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A trial of reduced carbohydrate diet to improve metabolic outcomes and decrease adiposity in obese peripubertal African American girls: does macronutrient profile matter?

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Abstract

Objectives—Obesity prevalence among African American (AA) girls is higher than that of other groups. As typical calorie-restriction obesity treatment strategies have had limited success, alterations in macronutrient composition might effectively improve metabolic outcomes in this population and impact future body composition trajectories. The objective was to evaluate the efficacy of a moderately restricted carbohydrate (CHO) versus a standard CHO diet on weight/fat loss and metabolic parameters in overweight/obese AA girls aged 9–14 years.

Methods—A total of 26 AA girls (ranging from 92nd BMI percentile and above) were assigned to either a reduced- (SPEC: 42% calories from CHO, n=12) or a standard- (STAN: 55% of calories from CHO, n=14) CHO diet (protein held constant) for 16-weeks. All meals were provided and clinically tailored to meet the estimated energy requirements ($REE \times 1.2$ in eucaloric phase and $REE \times 1.2 - 1000\text{kcal}$ in energy deficit phase). The first five-weeks encompassed a eucaloric phase evaluating metabolic changes in the absence of weight change. The subsequent 11-weeks were hypocaloric (1000kcal/d deficit) in effort to promote weight/fat loss. Meal tests were performed during the eucaloric phase for metabolic analyses. Dual-energy x-ray absorptiometry (DXA) was used to evaluate body composition.

Results—Both groups had reductions in weight/adiposity but the difference did not reach significance. The solid meal test indicated improved glucose/insulin homeostasis on the SPEC diet up to three hours post-ingestion. In addition, significantly lower triglycerides ($p < 0.001$) were observed on the SPEC diet.

Conclusions—Dietary CHO reduction favorably influences metabolic parameters but did not result in greater weight/fat loss relative to a standard diet in obese AA girls. Future research is needed to determine long-term effectiveness of a reduced CHO diet on glucose and insulin homeostasis and how it may apply to weight maintenance/fat loss during development alone and/or in combination with additional weight loss/metabolic improvement strategies.

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Conflicts of interest: None

Keywords

Puberty; diet; obesity; carbohydrate metabolism; race

INTRODUCTION

In addition to the increased probability of developing obesity in adulthood, excessive body fat accrual during developmental years is linked with earlier onset of chronic diseases (12) and, due to greater exposure, more adverse metabolic health outcomes (13;22;35). This trend is particularly salient in African American (AA) girls, who are disproportionately burdened by obesity as well as risk factors for the future progression to type 2 diabetes and cardiovascular diseases when compared to other racial/ethnic groups (34). Identification of strategies to limit excess adipose tissue acquisition and prevent metabolic perturbations early in the life course, particularly among AA girls is essential.

Adipose tissue deposition/accumulation is influenced by multiple metabolic pathways in which nutrition plays a central role (11;13;15). Thus, dietary modification strategies are commonly undertaken to control weight and fat accumulation in both overweight/obese adults and children (6–8;11;15). Carbohydrate (CHO), the main dietary component affecting insulin action, impacts the postprandial metabolic response contributing to weight and body composition changes. For example, in comparison to a low CHO meal, the response to a meal high in CHO results in an augmented metabolic cascade of events (increased serum glucose and insulin, and subsequent increased lipogenesis and glycogenesis) (24). The relatively more rapid absorption of glucose associated with this type of meal challenges homeostatic mechanisms, complicating the transition from the postprandial to the postabsorptive state. For example, hypoglycemia is more likely to occur following consumption of a high CHO meal, triggering counter-regulatory hormone pathways, thereby influencing satiety cues (43). In addition, the proportion of CHO influences fuel oxidation and energy expenditure, reflecting a shift from fat to glucose oxidation and concomitant down-regulation of energy expended at rest (41;43). Thus, high CHO intake can perturb metabolic pathways involved in fuel metabolism and may be permissive of unfavorable body tissue compartmentalization as well as conducive to impaired insulin homeostasis (27).

The response to a high CHO diet may be further amplified in an individual with inherently high insulin secretion (e.g. AA girls), particularly during sensitive times of growth and development (e.g. puberty), further increasing the potential for excess fat deposition (25;26). We have previously reported higher insulin secretion in AA girls as well as greater adipose tissue deposition (5). Accordingly, a dietary approach which minimizes the insulin-related altered/exaggerated cascade of events following meal ingestion, implemented during a critical period for adipose tissue deposition may be beneficial in improving metabolic outcomes and facilitating weight loss. This study was conducted, in obese peripubertal AA girls, to evaluate the efficacy a moderately-restricted versus a standard CHO diet on weight/fat loss and biochemical markers of insulin homeostasis and metabolic parameters.

