component of energy expenditure. The lack of association between SES and energy expenditure may have suffered from an insufficient power due to the small sample size ($n = 44$) in SES multiple regression models.

Conclusions

Hormonal indices of maturation did not influence energy expenditure or the relationship between ethnicity and energy expenditure in this group of prepubertal African

American and Caucasian children. Ethnic differences in hormone concentrations may be more apparent when children go through puberty. Future longitudinal investigation on maturation-related changes in energy expenditure is warranted.

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**A LONGITUDINAL STUDY OF BODY COMPOSITION AND RESTING ENERGY
EXPENDITURE DURING PUBERTY IN AFRICAN AMERICAN AND
CAUCASIAN CHILDREN AND ADOLESCENTS**

by

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Format adapted for dissertation

Abstract

Pubertal maturation is associated with notable physiological changes. The purpose of this study was to examine longitudinally how pubertal maturation is associated with fat mass (FM), lean mass (FFM), and resting energy expenditure (REE) and how these relationships differ by ethnicity and gender. This sample included 92 Caucasian children (28 boys and 64 girls, aged 4.9 to 12.0 yr) and 64 African American children (26 boys and 38 girls, aged 4.6 to 12.1 yr) from Birmingham, Alabama. Children had 2 to 5 yr of annual measurements in FM, FFM, and REE. Tanner stage ranged from 1 to 5. Mixed model repeated measures analyses were used to test the relationships between FM, FFM, and REE with increasing age and Tanner stage among ethnic and gender groups. Relative FM decreased from Tanner stage 1 to 3, 4, and 5, but not to 2. FM did not differ significantly by ethnicity or gender. FFM increased from Tanner stage 1 to subsequent Tanner stages. FFM was higher in African Americans than in Caucasians, and in boys than in girls. Compared with Caucasians, African Americans had a higher relative limb lean mass and lower relative trunk lean mass. REE decreased with increasing age and Tanner stage after adjusting for ethnicity, gender, FM, and FFM. The decrease in REE was significant from Tanner stage 1 to 3, 4, and 5, but not to 2. After adjusting for age, Tanner stage, FM, and FFM, REE was significantly higher in Caucasians than in African Americans, and in boys than in girls. In conclusion, there were ethnic and gender differences in REE after adjusting for age, Tanner stage, FM, and FFM. In our cohort, an ethnic difference in REE appeared after Tanner stage 2, and it was not explained by FFM distribution. Whether earlier onset of puberty and the associated decline in REE contribute to greater prevalence of obesity among African Americans remains to be determined.

Introduction

Puberty is a period of development marked by maturation of secondary sexual characteristics along with dramatic changes in hormone concentrations, body composition, and energy partitioning (46). The process of pubertal maturation is not linear, but proceeds in patterns (11). Growth changes from a stable and linear pattern in childhood to a pattern of accelerating velocity in puberty (7). These changes exhibit a sexually dimorphic pattern. Girls enter puberty earlier than boys. The growth spurt usually occurs in Tanner breast stage 2 in girls and Tanner stage 3 in boys (46). Lean mass (FFM) begins to increase during early puberty in both boys and girls (4, 46). In boys, FFM increases continuously throughout puberty. Fat mass (FM) remains constant or declines, accompanied by increased skeletal growth. Percentage FFM increases and percentage FM decreases. In girls, both FM and FFM increase with increasing pubertal stage. Relative FM and FFM do not change significantly. A cross-sectional study in 403 Caucasian children (4-20 yr of age) (6) reported significant differences in body composition in different maturation stages. Tanner stage was positively related to FFM in both boys and girls, and to FM and percentage FM in girls. In boys, percentage FM was significantly lower in Tanner stage 3 compared with Tanner stage 1 and in Tanner stage 5 compared with stage 3.

Because of the dynamic changes in body composition and hormonal status, energy metabolism may also change during pubertal maturation. Basal and resting energy expenditure (REE) increase during pubertal development because of the increase in body weight, both in FM and FFM (5). However, it is not known how REE changes with pubertal maturation and how it differs by ethnicity and gender independent of body compo-

sition. Morrison et al. (35) showed a negative effect of maturation stage on REE after adjusting for body size in early pubertal girls. In contrast, other cross-sectional studies (4, 34) have suggested that age, but not pubertal maturation stage, contributes to the decrease in REE independent of differences in FM and FFM in both boys and girls.

One explanation for the effect of maturation on REE is the different metabolic contributions of organ and muscle mass during pubertal growth. In an average human, it has been shown that organ mass (brain, liver, heart, and kidney) accounts for two thirds of metabolic energy, although total organ mass is less than 6% of body weight (22). The rest of the energy expenditure is mainly accounted for by skeletal muscle, which comprises 40-50% total body weight (22). The metabolic rate of the organs is 15-25 times that of muscle. The contribution of organs to REE (60%) is 2.4 times higher than that of muscle (20-25%) per unit mass (13, 22). A recent study (24) has concluded that organ size contributes significantly to, and explains the residual variance in, REE in young adults.

Trunk lean mass (TLM) and limb lean mass (LLM) have been suggested to serve as surrogates for mass of organs and muscles, respectively, that are not easily measured (21, 40). LLM is the main component of total lean mass and is the sum of skeletal muscle in arms and legs as defined in DXA scan analysis. TLM, because of the organ component, may contribute more to REE than LLM, which reflects mainly skeletal muscle (14).

African Americans are reported to have a greater total LLM than Caucasians after adjusting for height, body weight, age, and gender ($P = 0.0001$) (14). Thus, we hypothesized that African Americans have a higher muscle mass and a lower organ mass relative

to weight, a distribution that may contribute to a lower REE when compared with Caucasians (23).

Earlier cross-sectional studies of body composition and REE during puberty involved comparison of different subjects in different pubertal stages. Because growth cannot be assessed adequately with one measurement at one time, measurement should be done within the same subject over time. Because African American children begin puberty earlier than Caucasian children (20, 35, 36), the influence of maturation on body composition and REE may be apparent at a younger age in African Americans. To our knowledge, no longitudinal studies have examined the changes in body composition and REE during pubertal maturation in both African American and Caucasian boys and girls.

The hypotheses of this study were as follows: 1) In our cohort, maturation would have a positive relationship with FFM and a negative relationship with REE after adjusting for body composition, and 2) an ethnic difference in REE would become more apparent as the children matured. The first objective of this study was to examine FM, FFM, and REE across age and Tanner stage in African American and Caucasian boys and girls. The second objective was to identify the stage of puberty when differences in body composition and REE occurred. The third objective was to examine whether there was a difference in FFM distribution in African American children compared with Caucasian children and whether it explained any observed ethnic difference in REE.

Methods

The sample included 92 Caucasian children (28 boys and 64 girls, aged 4.9 to 12.0 yr) and 64 African American children (26 boys and 38 girls, aged 4.6 to 12.1 yr)

from Birmingham, Alabama. They participated in a longitudinal study investigating how intra-abdominal fat in children influences disease risk factors. The children were recruited by using newspaper advertisements and by word of mouth. Ethnicity was defined on the basis of the self-ascribed ethnicity status of children's parents and grandparents. The eligibility criteria were 1) >4 years of age; 2) not taking medications known to affect body composition or physical activity (e.g., prednisone, ritalin, growth hormone); 3) not previously diagnosed with syndromes known to affect body composition, fat distribution, or both (e.g., Cushing's syndrome, Down's syndrome, type I diabetes, or hypothyroidism); and 4) not diagnosed with any major illnesses since birth. We have also previously reported energy expenditure (41), body composition (17), and aerobic fitness (44) data in these children when they were prepubertal.

