CHILDHOOD OBESITY: THE ROLE OF GENETIC ADMIXTURE, PARENTAL FEEDING PRACTICES, AND THE SCHOOL FOOD ENVIRONMENT

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ABSTRACT

Disentangling the etiology of pediatric obesity continues to challenge researchers due to the interconnecting of behavioral, physiological, environmental, and genetic factors. Studies have focused on the role of the obesogenic environment, characterized by excess energy intake and sedentary behavior, as it relates to weight and fat gain in children. Although these factors certainly have an impact on fat accumulation, increased energy intake and decreased physical activity do not solely account for the increasing trends in obesity prevalence. Less explored genetic and environmental factors may contribute independently and/or synergistically to the current obesity epidemic, and that, these factors could vary in different groups. The objective of this dissertation is to explore the relationships of non-traditional factors related to the home and school environment and population variation that may contribute to excess fat accumulation and differences in body composition across populations. The specific aims were investigated in a cohort of 301 European-American (n=115), African-American (n=107), and Hispanic-American (n=79) children aged 7–12 (specific aim 1 and 3). A sub-cohort including the European-American children was also investigated and stratified by school status [home-schooled; n=47 vs. traditionally schooled; n=48] for specific aim 2. The first specific aim was to determine whether parental feeding practices differed by
race/ethnicity; whether parental pressure to eat and parental restriction were associated with adiposity levels; and to investigate the relationship between parental feeding practices and/or child adiposity with socioeconomic status. In specific aim 2, we aimed to compare and evaluate the relationship between dietary intakes and the school food environment on adiposity between home-schooled children and traditionally schooled children. In the third specific aim, we determined the extent to which individual estimates of African genetic admixture and European genetic admixture contribute to body composition and fat distribution. Our findings suggest that parental restriction, parental pressure to eat, and school status are modifiable environmental factors associated with child adiposity. African genetic admixture and European genetic admixture were related to various measures of body composition. These results have great public health significance and provide further understanding of putative factors that increase the risk for excess fat accrual in children.

Keywords: childhood obesity, body composition, parental feeding practices, school food environment, genetic admixture
DEDICATION

This dissertation is dedicated to my mother, Ivonne Garcia Sullivan, for her unconditional support and love, which has shaped my very existence.

“All that I am and ever will be, I owe to my angel mami.”

-The Puerto Rican version of Abraham Lincoln’s quote
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INTRODUCTION

PARENTAL FEEDING PRACTICES AND SOCIOECONOMIC STATUS ARE ASSOCIATED WITH CHILD ADIPOSITY IN A MULTI-ETHNIC SAMPLE OF CHILDREN

SUMMARY
INTRODUCTION

The prevalence of childhood obesity has increased over the last 40 years (1). Recent reports indicate 18% of children are obese (1), defined as a body mass index (BMI; kg/m$^2$) > 95th percentile for age and sex (2). Research has demonstrated that obese children are more likely to become obese adults, increasing an individual’s risk for obesity-related diseases (3-8). Earlier onset of obesity has been correlated with a higher risk for the development and severity of complications such as type 2 diabetes, metabolic syndrome, and dyslipidemia, as well as negative psychological outcomes including depression and poor body image (3-8). The adverse effects associated with pediatric obesity are a matter of great public health concern, and efforts toward understanding the etiology of childhood obesity are needed for the development of successful long-term treatment and prevention strategies (9).

Understanding the nature of body-weight regulation via energy balance is an essential component to understanding the development of obesity. Energy balance occurs when the amount of calories available from consumption (energy intake) is equivalent to the amount of calories expended (energy expenditure). A state of energy imbalance results in a decrease or increase of energy storage, reflected in body-weight changes and/or in body composition parameters (i.e., bone, lean mass, and fat mass) throughout the lifespan (10;11). The development of obesity reflects a state of positive energy balance, characterized by an excess of adiposity (12).
Some researchers have blamed the rise in obesity prevalence on the “toxic” environment (13) characterized by excessive consumption of food and decreased physical activity. According to Wadden and colleagues, the toxic environment refers to an unprecedented exposure to energy-rich foods that are highly accessible, convenient, and affordable (13). When combined with a sedentary lifestyle where a significant number of children watch at least 2–4 hours of television per day (14;15) and do not meet physical activity recommendations (15-17), excessive caloric consumption is accumulated as body fat. Although measurements of physical activity and caloric intake have been associated with the increased obesity trend (15;18;19), they do not solely account for the observed variability in adiposity in children (20;21).

Additional factors may be contributing independently and/or synergistically to the increase in obesity prevalence and individual variability in body fat. For example, an overfeeding study where individuals were experimentally put in positive energy balance showed that there is inter-individual variation in adiposity accrual and body composition (22). This suggests a differential predisposition to fat accumulation that could be based on genetics. It has been estimated 50% of the variation of body mass index can be explained by genetic factors (BMI = kg/m²); however, genetic association studies have been unsuccessful in accounting for more than 2% of BMI variation in the population (23). The lack of understanding of the contributions of genes and environments to adiposity measures support the need to explore other factors contributing to fat accrual. A comprehensive understanding of the environmental, behavioral, and genetic factors influencing energy balance may highlight potential prevention and intervention strategies for optimal body weight and body composition trajectories.
Racial/Ethnic and Socioeconomic Differences in Obesity Prevalence and Body Composition

Significant differences in obesity prevalence by race/ethnicity have been observed in the U.S. (1). Recent reports indicate that 22.5% of Hispanic children and 28.6% of African-American children aged 6–11 are obese compared with 13.5% of European-American children (1). Though BMI-for-sex-and-age percentiles (BMI%) is a widely used surrogate measure of adiposity, it is a measure of excess weight relative to height, rather than excess body fat (24). Therefore, simply looking at obesity prevalence rates or BMI% does not provide insight into fat accumulation or distribution.

Studies have evaluated whether the relationship between BMI% and adiposity differs across populations (24;25). In one such study, Freedman and colleagues used dual-energy X-ray absorptiometry (DXA) to determine body fat and assessed the accuracy of BMI% as an indicator of adiposity across populations. Similar to the trends observed by Ogden et al. (1), this study found that African-American and Hispanic-American children had the highest BMI-for-sex-and-age percentiles and the highest prevalence of obesity (24). Despite the high BMI% of African-American children, African-American boys had the lowest mean of percent body fat (17%), and African-American girls’ total adiposity paralleled that of European-American girls (25–27%) (24). Hispanic-American children, however, had the highest mean BMI% and percent body fat for both girls and boys (30% and 21%, respectively) (24). Among children with similar BMI%, adiposity levels differed up to 5% across racial/ethnic groups (24). It appears as though BMI% alone is not a precise measure for assessing adiposity when evaluating diverse populations.
In addition to disparities in BMI prevalence and body fat outcomes in minority populations, differences in other body composition parameters have been documented (26-28). For example, at a given BMI, African-American children have exhibited greater bone mineral content, greater lean mass, and less visceral and total fat relative to their European-American or Hispanic-American counterparts (24;26;29;30). This suggests that at a similar BMI%, body composition parameters may differ across racial/ethnic groups, potentially impacting health across individual’s lifespan.

Socioeconomic status (SES) appear also to impact obesity prevalence, with both parental income and education being cited as significant independent predictors of increased levels of obesity-related measures in adolescents (31). Low SES is associated with increased obesity prevalence across socioeconomic levels, though the relationship is complex and often varies by race/ethnicity (31-33). Longitudinal studies show that low SES in childhood is associated with BMI% in adolescence (34) and BMI in adulthood (35). Data demonstrate a contribution of SES to both dietary patterns (36;37) and physical activity (33;38) in children, without a conclusive understanding of their relationship to trends in pediatric adiposity in diverse populations.

A Multifactorial Approach to Obesity Research

As previously stated, caloric intake and physical activity have been shown to influence weight gain and fat accumulation in children (15;18;19) but do not account for all the population variation in adiposity (20;21). Although it is evident that genetic and environmental factors influence food intake and physical activity, more comprehensive approaches are needed to understand the underlying factors that could independently and
synergistically mediate the nature vs. nurture relationship of energy balance and fat acquisition. Research supports the use of comprehensive frameworks to understand fat accrual and to guide the prevention and treatment of childhood obesity. For example, The Obesity Society has recommended the “socio-ecological framework” (39), a comprehensive model recognizing that children should be perceived in the context of their families, communities, and cultures while accounting for the environmental, biological, and behavioral determinants of health. In this dissertation, we adapt the socio-ecological framework to evaluate non-traditional genetic and environmental factors that may contribute to variation in levels of pediatric adiposity. We use multifactorial approaches where we account for the effects of caloric intake and physical activity to address the hypotheses of interest. These factors are: parental feeding practices, the school environment, and ancestral genetic makeup. A description of these factors follows.

Parental Feeding Practices and Pediatric Body Composition

The home environment plays a primary role in the development and shaping of child eating behaviors (40-42). Parents may also contribute to energy intakes and satiety signaling via their respective feeding practices (43;44). Historically, parental feeding practices were developed in humans as survival responses to environmental barriers and threats (predominantly food scarcity) and have been passed down generationally (45). However, in the present-day environment, where an excess of energy-dense foods is readily available, convenient, and affordable (13), parental feeding practices have been hypothesized to play a role in energy imbalance and excess fat accumulation (46;47).
Research has identified parental feeding practices, particularly parental restriction and pressure to eat, as a potential modifiable predictor of childhood obesity. Parental restriction, defined as limiting a child’s intake of “unhealthy” or calorie-dense foods, has been hypothesized to limit a child’s ability to self-regulate energy intake, by hindering hunger and satiety cues, potentially leading to eating in the absence of hunger (EAH) (48). Research has demonstrated that when parents restrict children from eating particular foods, under their own belief that the given food is unhealthy, it increases a child’s risk for higher BMI, overeating, weight gain, and EAH (49-52). EAH has been associated with the development of obesity (53), and longitudinal analysis indicates higher parental restriction at age five was related to EAH and greater fat mass accrual over time, particularly in overweight children (49). Laboratory studies also have demonstrated that children who are exposed to a significant amount of parental restriction tend to eat large amounts of palatable food following a meal, and this EAH and excess energy intake is positively related to child weight (54). Thus, parental feeding practices are integral in the progression of autonomous feeding behaviors, which influence energy intakes and, therefore, also may impact energy utilization and storage (15). Therefore, parental restriction has the potential to limit a child’s ability to self-regulate energy intake and, consequently, energy balance. Studies could further elucidate the role of parental feeding as a putative contributor to excess adiposity in children.

Another component of parental feeding practices is pressure to eat, where parents encourage children to “clean their plate.” Pressure to eat also has been associated with decreased preference and consumption of the pressured food, decreased intake of fruits and vegetables, picky eating, and lower weight in children (55;56). This is supported by
longitudinal analyses reporting children who were pressured to eat ate less and displayed more negative reactions to foods they were pressured to eat relative to children who were not pressured to eat at their meals (57). Additionally, in a retrospective study, young adults reported disliking foods they had been pressured to eat as children (58). Though it would be expected that a behavior that enables a child to be at a lower weight status would be beneficial, it may be at the expense of a nutrient-dense diet and can lead to a long-lasting aversion to foods they were once pressured to eat (58).

However, it is important to note that previous work pertaining to parental feeding practices has been conducted primarily in highly educated, European-American mother-daughter dyads and is not representative of the general U.S. population. Research aimed at understanding the role of parental feeding practices in diverse populations, including those with lower SES, is limited and needed as it could provide further insight into mechanisms by which dietary patterns impact overall adiposity in underserved populations.

The School Food Environment and Pediatric Body Composition

The home environment has been shown to play a role in the development of childhood obesity (59-61). As children get older, many spend a significant amount of time away from the home, allowing for additional environments to influence children’s energy intakes and body composition (61;62). Therefore, it is important to explore other environments that may influence diet, physical activity, and energy balance.

At school, where children spend a great deal of time, they consume approximately 35%-47% of their daily food intake and expend up to 50% of their daily energy (22). The
Institute of Medicine identified the school environment as the primary setting for preventing childhood obesity by making environmental and policy changes (63). To inform policy makers, research has attempted to explore how the traditional school environment impacts energy balance. Studies have suggested the school environment affects energy balance by facilitating an increase in both the availability and the intake of energy-dense foods and beverages (23-26) and/or by decreased participation in physical education programs (27). Regardless, controversy pertaining to the effects of the school environment on child weight remains, as some studies found no association between the traditional school environment and child weight (64;65). The inconsistent results may be due, in part, to discrepancies in the measurement or classification of what constitutes a “school lunch.” Some researchers classify only that which is provided by the National School Lunch Program (64;65) as a school lunch. Other studies also include a la carte items, competitive foods, and other foods children have access to while at school as a “school lunch,” including foods that were brought from home (61;66). Measuring the school food environment is a complex issue, and its implications could affect policy and prevention programs for pediatric obesity. Therefore, in order to determine the influence of the school food environment on dietary patterns and energy balance, research evaluating the influence of the school food environment should include only school-accessible foods consumed by children that were not brought from home. One way in which to truly gauge the influence of the “traditional school environment” on energy balance and adiposity would be to compare it to a different educational environment.

In order to assess the role of the traditional school environment on child adiposity, it is conceivable that home-schooled children (HSC), who consume lunch at home, could
serve as a novel comparison group when compared to traditionally schooled children (TSC) who purchase and consume their lunch at school. Approximately 2 million children are currently being home-schooled, and this number is increasing every year (67). Though studies have characterized various aspects of home-schooled families (68;69), to our knowledge, research has not evaluated home-schooled children’s dietary intakes and adiposity levels. As HSC’s dietary habits and body composition have been overlooked in the obesity literature, inclusion of this population as a comparison group allows for a better understanding of how the school food environment plays a role in energy balance and adiposity. This is the first study to characterize and determine if differences exist between HSC and TSC.

Genetic Admixture and Pediatric Body Composition

Though genetic factors have been estimated to contribute approximately 50% of the variance in BMI within a given population (70-72), exploring the etiology of racial/ethnic differences in obesity-related parameters has been challenging. On one hand, race/ethnicity captures the non-biological characteristics shaped by one’s experiences and history, including language, culture, and religious traditions that could influence energy balance through dietary preferences and intakes (73;74). On the other hand, biological factors underlying self-identified race/ethnicity are known to confound genetic analyses through a process known as population stratification (79;80).