METHODS

Participants were 26 overweight/obese (ranging from 92nd BMI percentile and above) AA girls ages 9 to 14 years. Obesity was defined as >95th age- specific BMI percentile, with all but one girl (92nd percentile) falling in that range. The girls were pubertal stage II–V as assessed by a pediatrician according to the criteria of Marshall and Tanner (31;32). Exclusion criteria were medical diagnoses and/or taking medications known to affect body composition, metabolism, cardiac function, etc. Girls were recruited to participate in a

weight-loss study through newspaper advertisement, distribution of flyers (posted at various community partnerships) and by word-of-mouth. The nature, purpose, and possible risks of the study were carefully explained to the girls and parent(s), and informed assent and consent, respectively, were obtained. Full datasets were available on 26 of 36 girls recruited. Reasons for non-completion included difficulty complying with diet (6), lack of transportation preventing food-pick-up (2), relocation, and inability to perform blood draw (2). The protocol was approved by the Institutional Review Board for human subjects at the University of Alabama at Birmingham (UAB). All measurements were performed at the Participant and Clinical Interactions Resources (PCIR) and the Department of Nutrition Sciences at UAB between 2008 and 2009.

Protocol

Participants were block-randomized to one of two diets which they remained on for the duration of the study: reduced CHO (42% of calories from CHO; SPEC) or standard CHO (55% of calories from CHO; STAN). The 16-week intervention included two phases: a five-week eucaloric (weight-stable) phase and an 11-week hypocaloric (weight-loss, approximate 1000-calorie reduction) phase. All food was provided for both phases of the study, with overall calorie amount determined according to resting energy requirements (REE; assessed via indirect calorimetry) multiplied by an activity factor of 1.2 (averaging 2010 kcal/d for eucaloric and 1025 kcal/d for hypocaloric). A physical activity questionnaire (paq-a(10)) was used in effort to determine an appropriate activity factor. Based on reported low-level of physical activity, an activity factor of 1.2 was used for all participants during each phase (19). At baseline, participants attended two visits. During the first visit, a physical examination by the study pediatrician was performed, questionnaires on typical diet and regular physical activity were given and a whole-body dual energy x-ray absorptiometry (DXA) scan was performed. During the second visit, participants reported to the PCIR at 06h30 in the fasted state for metabolic testing. After 30 minutes of rest, participants underwent indirect calorimetry and a liquid mixed meal test (LMMT). Following the LMT, participants were provided meals according to prescribed diet and began the five-week eucaloric phase. Throughout this phase, calorie prescription was evaluated as participants were weighed twice per week to ensure weight stability. Weight changes exceeding two kg from baseline resulted in caloric modification in effort to maintain weight. At mid-point of the eucaloric phase, a solid meal test using the assigned breakfast was given to determine the metabolic response to a meal on the prescribed diet. At the end of the five-week eucaloric phase, participants repeated the DXA scan, indirect calorimetry, and LMMT. The liquid meal was administered to evaluate pancreatic response and changes over time to dietary modification whereas the solid meal was to evaluate the acute response to the actual meal. The LMMT and solid meal test were conducted only during the eucaloric phase. Given that high CHO intakes can alter metabolic pathways leading to impaired insulin and glucose profiles (26;28) it was important to evaluate the postprandial effects of the meal tests in the absence of weight loss to truly disentangle the effects of diet versus energy imbalance on body composition. During the 11-week hypocaloric phase, participants remained on prescribed diet but received 1000 kcal/day deficit. Participants were weighed twice per week to ensure compliance to the diet. Protocol design was such that a weight change reflecting greater than two kg would result in calorie modification, though this action was not necessary for any participant. Upon completion of the 11-week hypocaloric phase, an additional DXA scan was performed. Girls were encouraged to maintain their current level of physical activity for the duration of the study.

Assessment of Typical Diet and Physical Activity Patterns

A four-day food record was used to assess typical dietary patterns, particularly breakfast consumption and meal patterning. The paq-a was used to obtain a measure of regular daily physical activity.

Diets

Table 1 outlines the nutritional composition of the SPEC diet and the STAN diet. The SPEC comprised 42% of energy from CHO and 40% of energy from fat whereas the STAN comprised 55% of energy from CHO and 27% of energy from fat. Both diets contained ~18% of energy from protein. However, both comprised <10% saturated fat. A moderate reduction was chosen in order to include an acceptable diet to the study population, while maintaining a fat content <40% of the calories. Based on preliminary analyses as well as recent comparisons to NHANES data (36), the reduction in CHO of the SPEC diet represented an approximate 25% decrement in amount of CHO consumed by this population. Although diets were designed to be similar in CHO (e.g. fiber, fructose content) and fat quality (e.g. cholesterol), in an effort to attain macronutrient profile while providing cultural- and age- appropriate meals, there were differences in fructose, fiber and cholesterol ($p<0.01$).

Participants were instructed to remain on assigned diet throughout the duration of the 16-week intervention period: SPEC, specialized for the unique metabolic characteristics (high insulin secretion/low insulin sensitivity) of the sample population, or STAN, reflecting typical dietary composition among the same population. All meals were based on an energy level of 1600 kcal, and snacks in increments of 100 kcal were added to the menu as necessary for participants who required excess. All meals included culturally- and age-appropriate foods (no supplements or formulas) and were prepared and packaged in the research kitchen of the UAB PCIR. Non-caloric fluid intake of ≥ 64 oz/d was recommended.