The nature, purpose, and possible risks of the study were carefully explained to the parent before obtaining consent. This study was approved by the Institutional Review Board at the University of Alabama at Birmingham, and parents provided informed consent before testing began. All measurements were performed at the General Clinical Research Center (GCRC) or in Department of Nutrition Sciences at the University of Alabama at Birmingham between 1994 and 1999.

General Outline of Protocol

Children were admitted to the GCRC in the late afternoon for an overnight visit. On arrival, anthropometric measurements were obtained, and dinner was served (~1700). An evening snack was allowed, and after 2000, only water and energy-free, noncaffeinated beverages were permitted until after the morning testing. Tanner stage of

the children was examined by a pediatrician on the basis of breast stage and pubic hair development in girls (29) and genitalia development in boys (30), according to the criteria of Tanner. On the following morning upon awakening at 0500, after the overnight fast, resting metabolic rate was assessed by indirect calorimetry, using a Deltatrac Metabolic Monitor (Sensormedics Corp., Yorba Linda, CA). During testing, all subjects were instructed to lie as still as possible and remain awake. An adult-size canopy hood was used to collect the expired air. After a 10-min equilibration period, data on oxygen consumption and carbon dioxide production were collected continuously for 20 min. REE was calculated using the equation of de Weir (9). The subjects were allowed to urinate if necessary. If the subjects urinated, they were asked to rest for 15 min before resting metabolic rate measurement was taken.

Two weeks later, the children arrived at the Department of Nutrition Sciences at 0700 in the fasted state. Total and regional body compositions were measured by dual-energy X-ray absorptiometry (DXA) using a Lunar DPX-L densitometer previously validated in the pediatric body weight range (16, 38). Subjects were scanned in light clothing while lying flat on their backs with arms by the sides. DXA scans were performed and analyzed using pediatric software (version 1.5e), as previously described (16, 38). On the day of each test, the DPX-L was calibrated using the procedures provided by the manufacturer. Height was measured without shoes using a stadiometer. Weight was measured in light clothing on an electronic scale.

The participants came annually to the Department of Nutrition Sciences for measurements of body composition and REE. The start date for the cohort ranged from 1994 to 1999. In this analysis, the number of measurements for each child ranged from 2 to 5,

and the total follow-up duration for each child ranged from 1 to 4 yr. During the second year, 30% of girls were not examined.

Statistics

Means and standard deviations were calculated for each visit for age, weight, height, body mass index (BMI), FM, FFM, and REE. Analysis of variance (ANOVA) models were used to determine ethnic and gender differences in these variables in each visit year. Mixed model repeated measures analyses (PROC MIXED) were used to examine relationships between body composition and REE with increasing age and Tanner stage among ethnic and gender groups.

FM and FFM were examined across age and Tanner stage, adjusting for ethnicity and gender with these simplified models: $FM = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender}$; $\text{Relative FM} = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender} + \text{FFM}$; and $FFM = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender}$.

Absolute and relative LLM and TLM were examined using the following models: $LLM (\text{absolute}) = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender}$; $LLM (\text{relative}) = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender} + \text{TLM}$; $TLM (\text{absolute}) = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender}$; and $TLM (\text{relative}) = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender} + LLM$.

Relative LLM and TLM were obtained by adjusting each for the other in order to take body size into account. REE was examined with the model $REE = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender} + FM + FFM$.

Because of highly significant correlation between LLM and TLM ($r = 0.94$, $P < 0.0001$), a variable LIMTRK was created, based on the principle component method, to

describe FFM distribution: $LIMTRK = 0.707 \times LLM + 0.707 \times TLM$. LIMTRK contained all the information on both LLM and TLM and eliminated multicollinearity caused by the significant correlation between the two. The contribution of FFM distribution to REE was then examined using the model $REE = \text{Age} + \text{Tanner stage} + \text{Ethnicity} + \text{Gender} + \text{FM} + LIMTRK$.

Fixed class variables in these models were visit year, subject identifier, ethnicity, gender, and Tanner stage. Random variables were age, FM, FFM, LLM, and TLM. Because the changes from one Tanner stage to the next may not be linear, we recoded five Tanner stages into three dummy variables. Tanner stages 4 and 5 were combined because of small sample size in Tanner stages 4 and 5. These dummy variables were used to compare the difference in body composition and REE between Tanner stage 1 and Tanner stages 2, 3, and combined 4 and 5. Initially, Schwarz's Bayesian criterion was used to select an appropriate covariance structure suited for each mixed model (27). Three covariance structures were tested by PROC MIXED. Compound symmetry indicates that the correlation parameter estimate is the same regardless of the length of time interval between the measurements (28). This is not valid because measurements close in time have a higher correlation than measurements far in time. Unstructured covariance structure does not have a mathematical pattern imposed on the correlation matrix. Therefore, it does not assume a same correlation parameter estimate between measurements. Autoregressive covariance structure is built so that measurements close in time have a higher correlation than those that are far in time. These covariance structures were tested, and the one with the largest test value was used in the mixed models for FM, FFM, LLM, TLM, and REE. *F*-tests were used to determine the overall fixed effects of the models.

Overall least squared means plus/minus the standard errors of the least squared means were reported for each ethnic-gender subgroup. All statistical analyses were conducted using SAS software version 7.00 (Carey, NC), with a significance level set at $P < 0.05$ for all tests.

Results

Characteristics. The distribution of the participants in 5 yr is presented in Table 1. In the first visit year, there were 156 participants. Years of follow-up ranged from 2 to 5. During the second year, 30% of girls were not evaluated.

Table 1. *Distribution of participants in 5 years*

Visit Year	Caucasian boys	Caucasian girls	African American boys	African American girls	Total number of subject visits
1	28	64	26	38	156
2	25	40	22	31	118
3	21	61	22	32	136
4	16	37	20	25	98
5	0	21	16	21	58
Total number of observations	90	223	106	147	566

Table 2 shows the distribution of Tanner stage in each year. Tanner stage was not examined in four cases. During the first visit year, the majority of the children were pre-pubertal. The number of children who reached Tanner stage 4 or 5 during the five years of this study was relatively small.

Table 2. *Frequency of the Tanner stages of participants in 5 years*

Visit Year	Tanner stages				Total
	1	2	3	4 & 5	
1	145	9	1	1	156
2	89	21	6	1	117
3	60	57	14	4	135
4	26	44	22	5	97
5	10	13	16	18	57
Total number of observations	475	144	59	29	562

Table 3 shows the characteristics of the participants in up to 5 visit years. There was no significant ethnic or gender difference in age or height for all years. African American children were heavier than Caucasian children during the third visit year ($P = 0.03$). BMI was not different until the fourth and fifth year, with a higher BMI in African American children than in Caucasian children ($P < 0.05$). Body composition data in up to 5 visit years are shown in Table 4. There was no ethnic or gender difference in FM during 5 visit years. Caucasian children had significantly lower FFM than African American children during the third and fourth visit years ($P < 0.05$). Boys had significantly higher FFM and REE than girls during the first and second years ($P < 0.05$). African American children had a higher LLM than Caucasian children ($P < 0.01$), and boys had a higher LLM than girls ($P < 0.02$) only during the first visit year. There was no significant ethnic difference in TLM during all the visit years. Boys had a higher TLM than girls only during the first two visit years ($P = 0.03$). REE was not statistically significantly different in African Americans and Caucasians in all years. Boys had a higher REE than in girls only during the first 2 visit years ($P < 0.005$).