The social and cultural aspects underlying race and ethnicity have been known to influence obesity-related traits. For instance, affordability, acculturation, and availability of ingredients in traditional dishes may persuade newly immigrated parents to retain or
discard certain traditional foods. An example of this is represented by the Pima Indian women living in Arizona, who were found to have an average dietary-fat intake of 41% of total calories and a BMI of 37 kg/m² compared with 23% of calories from fat and a BMI of 25 kg/m² for woman remaining in Mexico (75). Evidently, culture has the potential to affect overall energy balance and, thus, body composition via mechanisms of eating behavior and dietary patterns (75). However, as an increasing proportion of the U.S. population describes themselves as “mixed” or “other” race/ethnicity (39;74), understanding the etiology of group comparisons becomes an issue of research concern.

Underlying racial/ethnic identity, there is a shared ancestral genetic component that responds to the history of populations. For example, in the U.S., the history of colonization resulted in the creation of admixed populations, including African Americans, European Americans, and Hispanic Americans. The ancestral genetic background of individuals in these admixed populations can be evaluated through the use of ancestry informative markers (AIMs) that differ in allelic frequency between parental populations (76). Parental populations are defined as those isolated populations that merged in the New World during the colonization period (i.e., Europeans, Amerindians, and Africans). Using AIMS, the proportion of an individual’s genome responding to each parental population could be estimated. These genetic admixture estimates represent an objective measure of ancestral genetic background that can be employed to explore the genetic contribution to racial/ethnic disparities in obesity. Although there is multicollinearity between the race/ethnicity and admixture variables, race/ethnicity measures a social/contextual construct (77), whereas genetic admixture measures genetic ancestral
background (78). Therefore, these two measures may provide insight into two different constructs and should not be used interchangeably (39).

In the United States, the contributions of West African, European, and Amerindian parental populations have been significantly associated with body composition parameters in adults (79-81). For example, European admixture has been inversely associated with BMI and percent body fat in Native American young adults (81). Studies in African-American adults have reported inverse relationships between African admixture and BMI and percent body fat, while European admixture has been positively related with percent body fat (79;80). In children, research suggests genetic admixture is related to bone mineral content and insulin-related parameters (82;83), but further investigations are warranted to evaluate the extent to which ancestral genetic background influences body composition. This could further elucidate whether and how genetic and environmental factors account for differences in obesity-related parameters across populations.

Experimental Aims

The work of this dissertation has been proposed to help inform potential factors influencing the etiology of pediatric adiposity, with the overall long-term goal of aiding in the development of effective childhood obesity intervention and prevention programs. This dissertation focuses on exploring factors related to the home and school environment and population variation that may impact aspects related to energy balance, contributing to the body of literature to further understand the etiology of fat accumulation and deposition in children. The following experiential aims were designed to investigate the
multifactorial relationships that may exist between genetic ancestry, race/ethnicity, socioeconomic status, parental feeding practices, the school food environment, and child adiposity. In this dissertation, child adiposity is used in reference to either total fat mass or percent body fat, unless otherwise specified. Figure 1 presents the main analyses for each experimental aim.

![Diagram of Experimental Aims]

**Figure 1. Outline of Experimental Aims**

*Experimental Aim 1*

The objective of this aim is to determine whether (1) parental feeding practices differed by race/ethnicity; (2) parental pressure to eat and parental restriction were associated with adiposity levels; and (3) to investigate the relationship between parental
feeding practices and/or child adiposity with socioeconomic status in a cross-sectional study of African-American, Hispanic-American, and European-American children.

Anthropometric techniques, dual-energy X-ray absorptiometry (DXA), and computed tomography (CT) scan were used to measure body mass index (BMI), trunk fat, and total abdominal adipose tissue, respectively. Parental feeding practices were assessed via the modified Child Feeding Questionnaire, and socioeconomic status was determined using the Hollingshead 4-factor index of social class. Structural equations modeling evaluated the relationships between parental feeding practices, socioeconomic status, and measures of child adiposity, after adjustment for child sex, pubertal status, African genetic admixture, and European genetic admixture.

**Experimental Aim 2**

The objectives of this aim are 1) to quantitatively measure and compare dietary intakes, physical activity, and body composition in home-schooled children and traditionally schooled children, and 2) to evaluate the relationship between overall dietary intakes, the school food environment, and physical activity on adiposity in European-American home-schooled children and traditionally schooled children.

Parents of the participants reported child school status (home-schooled vs. traditionally-schooled). Dietary intakes were obtained via two 24-hour recalls, and objective physical activity was assessed over a seven-day period using an accelerometer. Body mass index, fat mass, trunk fat, and percent body fat were determined by anthropometric techniques and dual-energy X-ray absorptiometry (DXA). Descriptive statistics were compared using analysis of covariance (ANCOVA) with Tukey’s post hoc
analysis. Multivariate linear regression models investigated the associations between school status, dietary intakes, and physical activity with adiposity variables after adjusting for age, pubertal status, socioeconomic status, and sex.

Experimental Aim 3

The objective of this AIM is to determine the extent to which individual estimates of African genetic admixture and European genetic admixture contribute to body composition and fat distribution in a cross-sectional study of African-American, Hispanic-American, and European-American children.

For this aim, genetic admixture estimates were obtained from genotyping approximately 142 ancestry-informative markers across the human genome informative for European, African, and Amerindian ancestry. Total fat mass, bone mineral content, and lean mass were assessed with dual-energy X-ray absorptiometry (DXA), and computed tomography (CT) scans were used to quantify the distribution of total abdominal adipose tissue, intra-abdominal adipose tissue, and subcutaneous abdominal adipose tissue. Multiple regression models were conducted to evaluate the contribution of admixture estimates to body composition and fat distribution after adjusting for sex, age, socioeconomic status, height, race/ethnicity, and pubertal status.
PARENTAL FEEDING PRACTICES AND SOCIOECONOMIC STATUS ARE ASSOCIATED WITH CHILD ADIPOSITY IN A MULTI-ETHNIC SAMPLE OF CHILDREN

by

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Format adopted and errata corrected for dissertation
ABSTRACT

Parental feeding practices have been associated with children’s weight status, but results have been inconsistent across populations. Research is needed to elucidate the relationship between parental feeding practices and adiposity in diverse populations. The present study tested if: (1) parental feeding practices differed by race/ethnicity (2) parental pressure to eat and parental restriction were associated with adiposity levels, and (3) to investigate the relationship between parental feeding practices and/or child adiposity with socioeconomic status (SES). Structural equations modeling was conducted to test the model in 267 children aged 7-12 years self-identified as African American (AA), European American (EA), or Hispanic American (HA) from economically diverse backgrounds. Dual energy X-ray absorptiometry and Computed Tomography scanning were used to determine body composition and abdominal fat distribution, respectively.

Parental restriction was a significant predictor of child adiposity while parental pressure to eat had an inverse relationship with child adiposity. HA parents reported significantly higher levels of restriction and pressure to eat, whereas EA parents reported the lowest. SES was positively associated with child adiposity and inversely related to parental restriction and pressure to eat.

Thus, parental feeding practices differ across racial/ethnic groups and SES and may contribute to population differences in child adiposity.

Keywords: obesity, restriction, pressure to eat, fat, race, ethnicity, diverse, admixture
INTRODUCTION

The prevalence of childhood obesity has been increasing over the last thirty years, with the most recent reports indicating an increase in both the degree and severity (Ogden et al., 2010). Research indicates that obese children are more likely to become obese adults (Magarey et al., 2003), and this tracking of pediatric obesity into adulthood may have detrimental implications for long term health (Field et al., 2001; Whitaker et al., 1997; Magarey et al., 2003).

Parental beliefs and practices regarding child feeding play a significant role in shaping eating behaviors. Parental feeding practices, particularly restriction of palatable foods and pressure to eat, have been related to children’s body weight and energy intakes. Parental restriction has been positively related with child body mass index (BMI), overeating, and weight gain (Fisher & Birch, 2002; Francis et al., 2001; Joyce & Zimmer-Gembeck, 2009; Birch & Fisher, 2000; Faith et al., 2004). It has been hypothesized that parental restriction may limit a child’s ability to self regulate energy intake and focus children’s attention on restricted foods, which may lead to overconsumption when the restricted foods become freely available (Fisher JO & Birch L.L., 1999). Parental pressure to eat, on the other hand, has been associated with decreased preference and consumption of the pressured food, lower fruit and vegetable intake, picky eating, and lower weight in children (Galloway AT et al., 2005; Galloway et al., 2006). However, the relationships between parental feeding practices and adiposity are not consistent across studies. Studies from Australia and the United Kingdom have found no association between parental restriction and child weight (Webber et al., 2010; Campbell et al., 2010; Carnell & Wardle, 2007) and longitudinal studies have shown no association
between pressure to eat and child adiposity measures (Webber et al., 2010; Spruijt-Metz et al., 2006). Although it has been theorized that parental beliefs and practices may be modifiable determinants to prevent pediatric obesity and excessive adiposity (Savage J.S. et al., 2007), it is evident that the relationships between parental feeding practices and risk for obesity in children are complex, and do not appear to carry the same effect among all.

The etiology of reported marked differences in obesity prevalence among members of diverse racial/ethnic groups in the United States is not clearly understood (Flegal et al., 2010). Limited research understanding the role of parental feeding practices in diverse populations, including those with lower socio-economic status (SES), has been conducted, generating null or mixed results (Mulder et al., 2009; Webber et al., 2010; Carnell & Wardle, 2007; Hennessy et al., 2010; Spruijt-Metz et al., 2006; Spruijt-Metz et al., 2002). Thus, SES and race/ethnicity may be possible contributors to underlying relationships between feeding practices and child weight. As minority and low-income populations continue to be disproportionately affected by overweight and obesity (Shrewsbury & Wardle, 2008; Ogden et al., 2010), the need to study diverse populations and to identify factors that may predispose minority and low-income children to pediatric obesity are paramount.

The objectives of this study were to test a model that focuses on the relationship between parental feeding practices on child adiposity in a socioeconomically and ethnically diverse population (Figure 1). This model is based on prior research in a homogenous sample (Fisher J.O. & Birch L.L., 1999; Fisher & Birch L.L., 2002; Galloway et al., 2006) and was inspired by the Social Determinants of Health and
Environmental Health Promotion model (Schulz & Northridge, 2004). We hypothesized that (1) parental feeding practices would differ by race/ethnicity, (2) parental pressure to eat would be inversely associated and parental restriction would be positively associated with adiposity levels, and (3) SES would be inversely associated with parental restriction, pressure to eat, and/or child adiposity in a multi-ethnic sample of children from economically diverse backgrounds.
METHODS

Subjects

This paper utilizes secondary data analysis from a cross-sectional study evaluating genetic and environmental factors underlying pediatric racial/ethnic differences in body composition and insulin-related outcomes. Participants were 267 children aged 7-12 years self-identified as AA (n=91), EA (n=116), or HA (n=60) from the Birmingham, Alabama area. Children were recruited at schools, churches, health fairs, and through newspapers, parent magazines, radio, and participant referrals. The children were peripubertal (pubertal stage ≤3 as assessed by a pediatrician according to the criteria of Marshall and Tanner) (Marshall WA & Tanner JM, 1969; Marshall WA & Tanner JM, 1970), and had no medical diagnosis or medications contraindicated for study participation (i.e. medication known to affect body composition, metabolism, cardiac function). Prior to study participation, the children and parents provided informed assent and consent, respectively, to the protocol, which was approved by the Institutional Review Board for human subjects at the University of Alabama at Birmingham. All measurements were performed at the General Clinical Research Center (GCRC) and the Department of Nutrition Sciences at the University of Alabama at Birmingham between 2005 and 2008.

Protocol

Subjects participated in two visits. On the first visit, pubertal status, anthropometric assessment, questionnaire data, and body composition were measured. Within thirty days
the children and their parents returned for the second visit, where blood for genetic admixture analysis was drawn.

Anthropometric measures

Anthropometric measurements for all participants were obtained by the same registered dietitian. Participants were weighed (Scale-tronix 6702W; Scale-tronix, Carol Stream, IL) to the nearest 0.1 kg (in minimal clothing without shoes). A single measure of height was recorded to the nearest 0.1 cm without shoes using a digital stadiometer (Heightronic 235; Measurement Concepts, Snoqualmie, WA). Children’s BMI-for-age percentiles were calculated as indicated by the Center for Disease Control and Prevention guidelines (Kuczmarski RJ et al., 2002).

Body Composition and Fat Distribution

Body composition was measured by dual-energy x-ray absorptiometry (DXA) using a GE Lunar Prodigy densitometer (GE LUNAR Radiation Corp., Madison, WI). Participants were scanned in light clothing, while lying flat on their backs with arms at their sides. DXA scans were performed and analyzed with pediatric software encore 2002 version 6.10.029. DXA has been found to be highly reliable for body composition assessment in children (Elberg et al., 2004). For the purpose of this study, only trunk fat (kg) was used in the analysis. Total abdominal adipose tissue (TAAT; cm²) was measured by computed tomography (CT) scanning with a HiLight/Advantage Scanner (General Electric, Milwaukee) as previously described (Kekes-Szabo T et al., 1994). A 5mm abdominal scan was taken at the level of the umbilicus. Scans were analyzed for
cross-sectional area (cm²) of adipose tissue using the density contour program with Hounsfield units for adipose tissue set at -190 to -30. CT has been shown to provide accurate measurements of body fat distribution in children (Fox K et al., 1993). The indicators of total abdominal adipose tissue, trunk fat mass, and BMI-for-age percentile were chosen for the Child Adiposity latent construct because they have been related to metabolic outcomes and obesity in children and adults (Bjorntorp P, 1988; Rexrode K et al., 1998; Casazza K et al., 2009; Edyta Suliga, 2009; Magarey et al., 2003).