Anthropometric Assessment

The same registered dietitian obtained anthropometric measurements on all participants. Participants were weighed (Scale-tronix 6702W; Scale-tronix, Carol Stream, IL) to the nearest 0.1 kg in minimal clothing without shoes. Height was also recorded without shoes using a digital stadiometer (Heightronic 235; Measurement Concepts, Snoqualmie, WA). BMI percentile was calculated using CDC growth charts based on these measurements (<http://apps.nccd.cdc.gov/dnpabmi/>).

Pubertal Status

The Tanner staging method has been demonstrated as a reliable indicator of pubertal development. Direct observation for the assessment of pubertal stage by a pediatrician, the 'gold standard' for differentiating among the five stages of maturity (9;18), was utilized. The staging based on the criteria of Marshall and Tanner (32;33) is according to both breast and pubic hair development. One composite number is assigned for Tanner staging, representing the higher of the two values defined by breast and pubic hair development (30).

Liquid Meal Test/Insulin Secretion

To determine insulin response to a standardized meal, insulin sensitivity and secretion was derived via frequent blood sampling following ingestion of a liquid mixed meal test (Carnation Instant Breakfast prepared with whole milk). To perform the test, a flexible intravenous catheter was placed in the antecubital space of the left arm. The dose for the liquid meal test was obtained according to amount of lean body mass (LBM) of the participant (1.75g carbohydrate/kg LBM) and provided a macronutrient distribution (24%

fat, 58% CHO, 18% protein) comparable to the STAN diet. Subjects were required to consume the meal within five minutes. Blood was drawn at baseline (three samples over 15 min) prior to the initiation of meal consumption at time zero. Subsequent blood samples were drawn every five minutes from time zero to 30 minutes, every 10 minutes from time 40 to 180 minutes, and at 210 and 240 minutes. Using the obtained measures, the insulin sensitivity index (SI) was calculated by oral glucose model (2).

Solid Meal Test

To examine the response of the subjects to an assigned diet meal, during week three of the eucaloric phase, the assigned diet breakfast was consumed at the PCIR. The breakfast meal was that which was assigned the day of the test and therefore was not exactly the same across participants. However, it was representative of the nutrient distribution of the assigned diet and provided a similar amount of calories (~500). Metabolites were analyzed before, during and after intake. To perform the test, a flexible intravenous catheter was placed in the antecubital space of the left arm. At time zero, subjects began meal consumption, completing within 20 minutes. Blood samples were collected at baseline, and at times 15, 60, 90, 120, 180, and 240 minutes. Samples were analyzed to determine if, as predicted, SPEC would result in a more stable blood glucose:insulin profile (60-minute insulin) than STAN.

Body composition and fat distribution

Whole body composition was measured by 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. Due to size limitations, girls not fitting within the scanning box were right-sided hemi-scanned with the left side estimated as per instrument protocol.

Resting energy expenditure

REE was determined in the fasted state upon arrival in the morning of the outpatient visit (Deltatrac, Sensormedics) in the PCIR. The instrument was calibrated before each test against standard gases. Before beginning testing, girls rested in attempt to return to “basal” state. During testing, all subjects were instructed to lie still while remaining awake. A canopy hood was used to collect expired air for 20 minutes after a 10-minute equilibration period, and oxygen consumption and carbon dioxide production were measured continuously during this time. After completion, REE was calculated using the equation of Weir (44).

Assay of Metabolites

Glucose was measured in 12 µl sera with the glucose oxidase method using a SIRRUS analyzer (interassay CV 2.56%). Insulin was analyzed using a TOSOH 1800 Automated Immunoassay Analyzer. Assay sensitivity is 15.42 pmol/L, mean intra-assay CV is 4.69%, and interassay CV is 6.0%. Triglycerides (TG) were assessed with the glycerylphosphate method. HDL-cholesterol (HDL) was analyzed using a two-reagent system involving stabilization of LDL, VLDL, and chylomicrons using cyclodextrin and dextrin sulfate, and subsequent enzymatic-colorimetric detection of HDL. Adiponectin and leptin were measured by RIA as described (3;17)

Statistical Analysis

Based on the data from a similar population, Linear Mixed Models (LMMs) using SAS PROC MIXED were fit in order to estimate the difference over the four-month time period while accounting for dependency and missing data. Age and BMI percentile at start of the

program were used as covariates. The standard errors for the difference were multiplied by the square root of their degrees-of-freedom to estimate the standard deviation of difference scores. For subsequent power analyses, this standard deviation of difference scores was assumed to remain constant over the $k = 3$ repeated measures of the proposed study. These results indicated that we would need at least $N = 22$ total subjects to have 80% statistical power at $\alpha = 0.01$. Statistical significance was determined as an $\alpha \leq 0.05$.

Differences in descriptive statistics were examined using t-tests. The contributions of diet to metabolic parameters, body composition and weight outcomes were evaluated using ANOVA to allow for inclusion of covariates. All models were adjusted for pubertal stage and height and for models evaluating HDL, triglyceride concentration was added as a covariate.

To conform to the assumptions of linear regression, all statistical models were evaluated for residual normality and logarithmic transformations were performed when appropriate. All data were analyzed using SAS 9.1 software.

RESULTS

Table 2 illustrates baseline sample characteristics, metabolites and body composition measurements of participants in the total sample and by diet group. Participants were similar across all variables except fat percent, which was statistically higher for leg fat and total percent fat ($p < 0.05$ for both) but no statistical difference in trunk fat ($p > 0.14$), among girls on STAN.