Table 3. *Characteristics of participants during study period*

	Visit year	Caucasian boys	Caucasian girls	African American boys	African American girls	ANOVA** P values
Age (years)	1	8.5 ± 1.8 (5.6, 12.0)	8.3 ± 1.3 (4.9, 11.0)	7.7 ± 1.5 (5.1, 10.2)	8.0 ± 1.9 (4.6, 12.1)	NS
	2	9.6 ± 1.7 (6.7, 13.0)	9.3 ± 1.5 (5.8, 12.3)	8.8 ± 1.5 (6.1, 11.2)	9.0 ± 2.0 (5.7, 13.0)	NS
	3	10.2 ± 1.6 (7.6, 13.0)	10.3 ± 1.4 (7.0, 13.5)	10.0 ± 1.5 (7.1, 12.2)	10.1 ± 1.9 (6.5, 14.1)	NS
	4	11.1 ± 1.5 (8.6, 13.3)	11.1 ± 1.4 (8.0, 14.5)	10.6 ± 1.7 (8.1, 13.3)	10.9 ± 1.8 (7.5, 13.0)	NS
	5		12.1 ± 1.4 (9.5, 14.2)	11.5 ± 1.6 (9.1, 13.8)	12.1 ± 1.8 (8.5, 14.1)	NS
*Weight (kg)	1	33.3 ± 10.8 (16.2, 61.7)	30.9 ± 9.8 (16.0, 64.8)	34.9 ± 13.1 (19.9, 67.1)	32.6 ± 12.2 (14.0, 71.7)	NS
	2	38.2 ± 13.5 (17.4, 66.9)	34.2 ± 11.7 (16.5, 77.1)	40.0 ± 16.1 (21.3, 74.0)	37.8 ± 14.3 (16.0, 74.7)	NS
	3	40.0 ± 14.3 (19.5, 78.0)	49.4 ± 14.2 (18.7, 85.8)	47.6 ± 17.4 (28.0, 83.3)	45.2 ± 15.2 (16.5, 73.7)	Ethnicity: 0.03 Gender: NS
	4	42.4 ± 12.9 (21.3, 64.2)	43.5 ± 14.8 (19.6, 102.8)	49.3 ± 19.8 (26.7, 93.0)	48.9 ± 16.5 (18.5, 85.6)	NS
	5		48.5 ± 13.0 (25.6, 69.9)	54.3 ± 20.8 (27.5, 104.1)	55.7 ± 19.5 (20.6, 100.5)	NS
Height (cm)	1	131.8 ± 12.6 (110.0, 160.0)	129.8 ± 9.6 (104.0, 152.5)	131.6 ± 10.5 (113.0, 153.0)	129.3 ± 14.1 (101.5, 163.8)	NS
	2	139.1 ± 13.0 (114.3, 166.3)	135.6 ± 10.1 (109.0, 159.3)	136.3 ± 11.6 (119.0, 157.0)	137.1 ± 14.1 (109.0, 162.5)	NS
	3	142.9 ± 12.1 (119.5, 170.0)	142.5 ± 9.5 (114.0, 166.0)	144.5 ± 9.5 (129.3, 161.0)	144.4 ± 13.6 (115.0, 164.0)	NS
	4	147.8 ± 11.6 (124.5, 165.5)	147.9 ± 10.5 (120.0, 174.0)	147.1 ± 10.6 (132.0, 167.3)	149.4 ± 12.8 (119.4, 166.0)	NS
	5		152.0 ± 11.5 (125.0, 169.9)	153.8 ± 11.3 (136.3, 173.5)	155.8 ± 11.3 (123.5, 171.0)	NS
BMI (kg/cm ²)	1	18.6 ± 3.6 (13.4, 25.6)	18.0 ± 3.5 (14.0, 28.8)	19.6 ± 4.9 (12.2, 35.2)	19.0 ± 4.7 (12.7, 34.1)	NS
	2	19.4 ± 4.5 (12.9, 26.9)	18.2 ± 4.2 (13.1, 30.4)	20.9 ± 5.7 (14.9, 36.5)	19.5 ± 4.9 (13.1, 31.3)	NS
	3	19.0 ± 3.8 (13.2, 27.0)	19.5 ± 4.7 (13.8, 32.2)	22.3 ± 6.1 (16.2, 39.6)	21.0 ± 5.2 (12.5, 33.0)	Ethnicity: 0.02 Gender: NS
	4	19.1 ± 4.3 (13.0, 28.1)	19.5 ± 4.6 (13.5, 33.9)	22.2 ± 6.5 (15.3, 40.3)	21.4 ± 5.6 (13.0, 34.8)	Ethnicity: 0.03 Gender: NS
	5		20.9 ± 5.2 (14.4, 33.1)	22.3 ± 5.8 (14.8, 37.6)	22.6 ± 6.9 (13.5, 38.6)	NS

The number of visits ranged from 2 to 5 in Caucasian girls. African American boys and girls, and from 2 to 4 in Caucasians boys. *Weight was obtained from the DXA.

**ANOVA, analysis of variance. NS: not significant. The class variables in ANOVA models are ethnicity and gender. There was no significant interaction between ethnicity and gender among groups at any visit year.