Parental Feeding Practices

The Child Feeding Questionnaire (CFQ) was developed by Birch et al. to measure parental feeding practices in EA children (Birch et al., 2001) and has been used previously in children of this age group (Spruijt-Metz et al., 2006; Campbell et al., 2010; Spruijt-Metz et al., 2002). For this study, the modified CFQ was given, which used questions from the original CFQ which were validated for use in HA and AA (Anderson et al., 2005). Restriction and pressure to eat were measured by a five-point Likert scale and responses ranged from agree (coded as a 5) to disagree (coded as a 1). The questions measuring parental restriction (RES1a, RES1b, and RES4a) were “I have to make sure my child does not eat too many sweets”, “I have to be sure that my child does not eat too many high fat foods”, and “I have to be sure that my child does not eat too much of his/her favorite foods”, respectively. The questions measuring “pressure to eat” (PE1, PE2, and PE3) were “My child should always eat everything on his/her plate”, “I have to be especially careful to make sure my child eats enough”, and “If I did not guide or regulate my child’s eating, he/she would eat much less than he/she should”, respectively.
Higher scores are indicative of higher engagement in feeding practices while low scores reflect little participation in child feeding practices. All parents participated in the questionnaire, and the Cronbach alpha for restriction and pressure to eat were 0.83 and 0.69, respectively.

Socioeconomic Status

Socioeconomic status was measured with the Hollingshead 4-factor index of social class (Hollingshead AB, 1975), which combines the educational attainment and occupational prestige for working parents in the child’s family. Scores range from 8 to 66, with higher scores indicating higher theoretical social status.

Pubertal Status

Tanner staging is an objective measure of pubertal development. Direct observation for the assessment of pubertal stage by the same pediatrician, was used for differentiating among the five stages of maturity (Marshall WA & Tanner JM, 1969; Marshall WA & Tanner JM, 1970). The staging based on the criteria of Marshall and Tanner is according to both breast and pubic hair development in girls and genitalia and pubic hair development in boys. One composite number is assigned for Tanner staging, representing the higher of the two values defined by breast/genitalia and pubic hair development (Malina RM & Bouchard C, 1991). Given that pubertal stage and biological sex plays a role in adiposity accrual and distribution (Goulding et al., 1996; Demerath et al., 2006), both were included as covariates in the analysis.
Genetic admixture

Genetic admixture better reflects genetic components of race/ethnicity than self-report (Fernandez & Shriver, 2004) and has been shown to influence body composition in children (Cardel et al., 2011; Casazza K et al., 2010). Thus, estimates of ancestral genetic admixture were used to adjust for the genetic contribution to body composition. The genetic admixture estimates were obtained for each child’s blood sample from genotyping ~142 ancestry informative markers (AIMs) across the human genome for European, African and Amerindian parental ancestry. Genotyping for the measures of genetic admixture was performed at Prevention Genetics (www.preventiongenetics.org) using the McSNP method and agarose gel electrophoresis, as previously described (Cardel et al., 2011). Individual admixture estimates were derived using maximum likelihood (ML) method, which estimates the proportion of genetic ancestry for an individual, using a range of proportions from 0 to 1 and identifies the most probable value of admixture based on the observed genotypes, as previously described (Fernandez & Shriver, 2004; Shaffer JR et al., 2007). The maximum likelihood method estimates the proportion of genetic ancestry for an individual, using a range of proportion from zero to one and identifies the most probable value of admixture based on the observed genotypes.

Statistical Analysis

Correlations were conducted for all variables of interest and descriptive statistics were analyzed among ethnic groups using analysis of variance (ANOVA) with Tukey’s post-hoc analysis. The normality of the distribution of values was evaluated and non-normal variables were log-transformed. Means for restriction and pressure to eat
between ethnic groups were analyzed with analysis of covariance (ANCOVA). The covariates are child sex, pubertal status (as assessed by tanner), African admixture, and European admixture. All above analyses were done using Statistical Analysis Software (SAS; version 9.1).

Structural equations modeling (SEM) using Mplus software (Muthen and Muthen, Los Angeles, CA) with ML estimation was used to model the relationships between parental feeding practices, SES, and child adiposity. SEM allows for latent variables and for simultaneous evaluation of multiple regression equations, providing estimates of the direct and indirect effects, while adjusting for covariates and colinearity among all variables. In the model (Figure 3), ovals are shown to represent latent variables. The black boxes represent observed variables and the gray boxes represent the indicators used to measure the latent variables. Direct paths between the variables are indicated by single-headed arrows estimated by linear regression coefficients. Correlations between the variables are shown by double-headed arrows. Significance for all relationships was set at a level of $p \leq 0.05$. Standard fit indices were used to test the fit of the model (Rex B.Kline, 2005): chi-square ($\chi^2$) test of model fit, its p-value and degrees of freedom (df); CFI (comparative fit index; good fit is $>0.90$); and RMSEA (root mean square error of approximation; good fit is $<0.05$).
RESULTS

Parental and Child Characteristics

Descriptive statistics are reported in {Table 1}. Socioeconomic status differed by race/ethnicity with EA having the highest level and HA having the lowest level. Indices of child adiposity differed by groups with HA children having a higher BMI-for-age percentile, greater TAAT, trunk fat mass, total fat, and percent fat when compared to EA and AA children. AA children had greater lean mass and were reproductively more mature relative to EA and HA children. A correlation matrix of key variables is described in {Table 2}.

Parental feeding practice scores by race/ethnicity are described in {Figure 2}. HA parents reported significantly higher levels of restriction and pressure to eat, whereas EA parents reported the lowest levels of restriction and pressure to eat.

SEM Model

The generally accepted statistical fit cutoffs (Rex B.Kline, 2005), indicate that the model was a good fitting model ($\chi^2=79.80$, df=62, p=0.06, CFI=0.98, RMSEA=0.03) {Figure 3}. Pubertal status and AFADM were significant covariates in the model. Child sex and EUADM were not significant predictors of Child Adiposity, when accounting for all other pathways in the model.

The standardized parameter estimates after adjustment for all other covariates in the model are displayed in {Figure 3}. These standardized path coefficients can be interpreted the same way one would interpret standardized $\beta$ weights in regression analyses. Restriction was a significant predictor of child adiposity ($\beta=0.26$, p<0.0001),
meaning that a 1 standard deviation (SD) shift in Restriction would result in a 0.26 SD increase in Child Adiposity, after adjusting for the contributions of genetic admixture, SES, child sex, and pubertal status. Pressure to Eat was inversely associated with Child Adiposity ($\beta=-0.37$, $p<.0001$), where a 1 SD increase in Pressure to Eat results in a 0.37 SD decrease in Child Adiposity, after controlling for all other factors in the model. There was a significant correlation between Pressure to Eat and Restriction ($r=0.46; p<.0001$). SES was inversely associated with Child Adiposity ($\beta=-0.28$, $p<.0001$), where a 1 SD increase in SES resulted in a -0.28 SD shift in Child Adiposity, after adjusting for all other predictors. SES was inversely associated with both Pressure to Eat ($r=-0.40; p<.0001$) and Restriction ($r=-0.20; p<.0001$).
DISCUSSION AND CONCLUSIONS

The overall purpose of this study was to investigate if parental feeding practices differed by race/ethnicity and evaluate the effects of parental feeding practices and SES on child adiposity. The findings indicate that parental feeding practices differ across racial/ethnic groups, and parental pressure to eat and restriction were associated with adiposity levels. Compared with children of higher SES, the use of controlling parental feeding practices was more prevalent in lower SES homes.

Pressure to eat has been reported to cause chaos and stress during mealtimes and to negatively impact children’s associations with food (Galloway et al., 2006). Considering that negative associations with food may influence eating behavior and child weight, the investigation of pressure to eat on child adiposity is paramount. When children are pressured to eat at mealtimes, they tend to eat less and make more negative comments about food than meals where pressure to eat does not occur. It appears that this relationship is not simply an acute response but pressure to eat promotes picky eating, food aversions, and lower fruit and vegetable consumption during childhood and young adulthood (Batsell et al., 2002; Batsell & Brown, 1998; Galloway et al., 2005). Though studies have identified the influence of pressure to eat on eating behavior, its relationship to BMI has been inconsistent. Studies in children from European descent have reported pressure to eat to be correlated with decreased child BMI-for-age percentiles in the United States (Faith et al., 2004; Matheson et al., 2006; Savage J.S. et al., 2007; Galloway et al., 2006) whereas no associations have been found with weight in children from the United Kingdom (Webber et al., 2010), suggesting a mediating role of cultural and/or geographical context. While studies have found that pressure to eat is associated
with decreased BMI (and, consequently, body weight), a different relationship has been observed with other adiposity measures, such as fat mass in EA children (Spruijt-Metz et al., 2006). In general, to the extent in which the individual measurement of fatness is better represented by a latent variable for child adiposity, our results show that pressure to eat is related to decreased adiposity among multi-ethnic children.

Our results also support previous findings related to the role of parental restriction and BMI-for-age percentiles as a surrogate for pediatric adiposity. It has been hypothesized that controlling a child’s food intake may limit a child’s ability to self-regulate consumption by hindering hunger and satiety cues, potentially leading to eating in the absence of hunger (EAH) (Huon GF, 1994; Fisher & Birch, 2002; Fisher J.O. & Birch L.L., 1999). EAH has been associated with obesity in children (Moens et al., 2007), and longitudinal analysis in EA girls have shown that higher parental restriction at age five relates to EAH by age seven and greater fat mass accrual over time (Fisher & Birch, 2002). However, it is important to note that other studies have found no association between parental restriction and child weight (Webber et al., 2010; Campbell et al., 2010; Carnell & Wardle, 2007; Gregory et al., 2010). It is possible that there is a bilateral relationship between child weight and parental restriction, in that, parents may increase restriction on children who they perceive to be at risk for overweight. This notion has been supported by the work of Rifas-Shiman and colleagues who demonstrated maternal restriction at age one was associated with a higher child BMI at age three before, but not after, adjusting for weight for length at age one (Rifas-Shiman et al., 2011), suggesting parents restrict the food intake of infants who are already overweight. As we scientifically move toward the understanding of behavioral practices
in obesity prevention and health, it will be pertinent to explore how the complex relationships among parental feeding practices, adiposity, weight regulation and health risk factors interact with diverse contexts of culture, locations and social class.

A most relevant aspect of our study is the inclusion of participants from various racial/ethnic groups and broad range of socio-economical factors, adding scientific evidence toward the unclear understanding of the impact of parental feeding practices across populations (Hennessy et al., 2010; Spruijt-Metz et al., 2006; Spruijt-Metz et al., 2002; Mulder et al., 2009). In our cohort, feeding practices differed across racial/ethnic groups, with HA parents reporting higher levels of Pressure to Eat and Restriction relative to AA and EA parents. As parental perceptions of appropriate child feeding varies across groups, with some parents reporting concern about diet quality and others focusing on satiety (Sherry et al., 2004), it is possible that different underlying feeding/food related dynamics are occurring among HA, AA and EA parents.

Racial/ethnic classifications are complex, and they reflect biological and nonbiological factors that might influence adiposity and health-related outcomes both behaviorally and physiologically. The inclusion of estimates of genetic admixture in this study accounts for the biological factors underlying such classification, bringing validity to our findings by reducing potential spurious associations driven by ancestral genes influencing body composition that could have confounded the association between parental practices and adiposity. When investigating the role of SES, we found an inverse association between SES and both parental Pressure to eat and Restriction. This finding suggests that low income populations, who are at increased risk for excess adiposity, may be at higher risk for exhibiting the studied parental feeding practices. National data has demonstrated that
children from low income homes are at higher risk for becoming overweight compared to those of middle income families (Ogden et al., 2010) and studies have reported SES to be inversely related to BMI-for-age percentile (Mikolajczyk & Richter, 2008; Goodman, 1999). Our findings suggest that parental feeding practices may be a contributing factor to childhood overweight in low income populations.

The findings of this research are pertinent in several regards. First, we studied a multi-ethnic and economically diverse group of children. Second, robust measures of body composition were used to assess adiposity. We were also able to control for the role of genetic admixture on adiposity, which is important given that genetic admixture has been shown to contribute to body composition differences in children (Cardel et al., 2011; Casazza K et al., 2010). Additionally, the use of structural equations modeling allowed the model to look at many relationships at once, though this study is limited because of the inability to stratify the model by race/ethnicity due to sample size limitations. Though our study is cross-sectional, and causality cannot be implied, it is important to note the possibility of reverse associations or bi-directionality in parental feeding practices, namely that child adiposity may elicit parental (attempts at) restraint or pressure to eat (Campbell et al., 2010).