Figure 1 illustrates the insulin (a) and glucose (b) response to the test meal given during week three. Girls on SPEC had a lower insulin ($p < 0.05$) concentration at 60 minutes following ingestion of the meal. The difference in insulin response remained significant for up to three hours after the meal challenge ($p < 0.01$, for all time points). Girls on the STAN diet had higher glucose levels at times 60, 90, and 120 minutes ($p < 0.01$).

Table 3 illustrates metabolic parameters assessed at baseline and after five weeks on the eucaloric diets. Changes in metabolic parameters over the five-week eucaloric phase by diet group, within diet group (time), and diet*time group interactions presented. After five weeks a time effect was observed, girls on SPEC had higher fasting insulin (6.7 mg/dl increase; $p = 0.02$) relative to their baseline values, but the insulin concentration did not differ by diet group ($p = 0.9$). The elevated TG (12.9mg/dl increase) on the STAN diet and concomitant decrease on SPEC (14.9mg/dl) was such that by the end of five weeks the girls on the SPEC diet had significantly lower TG relative to the girls on the STAN diet ($p < 0.001$). Girls on both diets decreased LDL (STAN=9.7; SPEC=4.8) and increased adiponectin levels (0.8=STAN; 0.04=SPEC) over five weeks, although the changes did not reach statistical significance ($p = 0.07, 0.06$, respectively). Because there were differences in fat and carbohydrate quality between the diets, the glycemic index and load, fat, fiber and cholesterol were included as independent variables to assess their contribution. However, significant contribution ($p < 0.10$) was not observed for any of these variables (data not shown).

Diet groups did not differ in amount of weight lost at the end of the intervention, although there was a trend towards greater weight loss on the SPEC diet relative to the STAN diet during the eucaloric phase ($p = 0.09$). This marginal difference in weight loss appeared to have been attributed to loss from the LBM compartment ($p = 0.11$; data not shown) as opposed to fat compartments. Girls on SPEC had less leg fat ($p = 0.04$) and marginally less percent fat ($p = 0.06$) at the end of the eucaloric phase relative to girls on STAN. A significant difference in fat compartments between the two diets was not detected at the end

of the hypocaloric phase or at the end of the intervention, suggesting the plausibility of greater leg and percent fat loss on the STAN diet, though not detected as a significant difference. It is unclear if the greater loss is attributable to greater amount present prior to the intervention. Table 4 illustrates the *change* over time in weight and fat parameters from baseline. Although diet groups also did not differ in total or compartmental fat loss over the intervention, there were significant changes in weight and fat loss on both diets.

DISCUSSION

High CHO diets have been associated with increased fat deposition (16;23;26). During the pubertal transition, the additional ‘insult’ of transient insulin resistance may augment fat mass accrual, especially among AA girls, who in general are more insulin resistant and more apt to gain fat during this period (5;21). The physiological premise of this study was that a dietary approach minimizing associated perturbations may improve long-term health outcomes. We hypothesized that a moderately-reduced CHO diet would improve body composition outcomes through attenuation of the insulin response, thereby increasing the capacity to control weight/fat accumulation in this population. There were no group differences in weight loss or body composition changes at the end of the intervention. A differential response in terms of insulin/glucose homeostasis was observed between diet groups, as those in the reduced CHO group experienced lower insulin response at 60 through 120 minutes and better glucose maintenance during a meal challenge. Individuals receiving the reduced CHO diet decreased TG concentrations to a greater degree than those who received a standard CHO diet. Our results suggest dietary CHO reduction favorably influences lipid parameters but did not result in greater weight or fat loss relative to a standard diet in peripubertal AA girls.

Low fat, high CHO diets have been consistently shown to induce hypertriglyceridemia (37;39;40). Moderate CHO reduction resulted in decreased TG, whereas the standard CHO diet (55% CHO; 27% fat), admittedly not a classic ‘high CHO diet’ (e.g. >75% CHO), resulted in marginally increased TG. Our findings suggest that even a modest reduction in CHO relative to “typical” intake has the capacity to reduce serum TG. Interestingly, both diets demonstrated a decrease in LDL and increase in adiponectin (albeit not significant), suggestive of improvement of at least some aspects of metabolism. Both diets were designed to be of higher quality in terms of macronutrient profile than what has been previously reported to be practiced by a similar pediatric cohort in the same geographic region (4;20). We speculate that metabolic improvements in both groups are at least partially attributed to this as well as the implementation of a more regular and healthier eating pattern than previously practiced (e.g., breakfast consumption and smaller, more frequent meal consumption). However, the contribution of macronutrient quality cannot be ignored. Indeed, carbohydrate quality (e.g. fiber, fructose) and fat quality (e.g. cholesterol) affect *de novo* fat synthesis. The beneficial effect of the SPEC diet on lipid profile (albeit with higher dietary cholesterol) supports the adverse effect of fructose and/or beneficial effect of fiber on endogenous cholesterol synthesis (37;39;40). Future studies on the extent to which the varying dietary quality exerts effects on metabolic parameters, particularly during formative years are warranted.