Table 4. *Body composition and REE in study period.*

Variables	Visit year	Caucasian boys	Caucasian girls	African-American boys	African-American girls	ANOVA <i>P</i> values
FM (kg)	1	9.8 ± 6.2 (2.6, 24.0)	9.8 ± 5.9 (2.9, 32.6)	10.6 ± 8.2 (3.2, 31.7)	10.5 ± 6.8 (2.7, 29.2)	NS
	2	10.7 ± 8.3 (1.9, 26.3)	9.9 ± 7.2 (1.5, 33.8)	12.0 ± 10.9 (1.7, 37.4)	11.4 ± 8.1 (3.1, 31.3)	NS
	3	10.7 ± 7.8 (2.1, 31.0)	12.8 ± 9.1 (2.3, 44.7)	15.2 ± 11.9 (2.7, 39.2)	14.4 ± 9.0 (2.2, 31.5)	NS
	4	11.2 ± 8.0 (2.8, 29.8)	13.4 ± 9.4 (2.3, 48.8)	14.4 ± 13.3 (2.3, 46.6)	15.3 ± 10.4 (2.9, 38.4)	NS
	5		14.8 ± 11.7 (1.8, 48.6)	14.8 ± 13.2 (1.8, 48.6)	18.2 ± 13.1 (3.3, 46.9)	NS
FFM (kg)	1	21.1 ± 5.0 (10.6, 40.2)	20.1 ± 4.1 (12.5, 34.4)	22.9 ± 5.1 (14.6, 34.5)	20.8 ± 5.8 (10.6, 40.2)	Ethnicity: NS Gender: 0.01
	2	26.1 ± 6.0 (14.8, 38.9)	23.1 ± 4.8 (14.4, 41.3)	26.5 ± 5.5 (17.1, 35.3)	24.9 ± 7.0 (12.1, 40.4)	Ethnicity: NS Gender: 0.04
	3	27.8 ± 6.9 (16.7, 45.3)	26.2 ± 5.7 (15.8, 45.5)	30.7 ± 6.0 (22.5, 42.9)	29.0 ± 7.2 (13.6, 42.2)	Ethnicity: 0.01 Gender: NS
	4	29.6 ± 6.9 (17.7, 42.4)	28.6 ± 6.4 (16.6, 51.3)	33.0 ± 8.0 (23.2, 49.2)	31.5 ± 7.4 (14.7, 44.8)	Ethnicity: 0.03 Gender: NS
	5		31.9 ± 5.6 (20.3, 42.6)	37.4 ± 9.8 (24.5, 56.7)	35.8 ± 7.7 (16.4, 51.7)	NS
LLM (kg)	1	9.8 ± 3.0 (4.4, 17.1)	8.6 ± 2.1 (4.7, 15.7)	10.8 ± 2.7 (6.2, 16.0)	9.9 ± 3.2 (4.1, 20.8)	Ethnicity: 0.006 Gender: 0.02
	2	11.7 ± 3.4 (5.3, 19.0)	10.2 ± 2.6 (5.2, 19.4)	12.4 ± 3.0 (7.7, 17.2)	12.0 ± 3.9 (4.8, 20.6)	Ethnicity: 0.02 Gender: NS
	3	12.8 ± 4.1 (6.2, 23.1)	11.9 ± 2.9 (6.2, 20.2)	14.8 ± 3.0 (10.1, 20.1)	14.1 ± 4.0 (5.3, 21.1)	Ethnicity: 0.001 Gender: NS
	4	13.9 ± 3.7 (6.9, 20.4)	13.1 ± 3.1 (6.6, 22.5)	16.1 ± 3.9 (10.6, 25.0)	15.3 ± 4.0 (6.1, 21.6)	Ethnicity: 0.003 Gender: NS
	5		14.7 ± 2.8 (8.6, 20.2)	18.6 ± 4.8 (11.2, 26.2)	17.9 ± 4.0 (7.0, 24.8)	Ethnicity: 0.01 Gender: NS
TLM (kg)	1	10.2 ± 2.3 (5.8, 15.9)	9.4 ± 2.0 (5.8, 16.7)	10.0 ± 2.6 (6.6, 17.6)	9.0 ± 2.6 (4.6, 17.1)	Ethnicity: NS Gender: 0.03
	2	12.0 ± 2.7 (7.1, 17.7)	10.7 ± 2.3 (6.7, 19.5)	11.7 ± 2.6 (7.0, 16.0)	10.9 ± 3.1 (5.4, 17.1)	Ethnicity: NS Gender: 0.03
	3	12.5 ± 2.8 (7.8, 19.7)	12.0 ± 2.9 (7.1, 22.7)	13.7 ± 3.3 (10.0, 22.0)	12.7 ± 3.1 (6.0, 18.2)	NS
	4	13.3 ± 3.1 (8.2, 19.7)	13.0 ± 3.3 (7.6, 26.1)	14.5 ± 4.2 (9.6, 23.3)	13.9 ± 3.5 (6.4, 21.2)	NS
	5		14.6 ± 2.8 (9.0, 19.6)	16.1 ± 5.1 (10.3, 27.0)	15.6 ± 3.6 (7.1, 24.4)	NS
REE (kcal/day)	1	1336 ± 261 (927, 1933)	1204 ± 161 (837, 1887)	1329 ± 281 (938, 2136)	1199 ± 198 (837, 1777)	Ethnicity: NS Gender: 0.001
	2	1433 ± 218 (1104, 1849)	1267 ± 219 (813, 2050)	1409 ± 289 (869, 1970)	1290 ± 232 (863, 1653)	Ethnicity: NS Gender: 0.002
	3	1446 ± 233 (1111, 2186)	1387 ± 251 (859, 2192)	1487 ± 293 (1152, 2370)	1408 ± 240 (894, 1917)	NS
	4	1456 ± 291 (1027, 2015)	1419 ± 259 (807, 2141)	1408 ± 261 (1174, 2160)	1377 ± 250 (815, 1901)	NS
	5		1496 ± 310 (1027, 2181)	1503 ± 353 (968, 2255)	1474 ± 309 (938, 1941)	NS

Table 5 shows the results of the tests for goodness of fit of covariance structures used in the mixed models. Unstructured variance structure was used for the mixed models for FM, FFM, absolute LLM, absolute and relative TLM, and REE. Auto-regressive covariance structure was used for the mixed model for relative LLM.

Table 5. *Selection of the appropriate covariance structure for mixed models using Schwarz Bayesian criterion*

Mixed models	Compound symmetry	Unstructured	Auto-regressive
FM *	-1616.0	-1500.0	-1551.0
FFM **	-1338.3	-1266.7	-1279.0
Absolute LLM *	-964.4	-915.8	924.9
Relative LLM †	-740.0	-715.9	-713.0
Absolute TLM *	-1006.7	-925.0	-938.5
Relative TLM ††	-756.6	-693.2	-717.1
REE ***	-3481.7	-3476.8	-3482.9

*Adjusted for age, Tanner stage, ethnicity, and gender. **Adjusted for age, Tanner stage, ethnicity, and gender. ***Adjusted for age, Tanner stage, ethnicity, gender, FM, and FFM. †Adjusted for age, Tanner stage, ethnicity, gender, and TLM. ††Adjusted for age, Tanner stage, ethnicity, gender, and LLM

Body composition and REE by Tanner stages. Table 6 shows the differences of least square means and standard errors for FM, FFM, LLM, TLM, and REE between Tanner stage 1 and subsequent Tanner stages. Absolute FM did not vary significantly by Tanner stage after adjusting for age, ethnicity, and gender. FM adjusted for FFM decreased significantly from Tanner stage 1 to 3, 4 and 5 ($P < 0.05$), but not to 2. FFM increased significantly from Tanner stage 1 to subsequent Tanner stages ($P < 0.005$). LLM adjusted for TLM increased significantly from Tanner stage 1 to 3, but not to 2, 4, and 5, after adjusting for age, ethnicity, and gender. TLM adjusted for LLM increased

Table 6. *Changes in FM, FFM, LLM, TLM, and REE from Tanner stage 1 to subsequent Tanner stages.*

	Tanner stages	Least square means \pm SE	P values
Absolute FM (kg)	2	-0.1 \pm 0.3	NS
	3	-0.5 \pm 0.5	NS
	4 & 5	-1.0 \pm 0.9	NS
Relative FM (kg)*	2	-0.3 \pm 0.3	NS
	3	-1.7 \pm 0.6	0.003
	4 & 5	-2.3 \pm 1.0	0.02
FFM (kg)**	2	0.6 \pm 0.2	0.001
	3	3.0 \pm 0.3	0.003
	4 & 5	3.2 \pm 0.6	< 0.0001
LLM (kg)†	2	0.2 \pm 0.1	NS
	3	0.4 \pm 0.2	0.03
	4 & 5	0.4 \pm 0.3	NS
TLM (kg)‡	2	0.1 \pm 0.1	NS
	3	0.4 \pm 0.2	0.006
	4 & 5	0.5 \pm 0.3	0.04
REE (kcal/day)‡‡	2	-18.2 \pm 17.2	NS
	3	-75.2 \pm 29.0	0.01
	4 & 5	-223.4 \pm 46.5	< 0.0001

Tanner stage 1 was the reference group. * Adjusted for age, ethnicity, gender, and FFM. ** Adjusted for age, ethnicity, and gender. † Adjusted for age, ethnicity, gender, and TLM ‡ Adjusted for age, ethnicity, gender, and LLM. ‡‡ Adjusted for age, ethnicity, gender, FM, and FFM. NS, not significant.

significantly from Tanner stage 1 to 3, 4, and 5 ($P < 0.05$), but not to 2. The change in REE from Tanner stage 1 to stage 2 was not significant. The decreases in REE from Tanner stages 1 to 3 (75.2 kcal/day, $P = 0.01$) and from 1 to 4 and 5 (223.4 kcal/day, $P < 0.0001$) were significant after adjusting for age, ethnicity, gender, FM, and FFM.

Body composition and REE in ethnic or gender subgroups. Table 7 shows the least square means and standard errors for body composition and REE in ethnic-gender subgroups.