In conclusion, the results of this investigation provide support that specific parental feeding practices, such as parental restriction or pressure to eat, are associated with child adiposity. Our findings suggest that Hispanic parents differ from AA and EA parents in their feeding practices and further research is needed to investigate if controlling feeding practices differentially impact adiposity in diverse populations. SES was inversely related to child adiposity and low income families report higher levels of
parental feeding practices, a possible contributor to child overweight. Thus, parental feeding practices appear to differ across racial/ethnic groups and socioeconomic levels and may be associated with population differences in child adiposity.
Figure 1. Theoretical model depicting influences of parental feeding practices and Socioeconomic status with child adiposity.
Table 1. Descriptive characteristics by racial/ethnic groups (mean, SD or n%)

<table>
<thead>
<tr>
<th></th>
<th>AA (n=100)</th>
<th>EA (n=119)</th>
<th>HA (n=86)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (y)</strong></td>
<td>9.7 (1.4)</td>
<td>9.7 (1.6)</td>
<td>9.4 (1.5)</td>
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<tr>
<td><strong>% Female</strong></td>
<td>45.3</td>
<td>48.4</td>
<td>47.7</td>
</tr>
<tr>
<td><strong>Tanner 3 (%)</strong></td>
<td>23.6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.3&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>SES</strong></td>
<td>38.2 (10.7)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>49.0 (9.8)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>26.0 (12.1)&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>BMI %</strong></td>
<td>63.0 (27.7)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>59.7 (27.1)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>77.2 (19.5)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>% fat</strong></td>
<td>20.5 (9.6)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>22.3 (8.4)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>28.2 (8.4)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>TAAT (cm&lt;sup&gt;2&lt;/sup&gt;)</strong></td>
<td>10.6 (9.5)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>12.1 (9.4)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>16.5 (8.4)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>EUADM</strong></td>
<td>0.15±0.01&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.96±0.01&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.35±0.02&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>AFADM</strong></td>
<td>0.82±0.01&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.01±0.00&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.09±0.01&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a, b, c</sup> superscripts indicate significant differences among self-identified racial/ethnic category

EA=European American; AA=African American; HA=Hispanic American; SES=Socioeconomic status; BMI%=BMI-for-age percentile, TAAT=Total Abdominal Adipose Tissue; EURADM=European admixture; AFADM=African Admixture
Figure 2. Mean Parental Feeding Questionnaire Scores by race/ethnicity

![Graph showing mean scores by race/ethnicity.]

a, b, c statistical difference $p \leq 0.05$ between ethnic groups
Table 2. Correlation Matrix between key variables

<table>
<thead>
<tr>
<th></th>
<th>RES</th>
<th>P2E</th>
<th>Trunk Fat</th>
<th>TAAT</th>
<th>BMI%</th>
<th>Sex</th>
<th>Tanner</th>
<th>AFADM</th>
<th>EUADM</th>
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<td></td>
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<td>P2E</td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>Trunk Fat</td>
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<td>-0.08</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>TAAT</td>
<td>0.06</td>
<td>-0.10</td>
<td>0.82*</td>
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<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>BMI%</td>
<td>0.17*</td>
<td>-0.02</td>
<td>0.55*</td>
<td>0.53*</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sex</td>
<td>0.06</td>
<td>0.03</td>
<td>0.10</td>
<td>0.17**</td>
<td>-0.01</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tanner</td>
<td>0.08</td>
<td>0.01</td>
<td>0.28*</td>
<td>0.14</td>
<td>0.09</td>
<td>0.17*</td>
<td>1</td>
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<tr>
<td>AFADM</td>
<td>0.14*</td>
<td>0.19**</td>
<td>-0.12</td>
<td>-0.20*</td>
<td>-0.02</td>
<td>-0.01</td>
<td>0.29*</td>
<td>1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>EUADM</td>
<td>-0.34*</td>
<td>-0.43*</td>
<td>-0.02</td>
<td>0.05</td>
<td>-0.12</td>
<td>0.01</td>
<td>-0.24*</td>
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<tr>
<td>SES</td>
<td>-0.20*</td>
<td>-0.40*</td>
<td>-0.21*</td>
<td>-0.18**</td>
<td>-0.23*</td>
<td>0.01</td>
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<td>-0.15**</td>
<td>0.50*</td>
<td>1</td>
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</tbody>
</table>

Res=Restriction construct; P2E=Pressure to Eat construct; TAAT=Total abdominal adipose tissue; BMI%=BMI-for-age percentile; AFADM=African Admixture; EUADM=European Admixture; SES=Socioeconomic Status

**p<0.05
*p<0.01
Figure 3. Structural equations model investigating the effects of parental feeding practices and socioeconomic status on child adiposity


SCHOOL FOOD ENVIRONMENT, DIET QUALITY, AND ADIPOSITY: COMPARISONS BETWEEN HOME-SCHOOLED CHILDREN AND TRADITIONALLY-SCHOOLED CHILDREN

by

MICHELLE CARDEL, AMANDA L. WILLIG, AKILAH DULIN KEITA, KRISTA CASAZZA, ANDREA CHERRINGTON, DOMINICK J. LEMAS, JOSE R. FERNANDEZ

Submitted to IJO

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ABSTRACT

Background: Children spend a significant amount of time in school; however the contribution of educational environments to pediatric obesity is unclear. Comparing dietary intakes among home-schooled children (HSC) to those of traditionally-schooled children (TSC) is a novel method to elucidate the extent to which educational environments influence diet and child adiposity. Objective: This study quantifies and compares the relationship between diet (with an emphasis on diet quality and lunch time meals) and physical activity on adiposity in HSC and TSC. Methods: Subjects were HSC (n=47) and TSC (n=48) aged 7 to 12. Dietary intakes were obtained via two weekday 24-hour recalls and physical activity was assessed with 7 days of accelerometry. Fat mass (FM), trunk fat, and percent body fat (%BF) were measured by dual-energy x-ray absorptiometry. Analysis of covariance determined whether there were significant differences in body composition and dietary intakes between school groups. Multivariate linear regression analyses evaluated whether school status was associated with diet quality and adiposity. Results: TSC had significantly higher BMI percentiles, FM, trunk fat, and %BF; consumed 120 kcal more per day; and had increased intakes of trans fats, total sugar, added sugars, calcium, and lower intakes of fiber, fruits, and vegetables when compared to HSC. No differences in physical activity were reported. At lunch, TSC consumed more calories (651 kcal) than HSC (533 kcal), and reported higher intakes of total sugar, sodium, potassium, and calcium relative to HSC. Being traditionally schooled was significantly associated with increased consumption of trans fat, total sugar, added sugars (p<.05); lower intakes of fiber, and fruits and vegetables (p<.05); and higher FM, %BF, and trunk fat (p<0.01). Conclusions: These results suggest that educational environments
influence dietary intakes, diet quality, and pediatric adiposity and the traditional school food environment may be related to increased risk of pediatric obesity.

Keywords: obesity, percent body fat, pediatric, school lunch, physical activity, school age children
INTRODUCTION

The prevalence of childhood obesity has increased over the last thirty years (1;2) and research demonstrates that obese children are more likely to become obese adults (3). Pediatric obesity that tracks into adulthood may have detrimental implications for long term health (3-5) and contributes to the development of chronic pediatric health complications such as type 2 diabetes (6), metabolic syndrome (7), and dyslipidemia (8). Given the limited successful treatment options for overweight and obese children (9), cross-sectional studies in pediatric populations that aim to characterize risk factors associated with obesity phenotypes may help inform obesity treatment and prevention programs.

The dramatic rise in the prevalence of pediatric obesity has been linked, in part, to a confluence of behavioral and environmental factors, and the school environment has been investigated as a potential contributor. Traditional school systems have been hypothesized to affect a child’s weight by facilitating an increase in both the availability and intake of energy-dense foods and beverages in schools (10-18) and/or by decreased participation in physical education programs (18). Interestingly, not all studies in pediatric populations have detected associations between traditional school environments and child weight (16;17) and the extent to which educational environments influence diet quality or child adiposity remains unclear.

In order to determine whether the traditional school environment is associated with diet quality and child adiposity, it is conceivable that home-schooled children (HSC) who consume lunch prepared and eaten at home could serve as an ideal and novel comparison group when compared with traditionally-schooled children (TSC) who consume a lunch prepared and obtained from school. In the United States, approximately 2 million
children receive an education in a home-schooled environment, and this number is increasing every year (19). Aspects of HSC and families (i.e. parental motivations to home-school, determinants of the student achievement, and physical activity levels) have been well characterized (20;21); however, to our knowledge, research has not evaluated HSC’s dietary intakes or body composition. Given that the dietary habits and body composition of HSC has largely been overlooked in the obesity literature, inclusion of this population as a comparison group would not only allow for a clearer understanding of the relationship between the school food environment, diet quality, and childhood adiposity but would also be the first study to characterize and evaluate whether dietary intakes and body composition among HSC differ from TSC.

Therefore, the objectives of this study were: 1) to quantitatively measure and compare dietary intakes, physical activity, and body composition in HSC and TSC, and 2) to evaluate the relationship between dietary intakes, diet quality, and physical activity on adiposity in HSC and TSC.
SUBJECTS AND METHODS

Subjects

Participants were 95 children aged 7-12 years self-identified as European American recruited in Birmingham, Alabama. Recruitment of study participants was conducted at schools, churches, health fairs, and through newspapers, parent magazines, radio, and participant referrals. Parents of the participants reported school status [home-schooled; n=47 vs. traditionally-schooled; n=48] for each child in this study. The children were peripubertal (pubertal stage ≤3 as assessed by a pediatrician according to the criteria of Marshall and Tanner) (22;23), and were not taking any medications contraindicated for study participation (i.e. medication known to affect body composition, metabolism, cardiac function). Prior to study participation, children and parents provided informed assent and consent, respectively, to the protocol, which was approved by the Institutional Review Board for human subjects at the University of Alabama at Birmingham. All measurements were collected by trained staff at the General Clinical Research Center and the Department of Nutrition Sciences at the University of Alabama at Birmingham between 2005 and 2008.

Anthropometric measures

Anthropometric measurements for all participants were obtained by the same registered dietitian. Participants were weighed (Scale-tronix 6702W) to the nearest 0.1 kg (in minimal clothing without shoes) and height was recorded to the nearest 0.1 kg without shoes using a digital stadiometer (Heightronic 235; Measurement Concepts, Snoqualmie,
Children’s BMI-for-sex-and-age percentiles (BMI %) were calculated according to the 2000 Center for Disease Control and Prevention growth charts (24).

**Adiposity**

Body composition was measured by dual-energy x-ray absorptiometry (DXA) using a GE Lunar Prodigy densitometer (GE LUNAR Radiation Corp., Madison, WI) with pediatric software encore 2002 version 6.10.029 while participants wore light clothing and laid flat on their backs with arms at their sides. Measured variables for this study included total fat mass (FM), trunk fat, and percent body fat (%BF).

**Socioeconomic Status**

Socioeconomic status (SES) was measured with the Hollingshead 4-factor index of social class (25), which combines the educational attainment and occupational prestige for working parents in the child’s family. Scores range from 8 to 66, with higher scores indicating higher theoretical socioeconomic status. Given that SES has been shown to impact body composition in children (26), SES was used as a covariate in all models related to child adiposity. The SES measure may not address qualification of free/reduced price meals for the National School Lunch Program (NSLP), which may impact diet (27). Thus, regardless of current child schooling status, parents also reported if their child was certified to receive a free/reduced lunch (FREE; 0=No; 1=Yes). In models evaluating school status on dietary intakes and diet quality, we adjusted for the FREE variable.
Dietary Assessment

In this study, we refer to the lunchtime meal as one that was prepared and consumed at home for HSC participants and prepared and consumed at school for TSC participants (whether purchased or given to the child for those who qualified for free/reduced lunch from the NSLP). Dietary composition for each participant was assessed using an average of two weekday 24-hour dietary recalls (24HR). Each 24HR was performed in the presence of the child’s parent and the “multiple pass” method was used, providing a cup and bowl size to help estimate portion sizes (28). Children and parents were asked the location of all meals the children consumed (i.e. home, school, a restaurant) and where each meal was prepared and/or purchased. All TSC included in this study had eaten a school lunch (which included anything that was purchased at school, including NSLP meals, competitive foods, and a la carte items). All HSC had consumed a lunch that was prepared and eaten at home. 24HR data for all children in this study was coded by a registered dietitian and entered into Nutrition Data System for Research version 2006 (Nutrition Coordinating Center, University of Minnesota, Minneapolis, MN), a dietary analysis program designed for the collection and analysis of 24-hour dietary recalls.

Physical Activity Assessment

Children wore an MTI ActiGraph accelerometer (GT1M, ActiGraph Health Services, Pensacola, FL) on their waist over the right hip for 7 days to objectively measure physical activity levels (and removal was limited to times when the child was sleeping, bathing, and/or swimming). Epoch length was set at one minute and physical activity
data in this study was expressed as counts per minute (counts min\(^{-1}\)). The daily counts per minute >1952 counts per minute were summed and analyzed as the average time children spent in moderate and vigorous physical activity (MVPA).

Statistical Analysis

The normality assumptions for the outcome variables were evaluated and non-normal variables were log-transformed. Differences in descriptive statistics between HSC and TSC were explored with independent samples t-tests. Analysis of covariance (ANCOVA) with Tukey’s post hoc analysis was used to evaluate the difference in obesity, dietary, and physical activity variables between HSC and TSC and controlled for age, sex, pubertal stage, and SES. The school variable was dichotomized in all analyses as 0=HSC and 1=TSC. Multivariate linear regression analyses were used to determine whether school status influenced diet quality among HSC and TSC, where homeschooled status was used as the reference group. In this model we also evaluated if qualifying for free/reduced lunch from the NSLP influenced dietary quality; with children who did not qualify for free/reduced lunch as the reference group. Other important covariates included the child’s age and sex, with male as the referent category. Additional multivariate linear regression models investigated whether adiposity variables that included BMI %, FM, trunk fat, and %BF were independently associated with school status, energy intake, and physical activity. Covariates included child age, pubertal status, SES, and child sex with males as the reference group. All analyses were conducted using SAS Version 9.2 (SAS Institute Inc, Cary, NC).
RESULTS

Demographics

Descriptive statistics comparing HSC to TSC are reported in Table 1. Though not statistically significant, the distribution of females was slightly higher in TSC (56.25%) than in HSC (42.55%). No significant differences were observed regarding SES or percent of children who qualify for free/reduced lunch between the two groups.

Body Composition, Dietary Intakes, and Physical Activity

HSC had significantly lower BMI%, FM, trunk fat, and %BF relative to TSC. Comparisons of daily dietary intakes demonstrate TSC (1951 kcal) consumed approximately 120 kcals more daily than HSC (1830 kcal) and reported significantly higher intakes of trans fat, total sugar, added sugars, calcium, and lower consumption of fiber, and fruits and vegetables relative to HSC. Evaluation of the lunchtime meals indicated similar trends with TSC consuming significantly more calories, total sugar, sodium, potassium, and calcium at lunch when compared to HSC. Based on the daily energy need requirements outlined in Dietary Guidelines for Americans (29), both TSC and HSC were consuming excess calories. TSC reported greater consumption of excess calories (420 kcals/day) relative to daily overconsumption by HSC (300 kcals/day). No significant differences were detected between TSC and HSC with respect to the amount of time spent engaged in moderate to vigorous physical activity.
School Status and Dietary Quality

The relationship between school status and dietary patterns was evident after accounting for qualification of free/reduced lunch status, age, and child sex (Table 2). Traditional school status was associated with increased intakes of trans fat ($p=0.0136$), total sugar ($p=0.0342$), added sugars ($p=0.0164$), calcium ($p=0.0109$), and decreased consumption of fiber ($p=0.0408$), and fruits and vegetables ($p=0.0136$). Being traditionally schooled was also marginally associated with increased energy intake ($p=0.0584$). Regardless of current school status, qualifying for a free/reduced lunch program was associated with significantly more percent of calories from fat ($p=0.0223$), and was marginally associated with increased percent of calories from carbohydrate ($p=0.0560$), increased trans fat ($p=0.0548$), and less fiber intakes ($p=0.0670$).