A significant dietary effect was not observed in weight-loss or body composition. However, it is possible that there were baseline differences in body composition that may have introduced confounding. For example, the difference in trunk fat, though not statistically different between the groups could have mechanistically influenced the metabolic ability to utilize substrates as well as mediate differences in metabolic and reproductive hormones (1). Although visceral adiposity was not assessed, waist circumference was not statistically different ($p=0.30$) between the STAN (102.4cm) and SPEC (96.2cm) groups. We must be

aware that a statistically significant difference is not the same as a difference of biological significance; i.e., lack of a statistically significant group difference in fat depots may not be transferred to indifference in metabolic risk. Though even in light of the current pediatric obesity epidemic literature is scant, Randle et al (38) demonstrated utilization of one nutrient inhibited the use of another directly and without hormonal mediation several decades ago. This remains an auspicious avenue for investigation.

Although composition of the diet is an important factor in the determination of glucose and fat oxidation, individuals prone to obesity have an overall lower fat oxidation rate as compared to non-obese individuals (1;14). Contributing to disturbances related to obesity is the degree of metabolic inflexibility likely existent in this population due to the demands of puberty (6). For example, in overweight 10-year old girls in the throes of puberty, high-fat feeding induced lower thermogenesis and higher fat deposition than isocaloric low-fat feeding (29). The higher fat accompanying the reduced CHO diet may have perturbed metabolic parameters related to fat oxidation which may supersede other beneficial effects related to fat loss. It is also possible that excess weight gain results in a degree of irreversible exacerbation of metabolic changes during the pubertal transition. Sunehag et al recently reported no effect of modifications in CHO intake on glucose and lipid metabolism in healthy *nonobese* prepubertal children (42), suggesting alteration in fat oxidation may be due to an already perturbed metabolic environment. Further, increased fasting insulin experienced by those in the reduced CHO group may have diverted fatty acids towards storage vs. oxidation as well as altering fatty acid utilization by the muscle. Reducing CHO intake can rapidly decrease the body's glycogen content, which may be responsible for elicitation of compensatory response (change in tissue partitioning/fuel utilization/water storage) due to the body's inability to store excess CHO. This may explain the observed reduction in LBM during the eucaloric phase. Observed LBM loss may have also been reflective of fluid shift in response to altered fuel utilization (i.e. decreased glycogen storage, resulting in decreased concomitant water storage). Interestingly, during the hypocaloric phase, we did not observe additional LBM reduction in individuals on SPEC, and LBM did not differ between groups by the end of the intervention. However, it is important to recognize that at baseline, STAN girls had significantly more total fat relative to SPEC girls, raising the possibility that lipid metabolism of STAN and SPEC girls may differ in utilization of glucose and fat oxidation and differences observed may be related to baseline fat mass. There is paucity in the literature centered on metabolic flexibility in the pediatric population. Indeed, obesity is associated with increased fatty acid concentration and poor suppression of lipolysis with increased insulin levels (1;5;28). Intriguingly, existing studies focus on glucose availability with little attention paid to lipid or protein availability, irrespective of the inefficient switch to these substrates in the insulin resistant state (1). Further, a differential impact among AA girls has not been evaluated. Though such hypotheses are in need of further confirmation, our findings indeed highlight the importance of longer-term tailored, rather than one-size-fits-all dietary strategies for weight management.

A major strength of this study was the robust measurement used to assess metabolism and physiology. Providing all food to participants provided additional strength to the study design of this intervention. Although we cannot ensure 100% compliance, bi-weekly weigh-ins helped increase adherence to the diet. Nevertheless, as a pilot study, we were limited by small sample size and inclusion of only one racial/ethnic group. Identification of metabolic changes following weight loss would have also been beneficial, as it would have been ideal to have no differences in fat mass between the two diet groups. However, when groups are randomized, there is also a risk that the groups may differ at baseline. Whereas substantial evidence exists reporting the effect of fat and carbohydrate quality on lipid profile, the variance and modest sample size may have precluded our observance of a significant

contribution. Future studies should include metabolic measures following the hypocaloric phase, a larger sample size, inclusion of multiple races/ethnicities, as well as inclusion of individuals with a wider range of body habitus. In addition, beyond gross macronutrient effects, the evaluation of quality of macronutrients represents an auspicious area of evaluation.

Conclusions

In summary, we observed comparable weight and fat loss in both diet groups and a significantly greater reduction in TG following the SPEC diet compared with the STAN diet over the 16-week intervention. The fact that the weight and fat loss for both groups were significantly reduced after 16-weeks, argues that caloric intake independent of macronutrient consumption can be beneficial for controlling excess weight and fat accumulation in a population at risk for adverse health consequences. Future research is needed to determine long-term effectiveness of CHO reduction, (with an added emphasis on CHO quality) on weight/fat-loss on obesity-related metabolic parameters.