Table 7. *Least squares means and standard errors for FM, FFM, LLM, TLM, and REE in ethnic-gender subgroups*

	Caucasian boys	Caucasian girls	African American boys	African American girls	<i>P</i> values
Absolute FM (kg)*	12.2 ± 1.2	11.2 ± 1.0	12.7 ± 1.3	12.9 ± 1.1	NS
Adjusted FM (kg) **	10.5 ± 1.2	10.4 ± 1.0	10.6 ± 1.2	11.7 ± 1.1	NS
FFM (kg)*	30.0 ± 0.8	28.4 ± 0.6	32.1 ± 0.8	29.4 ± 0.7	Ethnicity: 0.01 Gender: 0.0002
Absolute LLM (kg) **	13.9 ± 0.4	13.1 ± 0.3	15.9 ± 0.4	14.5 ± 0.4	Ethnicity: < 0.0001 Gender: 0.0005
Relative LLM (kg) †	12.5 ± 0.3	11.9 ± 0.2	14.0 ± 0.2	13.6 ± 0.2	Ethnicity: < 0.0001 Gender: 0.0005
Absolute TLM (kg) **	13.8 ± 0.4	13.2 ± 0.3	14.0 ± 0.4	12.8 ± 0.4	Ethnicity: NS Gender: 0.001
Adjusted TLM (kg) †	12.1 ± 0.1	12.2 ± 0.1	11.4 ± 0.1	11.5 ± 0.1	Ethnicity: < 0.0001 Gender: NS
REE (kcal/day)***	1274 ± 38	1208 ± 33	1228 ± 38	1168 ± 31	Ethnicity: < 0.01 Gender: < 0.0001
REE (kcal/day)‡‡	1278 ± 37	1210 ± 33	1230 ± 38	1167 ± 31	Ethnicity: 0.003 Gender: < 0.0001

* Adjusted for age, Tanner stage. ** Adjusted for age, Tanner stage, and FFM.

*** Adjusted for age, Tanner stage, FM, and FFM. † Adjusted for age, Tanner stage, and TLM. ‡ Adjusted for age, Tanner stage, and LLM. ‡‡ Adjusted for age, Tanner stage, FM, and LIMTRK. NS, not significant.

There was no ethnic or gender difference in FM after adjusting for age, Tanner stage, and FFM. There were significant ethnic and gender differences in FFM after adjusting for age and Tanner stages ($P = 0.01$ and 0.0002 , respectively). Figure 1 shows the least squares means in body composition, adjusted for age and Tanner stage, in ethnic and gender subgroups. On average, FFM was higher in African Americans than in Caucasians (by 1.4 kg, $P = 0.01$) and in boys than in girls (by 2.1 kg, $P = 0.0002$). Relative LLM was higher in African Americans than in Caucasians (by 1.6 kg, $P < 0.0001$) and in boys than in girls (by 1.0 kg, $P = 0.0005$). Relative TLM was higher in Caucasians than in African

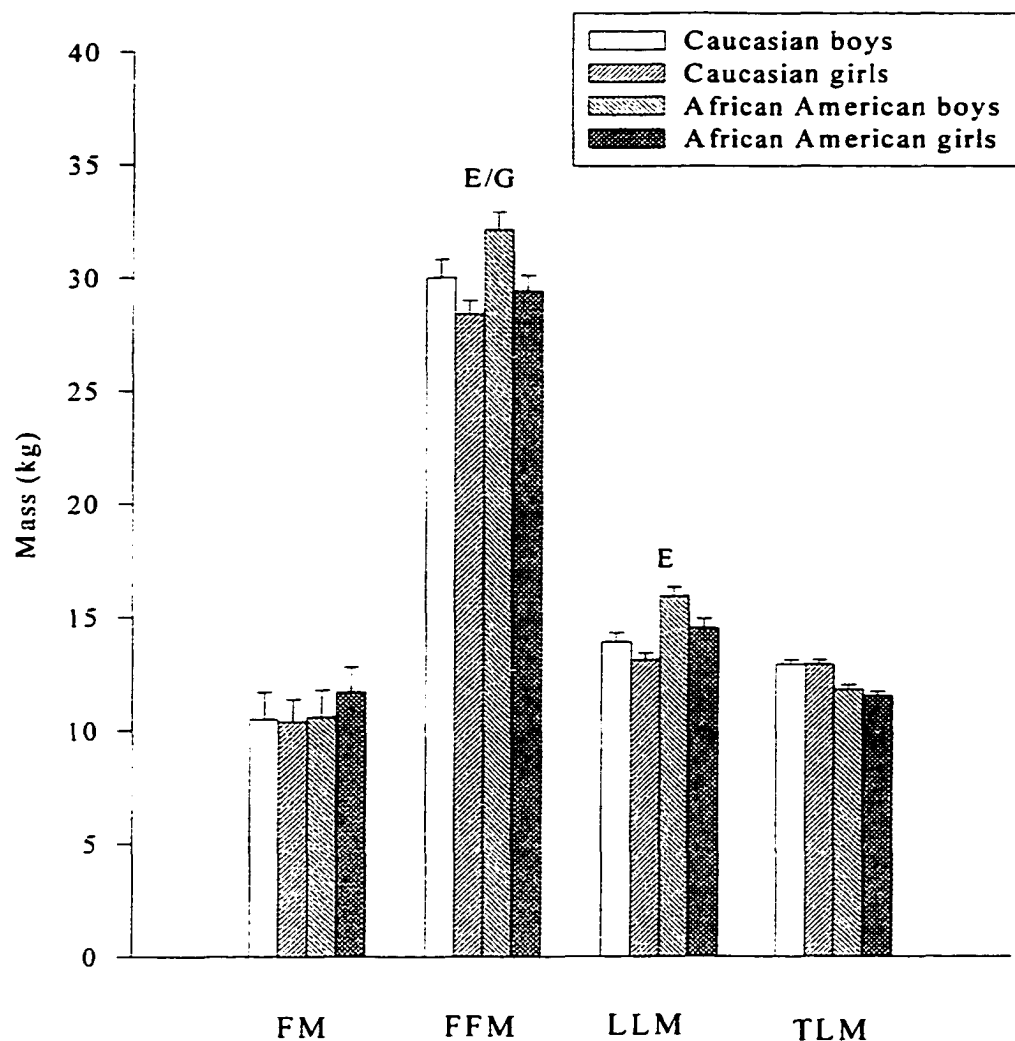


Fig. 1. Body composition adjusted for age and Tanner stage in African American and Caucasian children and adolescents. Tanner stage ranged from 1 to 5. FM, adjusted for FFM; LLM, adjusted for TLM; TLM, adjusted for LLM. E/G, significantly different by ethnicity and gender. E, significantly different by ethnicity.

Americans (by 1.0 kg, $P = 0.0005$). There were significant ethnic and gender differences for REE after adjusting for age, Tanner stage, FM, and FFM. Adjusted REE was higher in Caucasians than African Americans (by 41 kcal/day, $P = 0.001$) and in boys than in girls (by 63 kcal/day, $P = 0.0001$). REE adjusted for FM, LLM, and TLM (expressed as LIMTRK) showed the same pattern as the model adjusted for FM and FFM.

Body composition and REE in ethnic or gender subgroups during puberty. Figure 2 shows the gender-specific changes in FM and FFM during puberty. The change in relative FM from Tanner stage 1 to stages 4 and 5 was significantly different in boys and girls ($P < 0.005$). The amount of decrease from Tanner stage 1 to stages 4 and 5 was 5.4 kg in boys and 0.5 kg in girls. The increases in FFM from Tanner stage 1 to 2 and from 1 to 4 and 5 were significantly different in boys and girls ($P < 0.01$), but not from 1 to 3.

The amount of increase in FFM from Tanner stage 1 to 2 was not significant in boys and was 1.1 kg in girls. The amount of increase in FFM from Tanner stage 1 to stages 4 and 5 was 5.0 kg in boys and 3.5 kg in girls. There was no ethnic difference in the change in FM and FFM from Tanner stage 1 to subsequent Tanner stages.