School Status, Energy Intake, Physical Activity, and Body Composition

Regression analysis that tested for significant associations between adiposity variables with school group, energy intake, and physical activity, after adjustment for covariates SES, age, pubertal stage, and sex are reported in Table 3. TSC status was significantly associated with higher FM ($p=0.0011$), %BF ($p=0.0012$), trunk fat ($p=0.0013$), and was nominally associated with higher BMI% ($p=0.0631$). Physical activity was inversely associated with FM ($p=0.0218$), %BF ($p=0.0139$), and trunk fat ($p=0.0150$). Energy intake was not significantly associated with any adiposity variable tested.
DISCUSSION

This study characterized and compared dietary intakes (with an emphasis on diet quality and lunchtime meals), physical activity, and body composition between home-schooled children and traditionally-schooled children. We also evaluated the association between dietary intakes and physical activity on adiposity in home schooled children and traditionally-schooled children. Our findings indicate TSC had significantly higher energy intakes (both daily and at lunchtime meals), poorer diet quality, and higher levels of adiposity, relative to HSC, despite no differences in physical activity. To our knowledge, this is the first study to assess diet and body composition in HSC and is novel in its use of HSC as a comparison group to TSC with the objective of determining the extent to which the traditional school environment influences diet quality or adiposity in children. Thus, our findings indicate that the school environment is associated with a child’s diet quality and adiposity, independent of physical activity and other covariates.

Our analysis observed marked differences in adiposity and energy intake between TSC and HSC. Specifically, we found that TSC had greater FM, %BF, trunk fat, and BMI\% and consumed approximately 120 kcals more per day than HSC. We observed that the difference in daily caloric intake appears to be derived from a difference in calories at the lunchtime meal. This suggests that the lunchtime meals children are choosing or being provided at school are more energy dense than those consumed at home by TSC. This is consistent with previous research findings that describe foods in the National School Lunch Program (NSLP) as being low-nutrient, energy-dense foods (27), that were positively associated with increased caloric intake and higher BMI\% in children attending middle school (30).
Although TSC had increased calorie consumption at lunch relative to HSC, it is important to note that both TSC and HSC were exceeding energy intake recommendations (29). TSC exceeded energy intake recommendations by 420 kcal per day and HSC consumed an excess of approximately 300 kcal per day. The findings of this study are in agreement with data from the National Health and Nutrition Examination Survey 2003-2004 that reported children aged 6-11 consumed approximately 2100 kcal/day, which greatly exceeds this population’s calorie needs (31). Wang and colleagues indicate that on average, overconsumption of 131 kcal per day results in a 2 pound annual excess, suggesting if the observed trend of excess calories in our cohort continues it may increase the risks for overweight and obesity over time (32;33).

The diet quality of school lunches in the United States has been reported to fall below recommended Dietary Guidelines (6;14) and several studies have found diet quality to be inversely associated with adiposity (34;35). The relationship between school status and dietary patterns were evident after accounting for age, sex, and qualification for free/reduced lunch. Specifically, we found that being traditionally schooled was associated with increased consumption of trans fat, total sugar, added sugars, calcium, and lower intakes of fiber, and fruits and vegetables. Recommendations from the American Heart Association (36) suggest that total intake of trans fat should not exceed 1% of total calories, but TSC consumed an average of 6.75 g (which translates to 3.11% of total calories) and HSC consumed an average of 5.43 g (which translates to 2.07% of total calories). Given that increased intakes of trans fat has been consistently linked to coronary heart disease, it is concerning that the traditional school environment is related to higher consumption of trans fat in children of this cohort. Increased intake of sugar has also
been implicated in the development of hypertension, hyperlipidemia, and obesity (37) and American Heart Association recommendations for added sugars in children aged 7-12 range from 12-16 grams. Over 16% of total calories (75.52 g) in HSC and 19.5% of total calories (95.18 g) in TSC were derived from added sugars, both which greatly exceed current recommendations and may be placing both groups of children at higher risk for the development of cardiovascular disease (37). Higher intakes of calcium in the traditional school group are beneficial and may be protective against the development of obesity (38), though neither group is reaching current recommendations for daily calcium intakes (29). With the exception of TSC consuming more calcium relative to HSC, to the extent that study variables describe diet quality, it appears that TSC have poorer diet quality relative to HSC.

Previous studies demonstrate that physical activity levels are associated with differences in body composition in children (39-41), and in our cohort physical activity was inversely associated with FM, %BF, and trunk fat in the total sample. Additionally, we did not detect a difference in physical activity in our sample of TSC and HSC which was consistent with the findings of Welk et al (21). Given that interventions to increase physical activity in schools have shown to be successful in preventing excessive weight gain in youth (42), future research efforts and policy changes need to be made to increase the amount of physical activity and potentially combat the excess of calories being consumed in both TSC and HSC.

The strengths of this study are the use of robust measures used to determine body composition and objective physical activity. In children, DXA has been found to be highly reliable for body composition assessment (43;44) and Actigraph monitors exhibit
a high degree of inter-instrument reliability (45). The inclusion of HSC, a population whose dietary patterns and body composition had, to our knowledge, never been characterized, was important as they are a group that has been largely overlooked in the obesity research. Variables which independently may contribute to body composition including SES, sex, age, race/ethnicity, qualification for free/reduced lunch, or pubertal status did not differ among TSC and HSC. Additionally, we had the ability to account for where lunch time meals were purchased and/or consumed which allowed for evaluating the relationship between lunch time meals and adiposity in children. Although this analysis examined the relationship between the school environment, diet quality, and adiposity, it is cross-sectional and should not be interpreted as the school environment causes obesity in children. Rather, the characterization of HSC and consequently exploring the differences between TSC and HSC was meant to identify areas for further research that will address the question of how school lunches are associated with children’s diet quality and adiposity. This study was limited because sample size was small and limited to a specific geographic region; therefore results may not be generalizeable to all children. The homogeneous sample of European American children is a limitation, though overall demographics for HSC indicate this is typical for homeschooled families nationally (46). We also recognize that the use of HSC as a comparison group to TSC may bring about confounders that are not seen in this paper. However, our data demonstrated that TSC and HSC were homogenous with regards to age, socioeconomic status, qualification for free/reduced lunch, pubertal stage, race/ethnicity, and sex giving us confidence that our cohort of HSC are an appropriate comparison group for this study.
In summary, children spend almost half of their waking hours in educational environments where they expend up to 50% of daily energy and consume approximately a third of their daily caloric intake (10;47). Given the significant portion of time children spend at school; educational environments have the potential to impact nutritional intakes and physical activity levels. Our study suggests that school lunches are a primary source of increased energy intake and are associated with poorer diet quality in children. Additionally, being traditionally schooled was associated with increased adiposity and excess adiposity may increase the risk of cardiometabolic disease and other chronic diseases in children (6-8) and adults (3-5). We acknowledge the efforts that have been made to improve and regulate the school food environment (48;49), however, our results along with others (10-13) demonstrates that the efforts, while a step in the right direction, still fall short. These findings also highlight the need for state and federal policy changes and greater monitoring of the school food environment. Future research efforts and policy should address diet quality in schools and interventions are needed to increase physical activity in both TSC and HSC.

Conflict of Interest: The authors report no conflict of interest.
Table 1. Comparisons of child demographics, body composition, dietary intakes, and physical activity levels between home-schooled children and traditionally-schooled children

<table>
<thead>
<tr>
<th>Variable</th>
<th>Home-Schooled Children (n=47)</th>
<th>Traditionally-Schooled Children (n=48)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Demographics</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sex (% female)</td>
<td>42.55 ± 1.68</td>
<td>56.25 ± 1.69</td>
</tr>
<tr>
<td>Age (yrs)</td>
<td>9.54 ± 1.68</td>
<td>9.61 ± 1.69</td>
</tr>
<tr>
<td>Pubertal stage (Tanner)</td>
<td>1.32 ± 0.63</td>
<td>1.31 ± 0.59</td>
</tr>
<tr>
<td>Socioeconomic status</td>
<td>48.66 ± 10.67</td>
<td>49.38 ± 9.06</td>
</tr>
<tr>
<td>% Free Lunch</td>
<td>9.76</td>
<td>16.67</td>
</tr>
<tr>
<td><strong>Body Composition</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI percentile (%)</td>
<td>54.55 ± 27.52</td>
<td>63.33 ± 25.72*</td>
</tr>
<tr>
<td>Total fat mass (kg)</td>
<td>6.75 ± 3.54</td>
<td>8.41 ± 4.92*</td>
</tr>
<tr>
<td>Percent Body Fat (%)</td>
<td>20.26 ± 7.39</td>
<td>22.76 ± 8.79</td>
</tr>
<tr>
<td>Trunk fat mass (kg)</td>
<td>2.57 ± 1.61</td>
<td>3.36 ± 2.38*</td>
</tr>
<tr>
<td><strong>Daily Dietary Intakes and Physical Activity</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Energy Intake (kcal)</td>
<td>1830.25 ± 412.19</td>
<td>1950.80 ± 409.37*</td>
</tr>
<tr>
<td>Estimated Energy Requirement (kcal)</td>
<td>1533.33 ± 235.51</td>
<td>1529.17 ± 216.31</td>
</tr>
<tr>
<td>% Calories from carbohydrate</td>
<td>52.23 ± 7.07</td>
<td>51.89 ± 6.47</td>
</tr>
<tr>
<td>% Calories from protein</td>
<td>14.20 ± 3.59</td>
<td>13.96 ± 2.78</td>
</tr>
<tr>
<td>% Calories from fat</td>
<td>33.47 ± 5.74</td>
<td>34.12 ± 5.24</td>
</tr>
<tr>
<td>% Calories from SFA</td>
<td>12.15 ± 3.00</td>
<td>12.70 ± 2.38</td>
</tr>
<tr>
<td>Trans fat (g)</td>
<td>5.43 ± 2.33</td>
<td>6.75 ± 2.83*</td>
</tr>
<tr>
<td>Total sugar (g)</td>
<td>113.34 ± 35.68</td>
<td>127.23 ± 40.53*</td>
</tr>
<tr>
<td>Added sugars (g)</td>
<td>75.52 ± 37.46</td>
<td>95.18 ± 38.95*</td>
</tr>
<tr>
<td>Sodium (mg)</td>
<td>2964.11 ± 861.91</td>
<td>3157.78 ± 857.37</td>
</tr>
<tr>
<td>Potassium (mg)</td>
<td>2079.86 ± 682.79</td>
<td>2107.65 ± 573.05</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>804.58 ± 354.91</td>
<td>947.94 ± 279.82*</td>
</tr>
<tr>
<td>Fiber (g)</td>
<td>14.77 ± 5.84*</td>
<td>12.83 ± 4.67</td>
</tr>
<tr>
<td>Daily Fruit and Vegetable servings</td>
<td>4.98 ± 2.64*</td>
<td>2.59 ± 1.81</td>
</tr>
<tr>
<td>MVPA (min/d)</td>
<td>55.79 ± 30.94</td>
<td>61.65 ± 31.73</td>
</tr>
<tr>
<td><strong>Dietary Variables of Lunchtime Meal</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average caloric intake (kcal)</td>
<td>533.01 ± 244.46</td>
<td>650.72 ± 302.27*</td>
</tr>
<tr>
<td>% Calories from carbohydrate</td>
<td>51.67 ± 11.99</td>
<td>52.07 ± 11.99</td>
</tr>
<tr>
<td>% Calories from protein</td>
<td>15.24 ± 5.38</td>
<td>14.03 ± 5.10</td>
</tr>
<tr>
<td>% Calories from fat</td>
<td>32.95 ± 10.59</td>
<td>33.85 ± 10.59</td>
</tr>
<tr>
<td>% Calories from SFA</td>
<td>10.27 ± 4.82</td>
<td>11.70 ± 4.00</td>
</tr>
<tr>
<td>Trans fat (g)</td>
<td>1.73 ± 1.19</td>
<td>2.09 ± 1.30</td>
</tr>
<tr>
<td>Total sugar (g)</td>
<td>51.11 ± 38.97</td>
<td>63.05 ± 40.49*</td>
</tr>
<tr>
<td>Added sugar (g)</td>
<td>20.92 ± 19.45</td>
<td>25.74 ± 20.74</td>
</tr>
<tr>
<td>Sodium (mg)</td>
<td>937.68 ± 528.60</td>
<td>1085.13 ± 581.26*</td>
</tr>
<tr>
<td>Potassium (mg)</td>
<td>548.82 ± 291.57</td>
<td>818.29 ± 492.74*</td>
</tr>
<tr>
<td>Calcium (mg)</td>
<td>183.67 ± 117.98</td>
<td>321.65 ± 206.97*</td>
</tr>
<tr>
<td>Fiber (g)</td>
<td>4.41 ± 2.85</td>
<td>5.19 ± 3.61</td>
</tr>
</tbody>
</table>
* indicate differences between groups, p<0.05

\(^a\) Analysis of covariance (ANCOVA) with Tukey’s post hoc analysis was used to evaluate the difference in obesity, dietary, and physical activity variables between HSC and TSC and controlled for age, sex, pubertal stage, and SES

\(^b\) Analysis of covariance (ANCOVA) with Tukey’s post hoc analysis was used to evaluate the difference in obesity, dietary, and physical activity variables between HSC and TSC and controlled for age, sex

\(^c\) According to recommendations from Dietary Guidelines for Americans

\(^d\) In traditionally-schooled children, lunch time meal was a school lunch that was purchased/provided and consumed at school; In home-schooled children, lunch time meal was prepared and consumed at home

\(^\%\) Free Lunch=Percentage of the children whose parents report that qualify for a free/reduced National School Lunch Program Lunch; BMI percentile= BMI-for-sex-and-age percentiles; MVPA=Total min/day spent in moderate to vigorous physical activity; SFA=Saturated Fatty Acids
Table 2. The relationship between school status (traditional-schooled vs. home-schooled) and diet quality patterns among home-schooled and traditionally-schooled children