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Reference List

1. Aucouturier J, Duche P, Timmons BW. Metabolic flexibility and obesity in children and youth. *Obes Rev.* 2011; 12:e44–e53. [PubMed: 20977601]
2. Breda E, Cavaghan MK, Toffolo G, et al. Oral glucose tolerance test minimal model indexes of beta-cell function and insulin sensitivity. *Diabetes.* 2001; 50:150–158. [PubMed: 11147781]
3. Bush NC, Darnell BE, Oster RA, et al. Adiponectin is lower among African Americans and is independently related to insulin sensitivity in children and adolescents. *Diabetes.* 2005; 54:2772–2778. [PubMed: 16123368]
4. Casazza K, Dulin-Keita A, Gower BA, et al. Relationships between reported macronutrient intake and insulin dynamics in a multi-ethnic cohort of early pubertal children. *Int J Pediatr Obes.* 2009;1–8.
5. Casazza K, Goran MI, Gower BA. Associations among insulin, estrogen, and fat mass gain over the pubertal transition in African-American and European-American girls. *J Clin Endocrinol Metab.* 2008; 93:2610–2615. [PubMed: 18349063]
6. Casazza K, Thomas O. Do Dietary Modifications Made Prior to Pubertal Maturation Have the Potential to Decrease Obesity Later in Life? *ICAN.* 2009 In Press.
7. Chakravarthy MV, Booth FW. Eating, exercise, and "thrifty" genotypes: connecting the dots toward an evolutionary understanding of modern chronic diseases. *J Appl Physiol.* 2004; 96:3–10. [PubMed: 14660491]
8. Chaput JP, Tremblay A, Rimm EB, et al. A novel interaction between dietary composition and insulin secretion: effects on weight gain in the Quebec Family Study. *Am J Clin Nutr.* 2008; 87:303–309. [PubMed: 18258618]
9. Coleman L, Coleman J. The measurement of puberty: a review. *J Adolesc.* 2002; 25:535–550. [PubMed: 12234559]
10. Crocker PR, Bailey DA, Faulkner RA, et al. Measuring general levels of physical activity: preliminary evidence for the Physical Activity Questionnaire for Older Children. *Med Sci Sports Exerc.* 1997; 29:1344–1349. [PubMed: 9346166]
11. Dietz WH. Periods of risk in childhood for the development of adult obesity--what do we need to learn? *J Nutr.* 1997; 127:1884S–1886S. [PubMed: 9278575]

12. Flegal KM, Ogden CL, Yanovski JA, et al. High adiposity and high body mass index-for-age in US children and adolescents overall and by race-ethnic group. *Am J Clin Nutr.* 2010; 91:1020–1026. [PubMed: 20164313]
13. Franks PW, Hanson RL, Knowler WC, et al. Childhood obesity, other cardiovascular risk factors, and premature death. *N Engl J Med.* 2010; 362:485–493. [PubMed: 20147714]
14. Frislancho AR. Reduced rate of fat oxidation: a metabolic pathway to obesity in the developing nations. *Am J Hum Biol.* 2003; 15:522–532. [PubMed: 12820194]
15. Gillman MW, Rifas-Shiman SL, Kleinman K, et al. Developmental origins of childhood overweight: potential public health impact. *Obesity (Silver Spring).* 2008; 16:1651–1656. [PubMed: 18451768]
16. Goran MI, Gower BA. Longitudinal study on pubertal insulin resistance. *Diabetes.* 2001; 50:2444–2450. [PubMed: 11679420]
17. Gower BA, Nagy TR, Goran MI, et al. Leptin in postmenopausal women: influence of hormone therapy, insulin, and fat distribution. *J Clin Endocrinol Metab.* 2000; 85:1770–1775. [PubMed: 10843150]
18. Herman-Giddens ME, Slora EJ, Wasserman RC, et al. Secondary sexual characteristics and menses in young girls seen in office practice: a study from the Pediatric Research in Office Settings network. *Pediatrics.* 1997; 99:505–512. [PubMed: 9093289]
19. Heymsfield SB, Harp JB, Rowell PN, et al. How much may I eat? Calorie estimates based upon energy expenditure prediction equations. *Obes Rev.* 2006; 7:361–370. [PubMed: 17038130]
20. Keita AD, Casazza K, Thomas O, et al. Neighborhood-level disadvantage is associated with reduced dietary quality in children. *J Am Diet Assoc.* 2009; 109:1612–1616. [PubMed: 19699843]
21. Kimm SY, Barton BA, Obarzanek E, et al. Racial divergence in adiposity during adolescence: The NHLBI Growth and Health Study. *Pediatrics.* 2001; 107:E34. [PubMed: 11230615]
22. Lee JM, Pilli S, Gebremariam A, et al. Getting heavier, younger: trajectories of obesity over the life course. *Int J Obes (Lond).* 2010; 34:614–623. [PubMed: 19949415]
23. Loomba-Albrecht LA, Styne DM. Effect of puberty on body composition. *Curr Opin Endocrinol Diabetes Obes.* 2009; 16:10–15. [PubMed: 19115520]
24. Ludwig DS. Dietary glycemic index and the regulation of body weight. *Lipids.* 2003; 38:117–121. [PubMed: 12733742]
25. Ludwig DS. Clinical update: the low-glycaemic-index diet. *Lancet.* 2007; 369:890–892. [PubMed: 17368136]
26. Ludwig DS, Majzoub JA, Al-Zahrani A, et al. High glycemic index foods, overeating, and obesity. *Pediatrics.* 1999; 103:E26. [PubMed: 10049982]
27. Lustig RH. Childhood obesity: behavioral aberration or biochemical drive? Reinterpreting the First Law of Thermodynamics. *Nat Clin Pract Endocrinol Metab.* 2006; 2:447–458. [PubMed: 16932334]
28. Lustig RH. The 'skinny' on childhood obesity: how our western environment starves kids' brains. *Pediatr Ann.* 2006; 35:898–897. [PubMed: 17236437]
29. Maffei C, Schutz Y, Grezzani A, et al. Meal-induced thermogenesis and obesity: is a fat meal a risk factor for fat gain in children? *J Clin Endocrinol Metab.* 2001; 86:214–219. [PubMed: 11232003]
30. Malina, RM.; Bouchard, C. Growth, Maturation, and Physical Activity. Champaign: Human Kinetics Books; 1991.
31. Marshall WA, Tanner JM. Growth and physiological development during adolescence. *Annu Rev Med.* 1968; 19:283–300. [PubMed: 4297619]
32. Marshall WA, Tanner JM. Variations in pattern of pubertal changes in girls. *Arch Dis Child.* 1969; 44:291–303. [PubMed: 5785179]
33. Marshall WA, Tanner JM. Variations in the pattern of pubertal changes in boys. *Arch Dis Child.* 1970; 45:13–23. [PubMed: 5440182]
34. Ogden CL, Carroll MD, Curtin LR, et al. Prevalence of high body mass index in US children and adolescents, 2007–2008. *JAMA.* 2010; 303:242–249. [PubMed: 20071470]

