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The Relationships between Energy Balance Deviations and Adiposity in Children and Adolescents

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ACCEPTANCE

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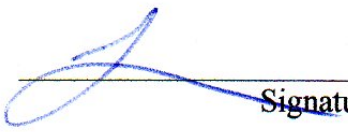

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ABSTRACT

THE RELATIONSHIPS BETWEEN ENERGY BALANCE DEVIATIONS AND ADIPOSITY IN CHILDREN AND ADOLESCENTS

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Background: Over the past decade obesity has doubled in children aged 6-11 and tripled among adolescents aged 12-19. One trend that has coincided with this increased obesity prevalence is decreased meal frequency, which may impact blood sugar, meal size, cortisol release, insulin release, and appetite controls that include the release of leptin and ghrelin. Ultimately, these changes may result in a simultaneous lowering of the metabolic (i.e., fat-free) mass and a rising of the fat mass. **Purpose:** To assess food/beverage intake in a way that would determine if large deviations in energy balance (EB) during the day were related to body fat percent (BF%). **Methods:** Using an IRB-approved protocol, male and female children were assessed using NutriTiming® (NutriTiming LLC, 2011) software, which simultaneously assesses energy intake from consumed foods and beverages and energy expenditure from activities with different intensities. A 24-hour recall questionnaire and interview, with at least one parent present, was used to obtain data, which represented a typical school day. BF% was assessed using an 8-mode bioelectrical impedance segmental body composition analyzer (Tanita, Model BC-418). Statistical analysis was performed with SPSS (ver. 18). **Results:** A total of 16 children ranging in age from 8-14 years were interviewed. Due to incomplete data on 4 subjects, 12 subjects (6 boys; 6 girls ranging in age from 9-14 years; mean=11.41 ± 1.5) were included in the data analysis. Energy intakes averaged 1,984 ± 510 kcal; and energy expenditure averaged 1,689 ± 351 kcal. Average BF% was 24.3 ± 4.9. Using Spearman correlation and independent group t-test (with the mean energy balance as the cut-point) traditional end-of-day energy balance (24-hr energy in vs. 24-hr energy out) was not statistically associated with body fat %, and there was no difference in BF% between those above and below the end-of-day EB mean. However, more hours spent in an energy surplus (EB > 0) was significantly associated with lower body fat % (r=-0.914; $P<0.001$), while spending more time in an energy deficit (EB < 0) was significantly associated with higher BF% (r=0.914; $P<0.001$). **Conclusions:** These data strongly imply that avoiding long periods of time in EB deficits would be useful for reducing body fatness in children. Long times in EB deficits may result from excessive time between eating opportunities, physical activity that results in faster energy expenditure, or any combination of these. Schools and organizations working with children should consider instituting periodic snacking strategies to assure a better dynamic relationship between energy consumed and energy expended throughout the day. This will reduce the chance that children may experience long time periods in EB deficits that, according to these data, are associated with higher BF%.

THE RELATIONSHIPS BETWEEN ENERGY BALANCE DEVIATIONS AND ADIPOSITY IN CHILDREN AND ADOLESCENTS

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ABBREVIATIONS

% kcal from CHO: Percent of kilocalories from carbohydrates

% kcal from FAT: Percent of kilocalories fat

% kcal from PRO: Percent of kilocalories from protein

AIR: Acute insulin response

ALT: Alanine aminotransferase

Anabolic to Catabolic: Ratio of hours spent in an anabolic state to hours spent in a catabolic state

AST: Aspartate aminotransferase

B: Standardized breakfast

CDC: Centers for Disease Control and Prevention

CDs: Current dieters

DEXA: Dual-energy X-ray absorptiometry

DI: Disposition index

EB: Eating breakfast

Energy Balance: End of day energy balance

FAST: Fasting

FFA: Free fatty acids

GERD: Gastroesophageal reflux disease

HEI-C: Healthy Eating Index-Canada

HIFREQMEAL: High-frequency meal pattern

Highest EB Surplus: Highest energy balance surplus

Hours + or- 400kcal: Hours spent in an energy balance above or below 400 kcal

Hours>400Kcal: Hours spent in an energy balance above 400 kcal

Hours < 0 EB: Hours spent in an a negative energy balance

Hours < 0 EB: Negative energy balance

Hours<400Kcal: Hours spent in an energy balance below 400 kcal

HRNDs: High restrained non-dieters

IAAT: Intra-abdominal adipose tissue

IMP: Impulsivity phase

INP: Inhibitory phase

ISI: Insulin sensitivity index

Kcal Consumed: Kilocalories consumed

Kcal Consumed/kg: Kilocalories consumed per kilogram of body weight

Kcal Expended: Total daily kilocalories calories expended

Kcal expended: Total daily kilocalories expended

LOFREQMEAL: Low-frequency meal pattern

Lowest EB deficit: Lowest energy balance deficit

LRNDs: Low restrained non-dieters

MetS: Metabolic syndrome

NAFLD: Non-alcoholic fatty liver disease

NB: No breakfast

NHANES: National Health and Nutrition Examination Survey

OB: Omitting breakfast

Ratio surplus to deficit: Ratio of energy balance surplus to energy balance deficit,

Trunk BF %: Trunk body fat percentage

WC: Waist circumference

CHAPTER I

INTRODUCTION

Childhood obesity prevalence is on the rise with an unprecedented velocity curve. Between the years of 1976-1980 and 2005-2006, the incidence of childhood obesity doubled in children aged 6-11 years and tripled among adolescents aged 12-19 years (1). Dietary trends that coincide with this increase include, increased snacking, portion sizes, consumption of fast foods and decreased meal frequency (2). Several studies have investigated meal frequency and found that increased meal frequency positively correlates with satiety and negatively with total caloric intake. Eating fewer meals, therefore results in greater hunger between eating episodes, which may explain the tendency for meal skippers to overcompensate calorically following the skipped meal/meals (3). Therefore, people who eat less frequently, and especially meal skippers, are prone to greater energy balance surpluses and greater weight gain than individuals who eat more regularly (3,4,5).