The decrease in REE was significantly different in boys and girls from Tanner stage 1 to 2 ($P < 0.0001$), but not to other Tanner stages, after adjusting for age, ethnicity, FM, and FFM. There was no significant ethnic difference in the change in REE from Tanner stage 1 to subsequent Tanner stages.

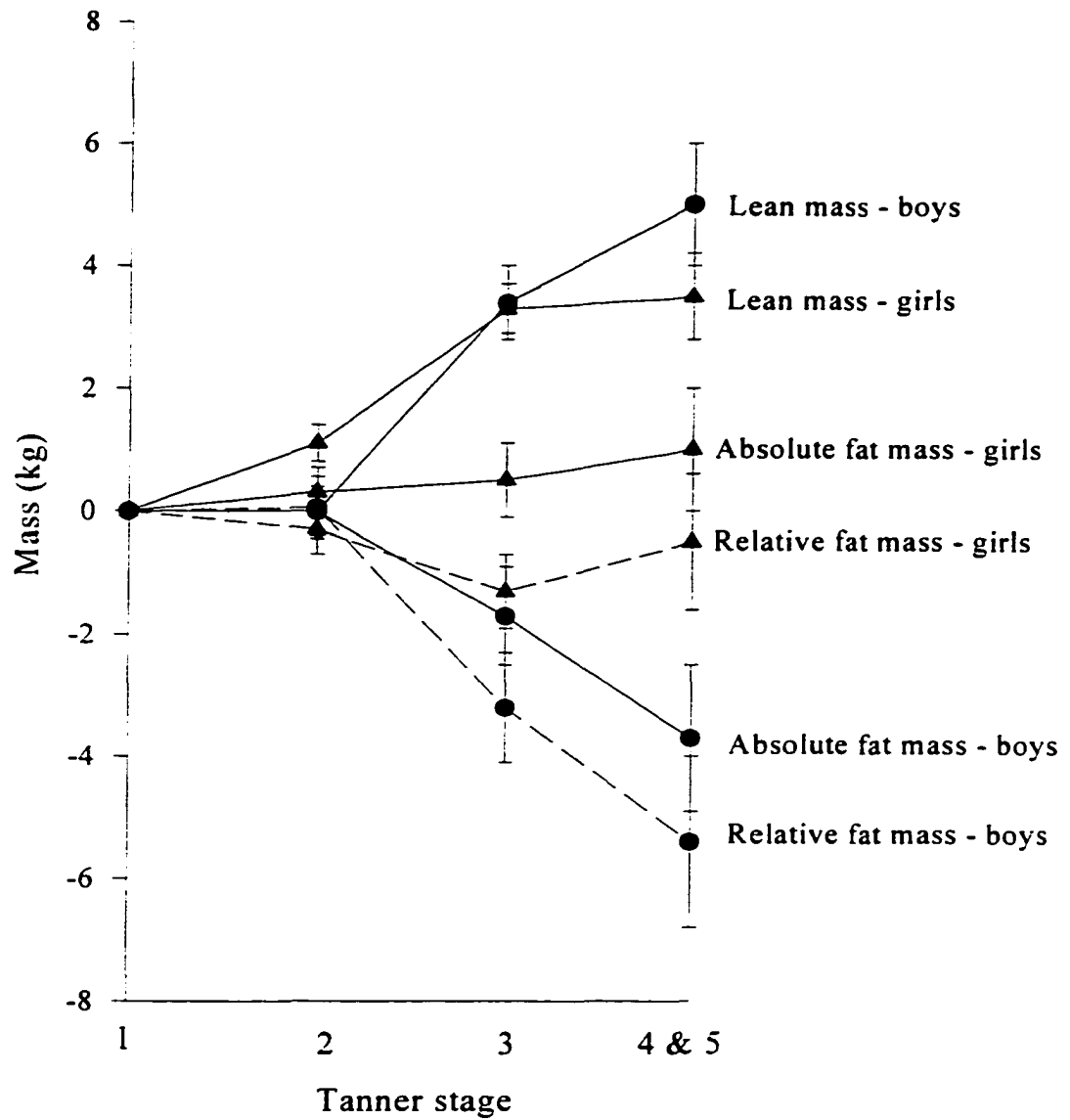


Fig. 2. Gender-specific changes in body composition during puberty, adjusted for age, Tanner stage, and ethnicity. Circles, boys; Triangles, girls. Solid lines, absolute value; broken lines, relative value. Relative FM, FM adjusted for FFM.

Discussion

This is the first longitudinal analysis to examine REE and body composition, particularly the distribution of FFM, during puberty in African American and Caucasian boys and girls. In this cohort, maturation was negatively related to the REE independent of FM and FFM after Tanner stage 2. African American children had a lower REE than their Caucasian counterparts after adjusting for age, Tanner stage, gender, FM, FFM or FFM distribution. The ethnic difference in REE became more apparent after Tanner stage 2.

In this study, we observed a positive relationship between pubertal stage and FFM, but not relative FM, after adjusting for age, ethnicity, and gender. FFM increased in both boys and girls with increasing Tanner stages. Boys had a later spurt but longer duration in FFM increment than girls. Relative FM remained constant in girls but decreased in boys after Tanner stage 2. These findings are similar to previous findings based on cross-sectional data (4, 46). In an earlier cross-sectional study (4), FFM was 15.0 kg and 9.1 kg higher in pubertal (Tanner stages 3 and 4 combined) than in prepubertal boys and girls, respectively, and 4.2 kg and 5.6 kg higher in postpubertal (Tanner stage 5) than in pubertal boys and girls, respectively. In the same study, relative FM decreased with increasing Tanner stage in boys but not in girls. The pattern of change in body composition during puberty has also been studied elsewhere (1, 6, 8, 10, 19, 33, 43). Age- and gender-specific patterns of change in body composition from 8 to 20 years of age were tracked using data from the Fels Longitudinal Study (19). In this study, rate of maturation was determined from skeletal age and chronological age. A PROC MIXED analysis was used to track the body composition of children from childhood into adult-

hood. There was a continuous increase in absolute FM, but a decline in the rate of increase with increasing age. There was an increase in FFM, the rate of which slowed with age in females, but remained constant in males until adulthood. Within a given age, children who had a more rapid maturation rate had a higher accumulation of both FFM and FM than children who had a slower maturation rate. Because African American children begin puberty earlier than Caucasian children (20, 35, 36), the influence of maturation on body composition and REE may be apparent at a younger age in African Americans. In the present study, this pattern may help to explain the higher FFM in African American children compared with Caucasian children at a certain age.

The accretion of FFM in both boys and girls can be explained by a lower proteolysis and protein oxidation, respectively, because of the action of insulin and insulin-like growth factor I in puberty compared with prepuberty (3). Different testosterone and estradiol ratio (T/E_2) (37) and leptin concentration (1, 10) are associated with gender difference in body composition during puberty. In boys, T/E_2 was substantially higher in puberty than in prepuberty because of the larger increments of testosterone than estradiol. In girls, T/E_2 was lower than in boys because of larger increments of estradiol than testosterone. Previous studies have shown that testosterone has a protein anabolic effect (2, 31, 32), which, in turn, is positively associated with FFM. Therefore, an interaction between T/E_2 could explain the gender-dimorphic changes in body composition.