35. Olshansky SJ, Passaro DJ, Hershow RC, et al. A potential decline in life expectancy in the United States in the 21st century. *N Engl J Med*. 2005; 352:1138–1145. [PubMed: 15784668]
36. Pan Y, Pratt CA. Metabolic syndrome and its association with diet and physical activity in US adolescents. *J Am Diet Assoc*. 2008; 108:276–286. [PubMed: 18237576]
37. Parks EJ, Krauss RM, Christiansen MP, et al. Effects of a low-fat, high-carbohydrate diet on VLDL-triglyceride assembly, production, and clearance. *J Clin Invest*. 1999; 104:1087–1096. [PubMed: 10525047]
38. Randle PJ. Endocrine control of metabolism. *Annu Rev Physiol*. 1963; 25:291–324. [PubMed: 13990766]
39. Roberts R, Bickerton AS, Fielding BA, et al. Reduced oxidation of dietary fat after a short term high-carbohydrate diet. *Am J Clin Nutr*. 2008; 87:824–831. [PubMed: 18400703]
40. Schwarz JM, Linfoot P, Dare D, et al. Hepatic de novo lipogenesis in normoinsulinemic and hyperinsulinemic subjects consuming high-fat, low-carbohydrate and low-fat, high-carbohydrate isoenergetic diets. *Am J Clin Nutr*. 2003; 77:43–50. [PubMed: 12499321]
41. St-Onge MP, Claps N, Heshka S, et al. Greater resting energy expenditure and lower respiratory quotient after 1 week of supplementation with milk relative to supplementation with a sugar-only beverage in children. *Metabolism*. 2007; 56:1699–1707. [PubMed: 17998024]
42. Sunehag AL, Toffolo G, Treuth MS, et al. Effects of dietary macronutrient content on glucose metabolism in children. *J Clin Endocrinol Metab*. 2002; 87:5168–5178. [PubMed: 12414888]
43. Veldhorst MA, Westerterp KR, van Vught AJ, et al. Presence or absence of carbohydrates and the proportion of fat in a high-protein diet affect appetite suppression but not energy expenditure in normal-weight human subjects fed in energy balance. *Br J Nutr*. 2010; 104:1395–1405. [PubMed: 20565999]
44. Weir JB. New methods for calculating metabolic rate with special reference to protein metabolism. 1949. *Nutrition*. 1990; 6:213–221. [PubMed: 2136000]

Figure 1.

A) Serum insulin concentration following consumption of the test breakfast meal on the specialized diet (SPEC) and standard diet (STAN). Test meal ingested at time=0. B) Blood glucose concentration following consumption of the test breakfast meal on the specialized diet (SPEC) and standard diet (STAN). Test meal ingested at time=0. * $p > 0.05$, ** $p > 0.01$

Table 1

Nutrition Composition of SPEC and STAN Diets

	Standard	Specialized
Calories	2019±67	2058±55
Carbohydrate (g)	287±10*	226±6
Fat (g)	60±2*	90±2
Protein (g)	91±3	102±3
Fiber (g)	19±0.6*	29±0.7
Fructose (g)	12±0.4*	20±0.5
Cholesterol (mg)	178±3.4*	302±4.3
Glycemic Index[†]	503±23.6*	346±29.8
Glycemic Load[†]	255±9.3*	129±7.4