<table>
<thead>
<tr>
<th>Variable</th>
<th>Total Energy (kcal)</th>
<th>Carbohydrate</th>
<th>Protein</th>
<th>Fat</th>
<th>Saturated Fat</th>
<th>Trans Fat (g)</th>
<th>Total Sugar (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>TSC †</td>
<td>1344853.35</td>
<td>0.035†</td>
<td>-1.23144</td>
<td>0.389</td>
<td>0.384</td>
<td>1.12812</td>
<td>0.3195</td>
</tr>
<tr>
<td>Free ‡</td>
<td>20.11053.31</td>
<td>0.357</td>
<td>-0.33323</td>
<td>0.5425</td>
<td>0.5425</td>
<td>0.82953</td>
<td>0.53129</td>
</tr>
<tr>
<td>Added Sugar (g)</td>
<td>Sodium (mg)</td>
<td>Potassium (mg)</td>
<td>Calcium (mg)</td>
<td>Fiber (g)</td>
<td>Fruit and Veg.</td>
<td>MVPA.</td>
<td></td>
</tr>
<tr>
<td>TSC †</td>
<td>30.468.34</td>
<td>0.614†</td>
<td>-0.394357.42</td>
<td>0.365</td>
<td>-0.394357.42</td>
<td>0.365</td>
<td>100.62324</td>
</tr>
<tr>
<td>Free ‡</td>
<td>-1.361.931</td>
<td>0.499</td>
<td>10.82593.39</td>
<td>0.3514</td>
<td>-13.32251.90</td>
<td>0.0585</td>
<td>3.91225.03</td>
</tr>
</tbody>
</table>

*Superscripts and bolded indicates a significant association with a cutoff at p<0.05
† Superscripts indicates a trend toward significant with a cutoff at p<0.10
a All models adjusted by age and sex
b Percentage of total calories
c Daily fruit and vegetable servings
d Moderate to Vigorous Physical Activity
e TSC=Traditional Schooled Children’s group
f Free=Qualify for Free/reduced National School Lunch Program
Table 3. The relationship between school group, energy intake, and physical activity with adiposity in home-schooled children and traditionally-schooled children

<table>
<thead>
<tr>
<th></th>
<th>BMI Percentile</th>
<th>Total Fat Mass</th>
<th>Percent Body Fat</th>
<th>Trunk Fat</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>β</td>
<td>p-value</td>
<td>β</td>
<td>p-value</td>
</tr>
<tr>
<td>TSC Group</td>
<td>12.2</td>
<td>0.0631</td>
<td>0.36</td>
<td>0.0011*</td>
</tr>
<tr>
<td>MVPA</td>
<td>0.07</td>
<td>0.5173</td>
<td>-0.27</td>
<td>0.0218*</td>
</tr>
<tr>
<td>Kcal/day</td>
<td>-0.00</td>
<td>0.8597</td>
<td>-0.24</td>
<td>0.3892</td>
</tr>
<tr>
<td>Pubertal Stage</td>
<td>12.3</td>
<td>0.0252*</td>
<td>0.23</td>
<td>0.0187*</td>
</tr>
<tr>
<td>SES</td>
<td>0.08</td>
<td>0.8127</td>
<td>-0.26</td>
<td>0.2667</td>
</tr>
<tr>
<td>Age</td>
<td>-</td>
<td>-</td>
<td>0.45</td>
<td>0.2202</td>
</tr>
<tr>
<td>Female</td>
<td>-</td>
<td>-</td>
<td>0.27</td>
<td>0.0433*</td>
</tr>
</tbody>
</table>

*Superscripts and bolded indicates a significant association with a cutoff at p<0.05  
† Superscripts indicates a trend toward significance with a cutoff at p<0.10

BMI percentile= BMI-for-sex-and-age percentiles; TSC Group=Traditionally-Schooled Children; MVPA= Average minutes/day spent in Moderate to Vigorous Physical Activity; Kcal/day=Average of total calories per day based on two week-day 24-hour recalls; SES=Socioeconomic Status
REFERENCE LIST


AFRICAN GENETIC ADMIXTURE IS ASSOCIATED WITH BODY COMPOSITION AND FAT DISTRIBUTION IN A CROSS-SECTIONAL STUDY OF CHILDREN

by

MICHELLE CARDEL, PAUL B. HIGGINS, AMANDA L. WILLIG, AKILAH DULIN KEITA, KRISTA CASAZZA, BARBARA A. GOWER, JOSE R. FERNANDEZ


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ABSTRACT

Objective: Although differences in body composition parameters among African American (AA), Hispanic American (HA) and European American (EA) children are well documented, the factors underlying these differences are not completely understood. Environmental and genetic contributors have been evaluated as contributors to observed differences. This study evaluated the extent to which African or European ancestral genetic background influenced body composition and fat distribution in 301 peripubertal AA (n=107), HA (n=79), and EA (n=115) children ages 7-12.

Design: Estimates of African admixture (AFADM) and European admixture (EUADM) were obtained for every subject using 142 ancestry informative DNA markers. Dual energy X-ray absorptiometry (DXA) and Computed Tomography (CT) scanning were used to determine body composition and abdominal fat distribution, respectively. Multiple regression models were conducted to evaluate the contribution of admixture estimates to body composition and fat distribution.

Results: Greater AFADM was associated with lower fat mass (p=0.0163), lower total abdominal adipose tissue (p=0.0006), lower intra-abdominal adipose tissue (p=0.0035), lower subcutaneous abdominal adipose tissue (p=0.0115) and higher bone mineral content (p=0.0253), after adjusting for socioeconomic status, sex, age, height, race/ethnicity, and pubertal status. Greater EUADM was associated with lower lean mass (p=0.0056).

Conclusion: These results demonstrate that ancestral genetic background contributes to racial/ethnic differences in body composition above and beyond the effects of
racial/ethnic classification, and suggest a genetic contribution to total body fat accumulation, abdominal adiposity, lean mass, and BMC.

**Key words:**

European genetic admixture, race/ethnicity, abdominal adiposity, bone mineral content, lean mass, obesity.
INTRODUCTION

Differences in body composition among African American (AA), European American (EA), and Hispanic American (HA) children have been well documented in the scientific literature (1-3). Studies have consistently shown that AA children have greater bone mineral content (BMC) and lean mass (LM) than EA children and HA children (4;5). Additionally, EA and HA children accumulate significantly greater amounts of total fat mass (FM) and intra abdominal adipose tissue (IAAT) relative to AA children (2;3;6;7). Population differences in adipose accumulation might impact pediatric growth and development, tracking into adulthood (8), and translating into risk factors for metabolic disorders. It is important, therefore, to identify and investigate factors that may contribute to early differentiation of individual’s body composition and fat distribution.

Genetic and environmental factors have been evaluated as contributors to body composition and energy balance in children. Genetic contributions to pediatric obesity have also been documented in children (9-12). Some candidate genes that contribute to obesity among adults have been identified in pediatric samples (12) and evidence suggests that the contributions of genes may increase obesity risk as age increases (13). Scientific research supports the role of socio-environmental factors in pediatric adiposity, particularly by their influence on physical activity and energy-dense food consumption (14-18). Evaluation of the influence of genes and environments on pediatric adiposity in children growing up during the obesity epidemic evidence a strong genetic influence despite the exposure to the obesogenic environment (9). As the biological integration of populations continues to increase, the extent to which ancestral genetic background
influences disease risk and contributes to genetic variability becomes increasingly paramount toward understanding differences in obesity-related outcomes.

Research initiatives have considered ancestral genetic background as an explanation for population differences related to obesity and body composition (19-25). Estimates of genetic admixture (ADM) quantify ancestral background using ancestry informative markers (AIMs) that differ in allelic frequency between parental populations who intermated at some historical time, creating new admixed populations. In the United States, the contributions of West African, European, and Amerindian parental populations have been significantly associated with body composition parameters in adults (20;24;26;27). However, little is known about how ADM affects body composition and fat distribution in children. The aim of this study was to investigate the extent to which individual estimates of African ancestral admixture (AFADM) and European ancestral admixture (EUADM) contribute to body composition and fat distribution in a cross-sectional study of AA, HA and EA children.
METHODS

Subjects and Design

Three hundred and one AA (n=107), HA (n=79), and EA (n=115) healthy children aged 7-12 years from Birmingham, Alabama, participated in this cross-sectional study. Race/ethnicity was assigned according to parental self-report. A pediatrician determined children’s pubertal status according to the criteria of Marshall and Tanner (28;29). Children were excluded from the study if they took any medications known to alter body composition, if the girls had started menstruation, or if the children were Tanner Stage >4. The majority of the children in the study were of normal weight (< 85th percentile for BMI as defined by the CDC at http://www.cdc.gov/healthyweight/assessing/bmi/childrens_BMI/about_childrens_BMI.html). All children and parents gave informed assent and consent, respectively, before participation. The study was approved by the University of Alabama at Birmingham Institutional Review Board for the use of human subjects in research.

Body composition, Anthropometrics, and fat distribution assessment

Total FM, LM, and BMC was assessed by dual-energy X-ray absorptiometry (DXA) using a GE Lunar Prodigy densitometer (GE LUNAR Radiation Corp., Madison, WI). Subjects were scanned in light clothing, while lying flat on their backs with arms at their sides. DXA has been found to be highly reliable for body composition assessment in children (30-32). Height was measured to the nearest centimeter using a wall-mounted stadiometer, and weight was measured on an electronic scale while children wore light clothing.
Computed Tomography (CT) scanning was used to quantify the distribution of abdominal adipose tissue as total abdominal adipose tissue (TAAT), IAAT, and subcutaneous abdominal adipose tissue (SAAT). A HiLight /Advantage scanner (General Electric, Milwaukee, WI) was used to perform a single slice (5mm) scan of the abdomen at the level of the umbilicus. The scan was analyzed as a cross-sectional area of adipose tissue using Hounsfield units for adipose tissue of –190 to –30. CT allows for accurate measurements of body fat distribution in children (33;34).

Race/Ethnicity

Self-reported “race/ethnicity” may not provide an accurate assessment of both the biological and environmental assumptions often associated with the term, making scientific evaluation of population-based differences challenging. Further, race/ethnicity is self-reported and can vary according to generation, historical periods, social dynamics, and as individuals become more admixed. In our analysis, statistical models include race/ethnicity as a control variable for social and cultural characteristics. Although there is multi-collinearity between the admixture variables and race/ethnicity, race/ethnicity measures a social/contextual construct (35) whereas genetic admixture measures genetic ancestral background (36). Therefore, these two measures may provide insight into two different measurement constructs and should not be used interchangeably (37).

Genotyping and determination of genetic admixture

Genetic admixture estimates were obtained from genotyping 142 AIMs across the human genome informative for European, African and Amerindian ancestry. Genotyping
for the measures of genetic admixture was performed at Prevention Genetics
(www.preventiongenetics.org) using the McSNP method and agarose gel electrophoresis,
as previously described (36). Molecular techniques for the allelic identification and
methodology for genetic admixture application have been described elsewhere (26).
Information regarding marker sequences, experimental details, and parental population
allele frequencies has been submitted to dbSNP (http://www.ncbi.nlm.nih.gov/SNP/)
under the handle PSU-ANTH. Individual admixture estimates were derived using
maximum likelihood (ML) method, as was previously described (38). The ML method
estimates the proportion of genetic ancestry for an individual, using a range of
proportions from 0 to 1 and identifies the most probable value of admixture based on the
observed genotypes.

Socioeconomic status

Socioeconomic status (SES) was determined according to the Hollingshead four
factor index of social status (39). This scale combines the education level and
occupational prestige for the number of working parents in each child’s family. Social
class scores using this scale range from 8 to 66, with higher values represent a higher
SES. SES have been shown to be related environmental factors, such as physical activity
(40) and diet (40;41). Therefore, SES served as a proxy for the environment in the
models.
Statistical analysis

For the purpose of descriptive comparisons, ANOVA was used to detect differences in the variables of interest according to racial/ethnic groups. Multiple linear regression models were used for statistical analysis to evaluate the relationship of ADM with body composition variables. Values of IAAT, SAAT, FM, TAAT, LM, and BMC were log transformed for normality after visual inspection of residuals from the regression equations. Exploratory models were conducted to test for significant contributions of AFADM, EUADM, sex, age, pubertal status, height, race/ethnicity, and SES on each body composition variable. The measured value for each genetic admixture component adds to one; therefore to avoid overspecification of the statistical models, only European and African admixture were included in the models. AFADM and EUADM were chosen because they have the greatest range of variability among the AA, HA, and EA participants in this sample. Data were analyzed using SAS statistical software version 9.1 (SAS Institute, Cary, NC, 2002). Statistical significance was set at $P<0.05$. 
RESULTS

Descriptive statistics for the children by self-reported race/ethnicity are presented in Table 1. AFADM levels ranged from 15 to 100% in children who self-identified as AA, from 0 to 29% in children self-identified as EA, and from 0 to 42% in children self-identified as HA. EUADM levels ranged from 0 to 71% in AA, from 47 to 100% in EA, and from 3 to 90% in HA children. SES differed among the three groups, with EA having the highest levels and HA having the lowest (p < .05). HA children had greater fat mass, percent body fat, BMI percentile, IAAT and SAAT than EA or AA children (p < .05). When compared to HA or EA children, AA children had greater lean mass and BMC (p < .05).

Results for the multiple regression models assessing the contribution of AFADM and EUADM to parameters of body composition and fat distribution are shown in Table 2. AFADM was inversely associated with FM (p=0.0163), TAAT (p=0.0006), IAAT (p=0.0035), SAAT (p=0.0115), and positively associated with BMC (p=0.0253). Greater EUADM was associated with lower LM (p=0.0056). These associations were independent of SES, age, pubertal status, height, sex, race/ethnicity, and total fat (where applicable, see Table 2). Neither self-reported race/ethnicity, nor SES was significantly associated with any fat parameter. BMC was inversely associated with SES.
DISCUSSION

Epidemiological research has demonstrated that there are racial/ethnic differences in body composition and fat distribution in children (2-7), which provides evidence that population-based differences in body composition parameters start early in the life course (3;7;42;43). The question, however, that remains unanswered is how early genetic and environmental factors influence those obesity-related parameters that account for population differences. Our results suggest that ancestral genetic background exerts an influence on levels of adiposity as early as 7 years of age and that this influence is independent of non-genetic factors. Research conducted among adults has shown that SES attenuates the relationship between admixture and diabetes but does not eliminate the significant contributions of admixture to diabetes status (44). Our results suggest that levels of body fat in early stages of development are controlled by ancestral genetic makeup, an observation that warrants further study and that might result in the development of intervention strategies targeted to individuals of admixed ancestry.