In this study, REE declined with increasing pubertal stage after adjusting for age, ethnicity, gender, FM, and FFM. In the current analysis, the decrease in REE was not significant from Tanner stage 1 to 2, but was significant from Tanner stage 1 to stages 3 and 4. An inverse relationship between REE and age was previously observed in 98

early-pubertal girls (35). REE adjusted for FFM was lowest in Tanner stage 3 girls. The difference in REE between stages 3 and 1 was significant in Caucasian girls. In African American girls, the difference between stages 2 and 1 was significant. This earlier study indicated that the change in REE during puberty was earlier in African Americans than in Caucasians. In another study, Weinsier et al. (45) reviewed 31 data sets with 1,111 subjects and found a decrease in REE with increasing age. REE was the highest in preschool children, followed by adolescents, then adults. In contrast to these findings, pubertal stage was not considered an important contributor to REE independent of body composition in several other cross-sectional studies (4, 34, 47). In 83 children and adolescents whose Tanner stage ranged from 1 to 5 (4), REE varied significantly with gender but not with pubertal stage independent of FFM. In another study (34), absolute REE increased with the advance of puberty. However, after adjusting for FFM and FM, the influence of puberty vanished in both boys and girls. During puberty, skeletal muscle is accumulated more rapidly than organ mass (21). The decrease in REE, adjusted for FFM, with maturation may be explained by the increase in proportion of the relatively less energy-demanding skeletal muscle component of FFM (22).

We observed a lower REE despite a higher FFM in African American children than in Caucasian children after adjusting for age, Tanner stage, gender, FM, and FFM. The magnitude of ethnic difference in REE in our cohort is approximately 60 kcal/d. An ethnic difference in REE was not previously observed in our cohort when children were prepubertal (39). The ethnic difference in REE became more apparent after Tanner stage 2. The relationships between REE and FFM in different Tanner stages are shown in Figures 3 and 4. Our data differ from cross-sectional studies in which a lower REE was ob-

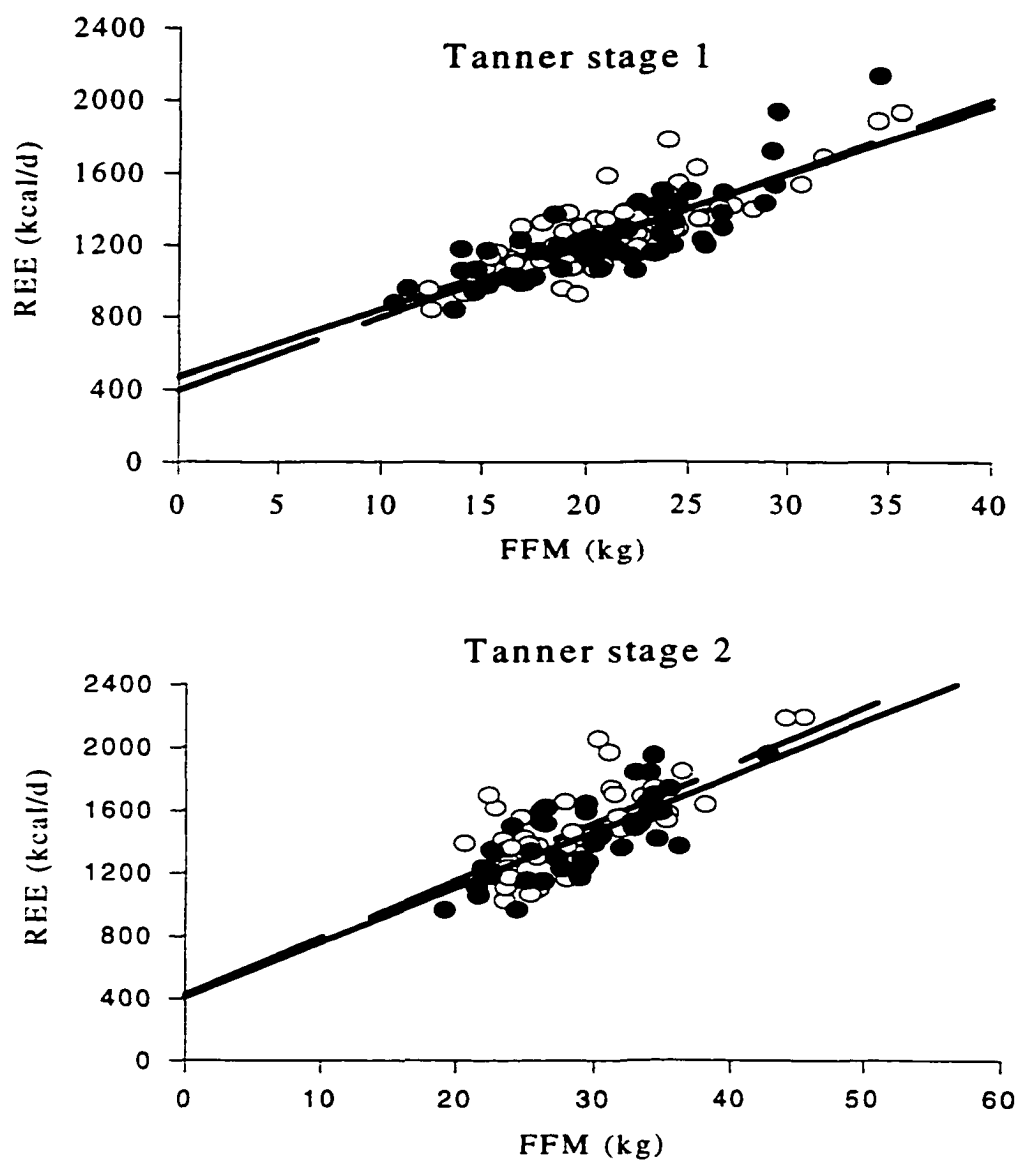


Fig. 3. Relationships between REE and FFM in Caucasian (dotted line, open symbols) and African Americans (solid line, filled symbols) children in Tanner stage 1 and 2. In Tanner stage 1, regression equations for Caucasian children: $\text{REE (kcal/day)} = 37 \text{ FFM (kg)} + 465$; for African American children: $\text{REE (kcal/day)} = 40 \text{ FFM (kg)} + 394$. In Tanner stage 2, regression equations for Caucasian children: $\text{REE (kcal/day)} = 35 \text{ FFM (kg)} + 432$; for African American children: $\text{REE (kcal/day)} = 35 \text{ FFM (kg)} + 406$.