[†] bread reference,

* significant difference between diets; p<0.05

Table 2

Baseline descriptive characteristics

	Total	STAN (n=14)	SPEC (n=12)
Age (yrs)	12.4±0.3	12.5±0.4	12.4±0.5
Tanner Stage	4.4±0.2	4.4±0.3	4.4±0.3
Height (cm)	159.5±1.5	159.3±2.0	160.3±2.3
Weight (kg)	89.6±4.6	93.8±7.5	85.1±4.8
BMI %	98.0±0.3	98.1±0.3	97.9±0.5
BMI z-score	2.4	2.5	2.2
Trunk Fat (%)	46.1±1.4	48.1±1.7	43.9±2.3
Leg Fat (%)	44.8±0.8	46.5 ^a ±1.1	42.9 ^b ±1.1
Total Fat (kg)	38.59±2.4	41.09±3.6	35.2±2.7
Percent Fat	43.9±1.0	45.8 ^a ±1.2	41.8 ^b ±1.5
Total Lean (kg)	46.0±2.2	47.4±3.4	44.2±2.3
Insulin (uU/ml)	16.0±2.0	17.1±3.6	14.7±1.4
Glucose (mg/dl)	96.2±1.6	96.2±2.0	96.2±2.6
SI ($\times 10^{-4} \text{ min}^{-1} / (\text{uIU/ml})$)	3.4±0.5	3.14±0.5	3.74±1.0
Cholesterol (mg/dl)	158.9±5.8	166.2±6.4	148.4±10.3
TG (mg/dl)	62.81±4.6	62.95±6.9	62.6±5.7
HDL (mg/dl)	41.91±1.8	43.53±2.5	39.5±2.3
LDL (mg/dl)	104.47±4.9	110.04±5.7	96.3±8.4
Energy intake (kcal/d)	2015	2031	1991
REE (kcal/d)	1667±61.0	1639±86.7	1700±88.4

^{a,b} superscripts indicate significant difference between diet groups $p < 0.05$. STAN = standard diet (55% calories from carbohydrate). SPEC = specialized diet (43% calories from carbohydrate)

Table 3

Comparisons in metabolic parameters at Baseline and at the end of the eucaloric phase by diet

	STAN			SPEC			p-values	
	Baseline	5-week	Baseline	Baseline	5-week	Diet	Time	Diet*Time
Fasting Insulin	17.3±3.0	18.9±2.6	14.3±3.3	21.0±2.9	0.900	0.019	0.139	0.139
Fasting Glucose	96.0±2.2	98.1±2.2	95.6±2.5	99.7±2.4	0.839	0.111	0.545	0.545
SI	3.0±0.7	2.8±0.7	3.9±0.8	3.7±0.8	0.413	0.680	0.914	0.914
Cholesterol	165.1±7.7	157.5±6.1	151.7±9.5	144.2±7.5	0.196	0.622	0.992	0.992
Triglycerides	63.6±6.6	76.5±7.1	64.3±8.0	49.4±8.7	0.207	<0.001	0.001	0.001
HDL	42.6±2.3	42.1±1.9	40.5±2.8	40.8±2.3	0.583	0.170	0.761	0.761
LDL	109.8±6.6	100.1±5.6	98.4±8.0	93.6±6.8	0.329	0.069	0.569	0.569
Adiponectin	6.2±0.7	7.0±0.7	4.3±0.9	4.7±0.8	0.064	0.100	0.359	0.359
Leptin	30.6±2.3	30.3±2.3	30.9±2.9	23.9±2.8	0.340	0.335	0.096	0.096

STAN = standard diet (55% carbohydrate). SPEC = specialized diet (43% carbohydrate). **Bold** values represent statistically significant differences, $p < 0.05$. *Italicized* values represent $0.10 > p > 0.05$. SI = insulin sensitivity index derived from mathematical modeling data of insulin response to a liquid meal test; HDL = high density lipoprotein cholesterol; LDL = low density lipoprotein cholesterol. p-values: diet column represents the comparison between the two diets; the time column represents the significance level of the change from baseline and the diet*time interaction term allows for the comparison of the changes over time between both diets.

Table 4

Change in weight and adiposity after five weeks (eucaloric phase), from week five to week sixteen (hypocaloric phase) and after sixteen weeks (over the course of the intervention).

	Eucaloric		Hypocaloric		Intervention	
	STAN	SPEC	STAN	SPEC	STAN	SPEC
Weight (kg)	-1.4±0.7*	-2.3±0.7**	-3.5±0.7***	-1.3±0.5**	-4.5±1.0**	-5.0±0.9***
Total fat (kg)	-1.4±0.4**	-1.4±0.4**	-1.8±0.8*	-1.9±0.9*	-2.9±0.9**	-3.4±1.0**
%fat	-0.9±0.3**	-0.3±0.4	-2.6±0.5***	-1.9±0.8*	-3.2±0.7**	-3.4±0.9*
Leg fat (kg)	-1.1±0.3**	-0.7±0.2**	-2.0±0.5**	-1.4±0.7*	-2.8±0.6**	-2.1±0.6**
Trunk fat (kg)	-0.9±0.4	-0.5±1.2	-3.3±0.6**	-2.0±1.6*	-3.7±0.8***	-2.6±2.4**

STAN = standard diet (55% of calories from carbohydrate). SPEC = specialized diet (43% of calories from carbohydrate).

p-value for significant change from baseline *<0.05,

**<0.01,

***<0.001.