The findings of this investigation also validate previous research showing that AFADM influences obesity-related outcomes among individuals of admixed backgrounds (20;25). Although no other studies have examined the relationship between admixture with total fat and lean mass in children, our team has previously reported associations between genetic admixture with bone mineral content (22), insulin sensitivity (23), and insulin-like growth-factor (19) as well as associations between AFADM and total fat and lean mass in adults (21;25). The association between higher levels of AFADM and lower SAAT, IAAT, and TAAT is supported by previous research indicating that most of the variability in abdominal adiposity is attributable to genetic
influences (9). Consistent with a previous study (27), EUADM was inversely associated with LM. Our results, however, suggest that at the early stage of life, EUADM is not predictive of fat accumulation or distribution.

To the extent to which SES serves as a proxy for environmental influences, our analyses indicate that during childhood the variability in body composition aspects that underlie racial/ethnic disparities is not accounted for by the “environment”. Although research has documented that higher SES operates as a protective factor for obesity in children from developed countries whereas the reverse trend occurs among developing nations (45-48), recent estimates suggest that the effects of SES on obesity are weakening in the United States (49). The observation of a weakening SES effect is in concordance with the results, and also supports previous work conducted in this cohort documenting that outcomes reflecting the social environment are not associated with body composition measures and do not appear to be salient factors at this early stage of the life course (50). The study and understanding of the environmental determinants, their interaction with genetic aspects and their influence on racial/ethnic differences in obesity-related outcomes is an area in need of further research.

While the findings of this study are important, it is not without limitations. The majority of previous studies on body composition in children have used anthropometrics to examine racial/ethnic differences in fat distribution (51); hence, our robust measures utilizing DXA and CT scans expand on previous findings. Although we accounted for racial/ethnic groups and SES in the models, it is important to stress that race/ethnicity and/or SES variables do not solely account for the extensive cultural and socio-demographic factors that differ by race/ethnicity and could potentially contribute to
variation in adiposity levels. Additionally, this study uses a cross-sectional design, and the findings will require further verification in a longitudinal analysis.

In summary, the data suggest a role for AFADM and EUADM in the etiology of racial differences in body composition and abdominal fat distribution during childhood. Further investigation is required to identify the specific genes that account for racial/ethnic variation in fat distribution and fat accumulation.
ACKNOWLEDGMENTS

This research was supported by National Institutes of Health grants R01 DK 51684-01, R01 DK 49779-01, National Institutes of Health CA 47888 Cancer Prevention and Control Training Program and National Institutes of Health CA 3R25CA047888-19S1 CURE supplement Cancer Prevention and Control Training Program; General Clinical Research Center grant M01 RR000032 from the National Center for Research Resources; and Clinical Nutrition Research Unit grant P30-DK56336.

Conflict of interest: The authors declare no conflict of interest.
Table 1. Characteristics of the study population by ethnic group (mean, SD).

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>EA (n=115)</th>
<th>AA (n=107)</th>
<th>HA (n=79)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>9.67 (1.67)</td>
<td>9.55 (1.49)</td>
<td>9.43 (1.59)</td>
</tr>
<tr>
<td>Sex (% female)</td>
<td>46.96</td>
<td>45.79</td>
<td>49.37</td>
</tr>
<tr>
<td>Tanner stage (% Tanner stage 3)</td>
<td>7.89&lt;sup&gt;b&lt;/sup&gt;</td>
<td>23.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>7.69&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>SES (Hollingshead 4-factor index)</td>
<td>49.12 (9.47)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>36.51 (11.71)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>25.87 (11.67)&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>BMI percentile</td>
<td>59.71 (26.83)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>62.79 (27.26)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>79.51 (18.11)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>35.56 (8.73)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>36.72 (9.86)</td>
<td>38.08 (10.09)</td>
</tr>
<tr>
<td>Height (cm)</td>
<td>140.21 (10.54)</td>
<td>140.63 (10.45)</td>
<td>137.38 (10.65)</td>
</tr>
<tr>
<td>Lean Mass (kg)</td>
<td>25.43 (5.0)&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>26.78 (5.55)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>24.77 (5.04)&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Fat Mass (kg)</td>
<td>8.17 (5.11)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>7.69 (5.72)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.26 (5.83)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Percent Body Fat</td>
<td>22.22 (8.48)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>19.92 (9.09)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>28.86 (8.17)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>BMC</td>
<td>12.40 (2.72)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>13.55 (3.52)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>12.50 (2.94)&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>IAAT (cm&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>34.28 (22.63)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>26.58 (18.56)&lt;sup&gt;c&lt;/sup&gt;</td>
<td>42.31 (24.50)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SAAT (cm&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>85.92 (73.71)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>79.73 (80.82)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>123.81 (62.91)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>TAAT (cm&lt;sup&gt;2&lt;/sup&gt;)</td>
<td>120.20 (94.12)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>106.32 (96.19)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>166.12 (83.62)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Percent European Admixture</td>
<td>96.19 (3.71)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>16.28 (14.91)&lt;sup&gt;c&lt;/sup&gt;</td>
<td>37.77 (17.97)&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Percent African Admixture</td>
<td>0.56 (1.35)&lt;sup&gt;c&lt;/sup&gt;</td>
<td>79.04 (15.48)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6.39 (8.14)&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Percent Amerindian Admixture</td>
<td>3.24 (3.47)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.68 (4.12)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>55.84 (19.17)&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a,b,c</sup> indicate significant differences between the groups with an alpha level of <0.05.

SES=socioeconomic status; BMI%= body mass index percentile; BMC = bone mineral content;
IAAT=intra abdominal adipose tissue; SAAT= subcutaneous abdominal adipose tissue;
TAAT=total abdominal adipose tissue
Table 2. Multiple Linear Regression results for the association of African and European Admixture with body composition and fat distribution.

<table>
<thead>
<tr>
<th></th>
<th>Total Sample (n=301)</th>
<th>African Genetic Admixture Estimate</th>
<th>European Genetic Admixture Estimate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat Mass</td>
<td>-0.48</td>
<td><strong>0.0163</strong>*</td>
<td>0.06</td>
</tr>
<tr>
<td>Total abdominal adipose tissue</td>
<td>-0.33</td>
<td><strong>0.0006</strong>*</td>
<td>-0.15</td>
</tr>
<tr>
<td>Subcutaneous abdominal adipose tissue</td>
<td>-0.42</td>
<td><strong>0.0115</strong>*</td>
<td>-0.08</td>
</tr>
<tr>
<td>Intra abdominal adipose tissue</td>
<td>-0.57</td>
<td><strong>0.0035</strong>*</td>
<td>-0.34</td>
</tr>
<tr>
<td>Bone mineral content</td>
<td>0.11</td>
<td><strong>0.0253</strong>*</td>
<td>0.08</td>
</tr>
<tr>
<td>Lean Mass</td>
<td>-0.04</td>
<td>0.1996</td>
<td><strong>-0.13 0.0056</strong>*</td>
</tr>
</tbody>
</table>

All models have been adjusted by sex, age, socioeconomic status, height, ethnicity, and pubertal status. The models for IAAT, SAAT, and TAAT have also been adjusted for total fat. The model for fat mass was adjusted by lean mass. Significance of alpha<0.05 is shown in bold with an *
REFERENCE LIST


(39) Hollingshead AB. Four factor index of social status. Yale University Press. 1975. Ref Type: Journal (Full)


GENERAL DISCUSSION

Excess adiposity is related to increased metabolic risk factors in children as young as 7 years of age (26) as well as in adults (3-6). Given the detrimental long-term effects associated with excessive fat accumulation and the limited long-term success with current treatment options (9), studies characterizing the etiological factors that contribute to increased adiposity are paramount in the development of strategies to potentially prevent and clinically treat obesity and its comorbidities. The overall objective of this work was to investigate less-explored genetic and environmental factors that contribute to variation in levels of pediatric adiposity after accounting for caloric intake and physical activity (factors that are traditionally evaluated as determinants of excess adiposity). This dissertation specifically focused on the evaluation of parental feeding practices, the school food environment, and genetic admixture as contributors to fat accrual in children. Particular attention was placed on understanding differences in body composition parameters across populations, including different racial/ethnic groups, diverse socioeconomic levels, and children who attended school in non-traditional settings.

The objectives of this study were centered in three specific aims. We aimed to investigate the relationship between parental restriction and parental pressure to eat on child adiposity (Aim 1), to evaluate the role of the school environment (home-schooled vs. traditionally schooled) on diet and child adiposity (Aim 2), and to determine the extent to which individual estimates of African genetic admixture and European genetic admixture contribute to body composition and fat distribution (Aim 3). The populations
investigated were a cohort of EA, AA, and HA children (mean age 9.6 ± 1.5 yr., mean BMI-for-age-and-sex percentile, 67.0 ± 24.4) (Aim 1 and 3). A sub-cohort of EA homeschooled children (mean age 9.5 ± 1.7 yr., mean BMI-for-age-and-sex percentile, 54.6 ± 27.5) and EA traditionally schooled children (mean age 9.6 ± 1.7 yr., mean BMI-for-age-and-sex percentile, 63.3 ± 25.7) composed the sample of Aim 2.

The main outcome variable of this study was total adiposity (with abdominal adiposity included in Aim 3), given the association of excessive body fat with risk for cardiometabolic diseases. We deviated from considering BMI% as the main outcome variable. Although BMI% is a widely used surrogate measure of body fat, it is a measure of excess weight relative to height, rather than excess adiposity (24). This was qualified by our preliminary analyses showing that the relationship between BMI% with percent body fat and total fat mass was not statistically strong (r=0.6011 and 0.5590, respectively). Our analyses also supported previous observations of the relationship between BMI% and adiposity differing across populations (24;84). When stratified by race, we found correlations between BMI% and percent body fat of r =0.5488 in EA, r = 0.6128 in AA and r = 0.5351 in HA, and correlations between BMI% and total body fat of r = 0.5151 in EA, r = 0.5919 in AA; r = 0.4911 in HA. This suggests that, in the best-case scenario, BMI% accounts for only 37% of the variation in measurements of adiposity in our cohort, highlighting the limitations of its use, particularly when studying diverse populations.

In line with previous studies demonstrating racial/ethnic disparities (28;85-87), differences in body composition parameters were observed across populations (Aim 1 and 3). HA children have significantly higher percent body fat (%BF), total fat mass
(FM), intra-abdominal adipose tissue (IAAT), subcutaneous abdominal adipose tissue (SAAT), and total abdominal adipose tissue (TAAT), relative to EA and AA children. AA children had significantly lower IAAT, greater bone mineral content (BMC), and greater lean mass (LM) compared to EA and HA children. The relationships observed are similar to work demonstrating that at a similar BMI%, EA and HA children have significantly more total fat and intra-abdominal fat and less lean mass and bone mineral content relative to African-American children (28;29;85;87).

The biological basis of the etiology of the observed body composition differences by race/ethnicity were explored by the use of the genetic admixture approach. Specifically, we determined the extent to which individual estimates of African genetic admixture and European genetic admixture contributed to body composition and fat distribution differences across our diverse populations. The findings of our study support the importance of including genetic admixture in clinical research exploring the role of body composition in disease risk. As the biological integration on populations continues to increase (74), accounting for the extent to which ancestral genetic variability influences adiposity becomes paramount to understanding differences in body composition across populations. Our approach also serves as an example of how race/ethnicity could be statistically modeled by biological and non-biological factors. Though there is multi-collinearity between the admixture variables and race/ethnicity, race/ethnicity accounts for measures of social/cultural constructs (77), whereas genetic admixture objectively measures ancestral background based on genetic information (78).

In Aim 3, European genetic admixture was inversely associated with lean mass, which is consistent with previous work conducted in older adults (80), but was not
associated with any adiposity variable. This suggests European genetic admixture is not related to fat accumulation or distribution this early in the life course and is supported by our association between European genetic admixture and the child adiposity latent variable in Aim 1. Conversely, in the analyses for both Aims 1 and 3, our results showed increased African genetic admixture associated with lower child adiposity and negatively associated with abdominal adiposity (Aim 1). The association with TAAT, SAAT, and IAAT is consistent with previous work demonstrating that the majority of the variation in abdominal adiposity is attributed to genetic influences (88). The relationship between African genetic admixture and body composition was also supported by a positive association between admixture and BMC. Taken together, our findings suggest that ancestral genetic background impacts body composition parameters – an influence that can be detected as early as 7 years of age, independent of non-genetic factors. Based on our results, it appears that African admixture may be protective against the development of excess adiposity, particularly abdominal adiposity, a fat depot highly correlated with metabolic risk for disease. At first glance, this is inconsistent with reports showing AA children have the highest rates of obesity prevalence (1). However, it is important to keep in perspective that traditional cut points for overweight and obese classification represent different levels of body fat at the same BMI% in AA and HA children relative to EA children (24;89). To the extent to which body fat represents the risk factor for disease, there is a need to create scientific awareness of the limitations of using BMI% in epidemiological studies to draw conclusions that could result in inaccurate public health recommendations not applicable to all sectors of the population.
One aspect of the environment assessed as a potential predictor of pediatric adiposity was the home environment, focusing on the evaluation of the relationship between parental restriction and parental pressure to eat on body fat in children. Our results indicate that parental pressure to eat was associated with decreased child adiposity, even after adjustment for genetic admixture, socioeconomic status, child sex, and pubertal status. This may be because pressure to eat can cause stress and chaos during mealtimes, which can decrease consumption of the pressured food (57) by developing an aversion toward the foods children were pressured to eat (56). Although it may be assumed that a behavior that is associated with decreased adiposity could be beneficial, this behavior tends to have a negative impact in health because parents pressure their children to eat fruits and vegetables, which might result in a reduction of a nutrient-rich diet (90). Given that this study is cross-sectional, cause and effect cannot be assumed, and future studies should evaluate this observation prospectively to determine the long-term relationship between parental pressure to eat, adiposity, and food aversion. Nonetheless, overcoming the detrimental consequences of pressure to eat will require awareness and behavioral practices. It has been suggested that increasing exposure to new foods, sometimes up to 20 times, might facilitate children’s acceptance and consumption of new foods (90-93), an approach that might be incorporated as a potential strategy to prevent the consequences of pressure to eat.