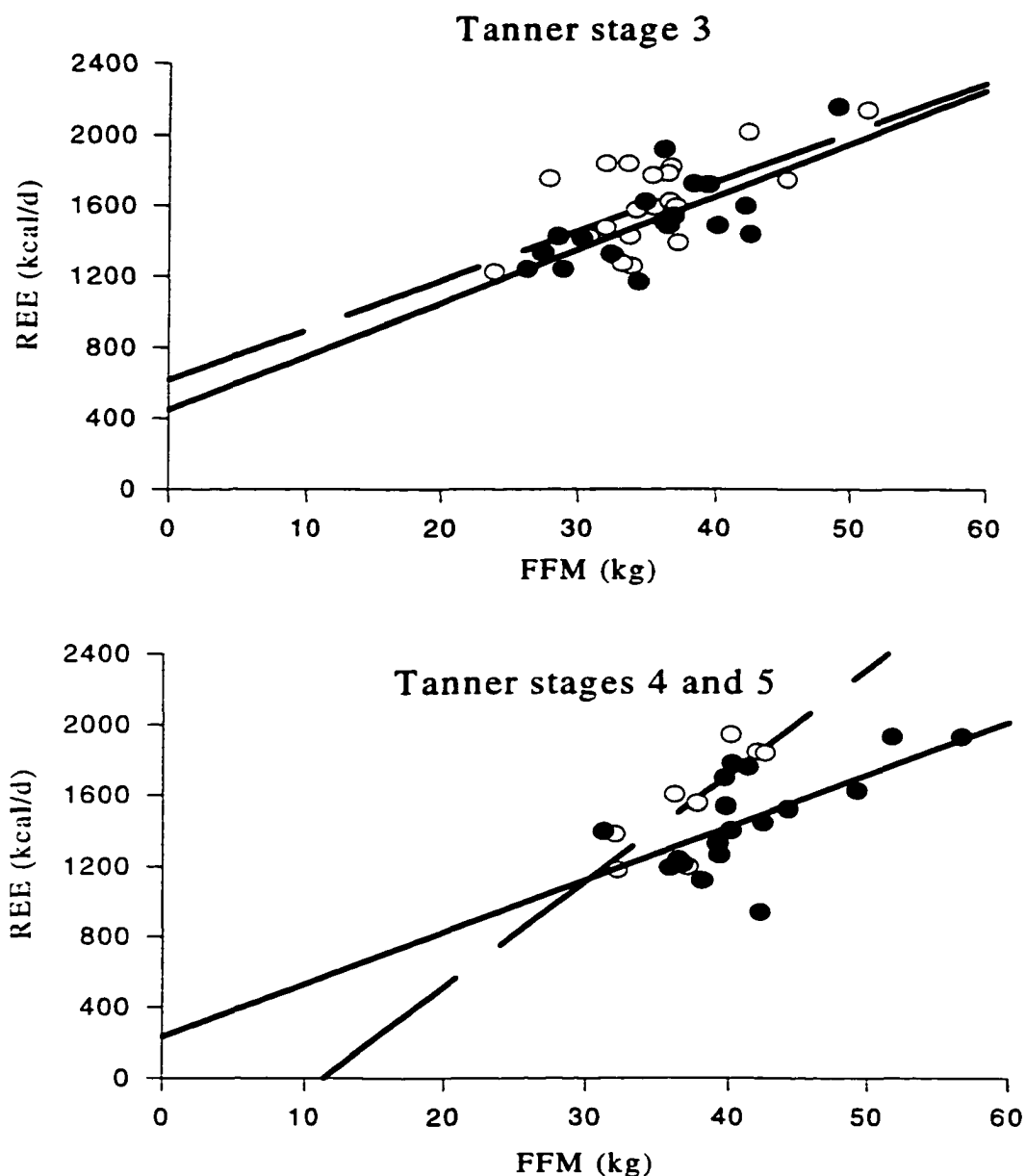


Fig. 4. Relationships between REE and FFM in Caucasian (dotted line, open symbols) and African Americans (solid line, filled symbols) children in Tanner stage 3 and 4/5. In Tanner stage 3, regression equations for Caucasian children: $\text{REE (kcal/day)} = 28 \text{ FFM (kg)} + 617$; for African American children: $\text{REE (kcal/day)} = 30 \text{ FFM (kg)} + 446$. In Tanner stage 4 and 5, regression equations for Caucasian children: $\text{REE (kcal/day)} = 60 \text{ FFM (kg)} - 682$; for African American children: $\text{REE (kcal/day)} = 30 \text{ FFM (kg)} + 232$.

served in African Americans than in Caucasians in both prepubertal (26, 35, 48) and pubertal children (47). The cause for this inconsistency remains unclear. It may be due to failure to accurately assess Tanner stages in different ethnic groups that mature at different rates, or to subtle differences between physical maturation and physiological maturation that cannot be easily assessed. Tanner stage and associated physiological changes are not a continuous process but proceed in patterns.

As mentioned earlier, differences in energy expenditure between organ and muscle may help to explain the decline in REE with maturation. Previous studies in adults have shown conflicting results regarding the contribution of FFM distribution to REE (12, 13, 15, 24, 40, 42). Garby et al. (15), who analyzed data from cadavers, showed that organ weights contributed significantly to REE, after adjusting for body weight or FFM. Other studies have found that, although composition of FFM itself was a determinant of REE, an independent contribution of organ weight was not found, or was found to be weak, after FFM was taken into consideration (12, 40). In the present cohort, African American children had a higher relative LLM and lower relative TLM than Caucasians. Both relative LLM and relative TLM contributed significantly to REE. However, including FFM distribution in the model did not explain the lower REE in African American children beyond FFM *per se*. A recent study in premenopausal women from our group found that a racial difference in REE persisted after adjusting for LLM, but disappeared after adjusting for TLM (23). In the present investigation, we chose to create a new variable by using principle component analysis that incorporated information on both LLM and TLM and avoided multicollinearity between the two. This new variable, LIMTRK, allowed us to examine the relative contribution of LLM and TLM to REE. Since addition

of relative FFM distribution did not eliminate the significant ethnic contribution to REE, it does not appear that the lower relative TLM in African Americans relative to Caucasians explained the lower REE in this cohort. The limitation of this study is that LLM and TLM are only crude surrogates of muscle and organ mass. Further studies are needed in investigating the actual mass in organ and muscle and the relative contribution of each to REE. In addition, this kind of analysis is confounded by multicollinearity, so we were not able to separate the two components and examine independent contributions of each.

Puberty is a period during which children are susceptible to the development of obesity. If future research confirms a lower REE in African Americans than in Caucasians during pubertal development, the lower metabolism may be related to the development of obesity among African Americans. Previous longitudinal study from our laboratory has shown that REE does not predict fat gain in Caucasian prepubertal children over 5 yr (18) and in African American children (25). Whether a lower REE predisposes African Americans to a higher obesity rate needs to be further examined in children in later Tanner stages when REE starts to decline. Other factors, such as food intake and physical activity, may be more significant with regard to weight gain. The ability of future research to assess energy balance is important. One important concern in this kind of study is the definition of maturation stage. Studies reviewed in this paper have used different staging criteria. Tanner stage is based on physical examination alone, and criteria may vary slightly among clinicians. Three pediatricians participated in our study. Where discrepancies existed (five cases), we used the lower Tanner stage. One limitation of our study was that we did not use bone age, an important determinant of maturation (39). Use of bone age may improve the ability to gauge maturation.

In conclusion, current results indicate a negative relationship between pubertal maturation and REE after adjusting for FM and FFM. African Americans, on average, had a lower REE than Caucasians. In this cohort, the ethnic difference in REE started to emerge after the onset of puberty. The ethnic difference in REE was not explained by the relative distribution of LLM and TLM. Whether earlier maturation and concomitant decline in REE predispose African American children to obesity remains to be determined.

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SUMMARY AND CONCLUSIONS

The goal of this dissertation work was to examine body composition and REE in African American and Caucasian children during puberty. Specifically, we examined how pubertal maturation contributed to the change in body composition and REE and how these relationships differed by ethnicity and gender.

There was no ethnic difference in any of the components of energy expenditure in prepubertal children after adjustment for body composition. When hormonal indices of maturation were adjusted for, we still did not observe an ethnic difference in REE in this group of prepubertal children. By examining these children through puberty in a longitudinal analysis, we found a positive relationship between pubertal maturation and lean mass after adjusting for age, and a negative relationship between pubertal maturation and REE after adjusting for age, ethnicity, gender, FM and lean mass. African American children, on average, had a lower REE than in Caucasians. This ethnic difference in REE started to emerge after the onset of puberty in our cohort. However, this ethnic difference was not further explained by relative distribution of LLM and TLM, surrogates for muscle and organ mass.

Whether earlier maturation and the concomitant decline in REE predisposes African American children to obesity remains to be determined. If future research confirms a lower REE in African American children than in Caucasian children during pubertal development, the lower metabolism may be related to the propensity to obesity among African Americans in later years.

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I certify that I have read this document and examined the student regarding its content. In my opinion, this dissertation conforms to acceptable standards of scholarly presentation and is adequate in scope and quality, and the attainments of this student are such that _he may be recommended for the degree of Doctor of Philosophy.

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