Adding to previous work on the association between parental restriction and BMI% (51;52;94;95), our study enhanced the scientific literature by evaluating the role of parental restriction in a composite measure of child adiposity. To the extent that the child adiposity latent variable (comprised of BMI%, TAAT, and trunk fat) represents
adiposity and fat distribution, our results show that increased parental restriction is related to both increased child weight and body fat outcomes. Although this might seem counterintuitive, restriction of certain foods in children, particularly when intended to follow a pre-structured meal time schedule, may impair the child’s ability to self-regulate hunger and satiety cues. This restriction can also promote eating in the absence of hunger, particularly of those restricted foods. The impact of parental restrictions to eat can limit a child’s ability to recognize their own hunger, to eat when hungry, and to stop when full, which could result in children’s inability to self-regulate their energy intake, resulting in energy imbalance and potential fat accumulation (48;94;96).

It is important to acknowledge that other studies have not found an association between parental restriction and child weight (59;97-99). The cross-sectional nature of most investigations may be a factor contributing to the inconsistency of results across studies. In a cross-sectional study, it is difficult to establish sequential causality between restriction and adiposity and to determine whether the restriction occurred as a consequence of parents perceiving their children to be at risk for overweight, or whether, in fact, the overweight resulted as a consequence of the parental restriction. Prospective work by Rifas-Shiman and colleagues demonstrate that maternal restriction of food in children at one year of age was positively associated with child BMI at age three before, but not after, adjusting for weight-for-length at age one (100). In our study, however, the relationship between parental restriction and child adiposity continued to be significant even after controlling for both the child’s actual weight and the parent’s perceived risk of the child becoming overweight (data not shown). The understanding of the role of parental restriction of food intake in adiposity merits further investigation. Understanding
the true effects of parental restriction on child adiposity will require large, prospective studies that use observations of parental feeding practices (rather than relying on parental report).

An additional contribution to the scientific literature from our study is the exploration of parental feeding practices by race/ethnicity, where Hispanic-American parents reported the highest levels of parental restriction and pressure to eat and European-American parents reported the lowest levels. Research has shown that parental perception and participation in child feeding behaviors differ across populations (73). This may suggest that feeding and food dynamics may be different among Hispanic-American, African-American, and European-American parents, which may also impact parental feeding practices (as demonstrated by our data). Our study incorporated a measure of genetic admixture in our statistical models, accounting for the biological factors underlying racial/ethnic classification (101). Despite the considerations of our study to avoid confounding factors underlying race/ethnic categorization, further research is needed to further understand the extent to which these feeding practices differentially impact adiposity in minority populations.

The evaluation of dietary practices on adiposity between children who were home-schooled (HSC) to those who were traditionally educated in a school environment (TSC) represented an innovative approach to explore feeding behaviors in children outside of their home environment. Our data suggest that the lunchtime meals children are consuming at school are more energy dense and have lower diet quality than those consumed at home, and that exposure to the school environment contributes to increased levels of adiposity in children.
Traditionally, physical activity and caloric consumption have been the factors evaluated as contributors to adiposity measures. Although physical activity did not differ between the two school groups, it contributed to increased adiposity in the entire sample. Energy intake was not significantly associated with adiposity variables; however, our data show that TSC had increased consumption of trans fat and sugars and decreased consumption of fiber, fruits, and vegetables. Statistical models evaluating the effect of diet quality in adiposity measures demonstrated an association between both saturated fat intakes and increased consumption of total sugar with adiposity variables. These data collectively suggest that differences in diet quality, rather than total energy intake, may explain why being traditionally schooled is associated with increased adiposity. The possibility that levels of adiposity in children may be influenced by diet quality is an aspect in nutrition research worth exploring due to its significant potential applications to the prevention of pediatric obesity.

The overall picture of how traditional school environments relate to increased adiposity is far from complete and is limited in our study by the small sample size and the potential limitations of utilizing a homogeneous EA sample in outcomes known to differ among racial/ethnic groups. We did not control for admixture in this study, but perhaps the HS children had increased African admixture, which was shown to be protective against excess adiposity in aim 1. It is also possible that TSC were exposed to more parental restriction at home, also increasing their risk for excess adiposity. However, perceived parental restriction did not differ between the two groups (data not shown), making it unlikely. Parents of TSC also had less actual control over what the children ate while they were at school. Whereas, in the case of the HSC children, the parents were at
home with them during the school day and could, presumably, have more control over their dietary intakes. We were not able to account for this possibility, and it is a limitation of our study.

In summary, educational environments have the potential to impact nutritional intakes and physical activity levels. Our study suggests lunchtime meals that children are consuming in a traditional school setting are a primary source of increased energy intake and are associated with poorer diet quality in children. Additionally, being traditionally schooled was associated with increased adiposity, and excess adiposity may increase the risk of cardiometabolic disease and other chronic diseases in children (102-104) and adults (3-5). We realize steps have been made to improve and regulate the school food environment in recent years (105;106). The National School Lunch act was amended in 2008, calling for more rigorous control of the diet quality of school foods. It is a limitation of our study that data collection was stopped in 2008, because it may not be representative of the food available currently in the Birmingham school systems. However, the National School Lunch act is reflected only in the National School Lunch program meals, which are regulated by the USDA. Many schools still allow vending machines, a la carte items, and competitive foods in the school food environment. Until those foods are also accounted for and monitored by either the USDA or by the schools themselves, those foods may continue to contribute to the reported low diet quality of the school food environment reported in Aim 2.

Given that low-income populations continue to be disproportionately affected by overweight and obesity (1;34;107), our studies evaluated the role of SES on adiposity by accounting for SES as a covariate in statistical models. In Aim 1, we observed a
significant inverse relationship between SES and the child adiposity latent variable. In contrast, Aims 2 and 3 demonstrated SES was not associated with adiposity. This observed discrepancy may be due to differences in both the methodology used and the demographics of the samples. There has been some debate regarding the influence of SES on adiposity in the United States. In 2007, Wang and colleagues suggested that the effects of SES on obesity were weakening in the United States (108). This observation is in concordance with the results of Aim 2 and 3 and supports previous work conducted in our group documenting that outcomes reflecting the social environment are not associated with body composition measures and do not appear to be salient factors at this early stage of the life course (109). However, the significant relationship between SES and adiposity in Aim 1 is consistent with recent longitudinal reports indicating SES continues to be associated with obesity prevalence in children and adolescents. The relationship between SES and adiposity in Aim 1 may be due, in part, to the higher levels of parental restriction reported by parents in low SES groups, which was positively associated with adiposity. SES has also been shown to impact energy balance via dietary patterns (33;110;111) and/or physical activity in children (33;38). However, a conclusive understanding of the relationship between SES to trends in pediatric adiposity in diverse populations remains unclear, and further investigation is warranted.
SUMMARY

This study provided further insight into understanding the factors associated with adiposity and body composition differences across populations. Figure 1 illustrates the study variables associated with body composition parameters in this study. The results of this dissertation indicate that genetic ancestral background, specifically African genetic admixture, can be protective against the development of excess fat accumulation and appears to explain the racial/ethnic differences in body composition. However, as George Bray succinctly described the etiology of obesity (112): “Genes load the gun, the environment pulls the trigger.” A variety of environmental factors in our study were also associated with increased adiposity, including: parental restriction, diet quality, SES, and being traditionally schooled. Parental restriction was associated with increased adiposity, raising the need for further research investigating the extent to which parental feeding practices impact adiposity and how such impact may differ among members of diverse populations.

The explorations on the role of energy intake in adiposity demonstrated no associations, even though energy intake could differ among particular groups (HSC and TSC). Our data suggest that diet quality may be a better outcome when evaluating the role of energy intake variables in fat acquisition and accumulation. Previous work has demonstrated that diet quality is independently associated with children’s weight status (113). When integrating the results of our studies and identifying future directions, it is evident that prospective studies are needed to investigate the relationship among parental
feeding practices, diet quality, and child adiposity in a multi-ethnic sample that reflects the diversity of the U.S. population.

Factors that were associated with decreased adiposity levels were increased physical activity and parental pressure to eat. Though parental pressure to eat was associated with less fat mass, the dislike for foods and the potential reduction in nutrient intake that accompanies this behavior can be detrimental to overall health in children (58;114). Educational initiatives should be developed to prevent parents from engaging in this behavior as a potential means to decrease adiposity in children. Given that interventions to increase physical activity have shown to be successful in preventing excessive weight gain in youth and conserving overall health (115), and that physical activity was inversely associated with adiposity, interventions incorporating physical activity could be beneficial in decreasing a child’s risk for excess adiposity (116).

There were many limitations of this dissertation. One issue that was outlined in our work is that there are clear differences in energy intakes, diet quality, and adiposity in TSC and HSC. Given that over one third of our EA sample is comprised of HSC, it raises the question of whether we should control for school status when evaluating dietary or body composition variables in this sample. It also highlights that results found with the cohort of EA children that includes home-schooled EA children may not be generalizeable to all EA children. Additionally, given that alterations in energy expenditure can predict weight changes (117;118), one limitation of this dissertation is that we didn’t account for resting energy expenditure in any of our studies, the largest contributor to total energy expenditure. This study also did not account for gene and
environment interactions, and future work will investigate these interactions in our cohort.

Studies focusing on the evaluation of both genetic and environmental factors have provided great insight into the etiology of obesity. It has been proposed that the heritability of obesity-related traits increases with age (119), suggesting that at an early stage of the lifespan, adiposity is highly influenced by environmental factors, probably highly influenced by the home and school environment, and highlighting a potentially crucial time for interventions. Children cannot change their genetic ancestral makeup or their genes, but modifications in their environment can be made. This dissertation has identified some modifiable environmental factors that are associated with increased adiposity, potentially contributing to the increased prevalence of obesity. Large-scale, multi-ethnic prospective studies using robust measures of adiposity, however, are needed to investigate the true effects of parental feeding practices and the school food environment.
Figure 1. Study variables associated with the development of childhood obesity.


APPENDIX A

INSTITUTIONAL REVIEW BOARD APPROVAL
September 13, 2012

MEMORANDUM

FROM: Cari Oliver, CIP
      Assistant Director, OIRB

RE: Michelle I Cardel’s involvement in Protocol Number X040109007 “Admixture Mapping for Insulin Complex Outcomes (AMERICO STUDY)”

To Whom It May Concern:

Michelle Cardel is listed as an investigator in the above-mentioned study conducted by Dr. Jose R. Fernandez’ study. Her dissertation is entitled “Childhood Obesity: The Role of Genetic Admixture, Parental Feeding practices, and the School Food Environment”.
Protection of Human Subjects
Assurance Identification/IRB Certification/Declaration of Exemption
(Common Rule)

Policy: Research activities involving human subjects may not be conducted or supported by the Departments and Agencies adopting the Common Rule (55FR2003, June 18, 1991) unless the activities are exempt from or approved in accordance with the Common Rule. See section 101(b) of the Common Rule for exemptions. Institutions submitting applications or proposals for support must submit certification of appropriate institutional review board (IRB) review and approval to the Department or Agency in accordance with the Common Rule.

1. Request Type
   [ ] ORIGINAL
   [ ] CONTINUATION
   [x] GRANT
   [ ] CONTRACT
   [ ] FELLOWSHIP
   [ ] COOPERATIVE AGREEMENT
   [ ] EXEMPTION
   [ ] OTHER:

2. Type of Mechanism:
   [ ] Federal
   [ ] State
   [ ] Local
   [ ] Other:

3. Name of Federal Department or Agency and, if known, Application or Proposal Identification No.:

4. Title of Application or Activity:
   Admixture Mapping for Insulin Complex Outcomes (AMERICO STUDY)

5. Name of Principal Investigator, Program Director, Fellow, or Other:
   FERNANDEZ, JOSE R

6. Assurance Status of this Project (Respond to one of the following):
   [x] This Assurance, on file with Department of Health and Human Services, covers this activity:
   Assurance Identification No. FHA000005960, the expiration date, 01/24/2017, IRB Registration No. IRB000000729

   [ ] This Assurance, on file with (agency/dept.),
   Assurance No.________________, the expiration date__________, IRB Registrar/Identification No.__________________ (if applicable)

   [ ] No assurance has been filed for this institution. This institution declares that it will provide an Assurance and Certification of IRB review and approval upon request.

   [ ] Exemption Status: Human subjects are involved, but this activity qualifies for exemption under Section 101(b), paragraph:__________

7. Certification of IRB Review (Respond to one of the following if you have an Assurance on file):
   [ ] Full IRB Review on (date of IRB meeting) ____________
   [ ] Expedited Review on (date) ____________
   [ ] If less than one year approval, provide expiration date ______

   [ ] This activity contains multiple projects, some of which have not been reviewed. The IRB has granted approval on condition that all projects covered by the Common Rule will be reviewed and approved before they are initiated and that appropriate further certification will be submitted.

8. Comments
   Protocol subject to Annual continuing review.
   Admixture Mapping for Insulin Complex Outcomes (AMERICO STUDY)

9. IRB Approval Issued:
   U-13-12

10. Name and Address of Institution
   University of Alabama at Birmingham
   701 20th Street South
   Birmingham, AL 35284

11. Phone No. (with area code) (205) 934-3789
12. Fax No. (with area code) (205) 934-1301
13. Email: irb@uab.edu

14. Name of Official
   Leslie Cooper, CRP

15. Title
   Assistant Director, IRB

16. Signature
   Leslie Cooper, CRP

17. Date
   U-13-12

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