One explanation for increased weight gain in individuals who skip meals is their tendency to consume toward a high level of fat and lower quality foods. Several studies have observed this trend between meal skipping and high fat intake (6,7). However, it is unclear which behavior influences the other. On the one hand, the increased-satiety level of the high fat diet may enable an individual to take more time between eating occasion

(5). On the other hand, meal skipping may encourage behaviors that require more energy dense meals as a way to compensate for the meal or meals that were skipped (8). Regardless of the cause, a consequence of meal skipping is often an increase in adiposity and the adverse health effects that accompany excess weight gain (7).

Other consequences of infrequent meals are increased prevalence of cardiometabolic risk factors. Several studies have found that individuals who skip meals present with both higher total cholesterol and higher LDL cholesterol (7,9), and other studies indicate an increase in both total and systolic blood pressure (10,11,12). There is also evidence that people subjected to prolonged periods of fasting, have decreased insulin sensitivity (9,13,14). This lower insulin sensitivity translates into many metabolic disturbances, one of which is delayed ghrelin suppression. Ghrelin stimulates appetite, therefore, it is hypothesized that the internal signals intended to turn off appetite are compromised when a meal is skipped and the time between eating opportunities is lengthened (15,16). Finally, when meal skipping becomes habitual, it is possible that leptin levels drop to increase appetite as a mechanism of survival. It appears that irregular energy intake causes a hormonal adaptation that signals the body to store energy, even when it is not necessary (17,18).

Children who are overweight are 1.5 to 2 times more likely to be overweight as adults than their normal weight peers (19). This higher risk of adult obesity also means a higher risk of developing chronic diseases. There is strong epidemiological evidence implicating overweight and obesity in many inflammatory conditions, such as diabetes, cardiovascular disease, and asthma (20,21,22,23). Other conditions associated with childhood obesity include metabolic syndrome (MetS), and non-alcoholic fatty liver

disease (NAFLD). Fortunately, the severity of these diseases can be greatly reduced, or the disease itself eliminated with weight loss (24).

Studies have investigated the connection between low meal frequency and the rise in overweight and obesity; however, no studies have looked at the relationship between body fat percentages and with-in day energy balance in children. Also, no research has been conducted to quantify the amount of time children spend in energy balance deficits and energy balance surpluses during a full 24-hour period. This study tracked the amount of time children spent in energy deficits and surpluses to determine if sporadic meal patterns positively correlated with increased body fat.

Hypothesis 1: Children who experience greater energy balance deficits have higher adiposity than children who do not experience these deficits.

Null Hypothesis 1: Children who experience greater energy balance deficits do not have higher adiposity than children who do not experience these deficits.

Hypothesis 2: Children who experience greater energy balance surpluses have higher adiposity than children who do not experience these surpluses.

Null Hypothesis 2: Children who experience greater energy balance surpluses do not have higher adiposity than children who do not experience these surpluses.

CHAPTER II

REVIEW OF LITERATURE

The Effects of Energy Balance Deviations on Dietary Quality and Habits

One factor associated with decreased meal frequency is the migration toward less nutrient dense foods. In the 1999-2000 National Health and Nutrition Examination Survey (NHANES), 20.5% of 9 to 13 year-old children and 36.1% of 14 to 18 year-old adolescents skipped breakfast. Furthermore, these children who were skipping breakfast also had reduced intake of essential nutrients and recommended fruits and vegetables (25). Therefore, the foods that they are eating are lower in fiber and complex carbohydrates, both of which are shown to improve satiety and maintain a more stable insulin response (26).

Another habit that has increased as meal frequency has decreased is the replacement of meals with low nutrient dense snacks (2). Furthermore, these low nutrient dense snack foods like pastries and cakes are also high in hidden fats. Hidden fats lack the same sensory characteristics of creaminess or crispness that visible fats have such as ice cream or chips (5). In adult obesity studies, a diet high in hidden fats was most often associated with overweight and obese individuals (4). A diet high in hidden fats is likely to lead to overweight or obesity, because hidden fats do not have the same affect on satiety that visible fats do (5). Therefore, the subjects ingesting hidden fats took longer to reach perceived fullness, ingested more calories and had greater energy surpluses. Furthermore,

overconsumption of visible fats in one meal led to less energy consumption in the preceding meal, whereas overconsumption of hidden fats did not lead to the same energy corrections (5).

Taking into account the results of the previous studies one can assume that individuals consuming diets high in hidden fats are also consuming a greater amount of fat throughout the day than those who eat more visible fats. Melhorn et al. (2010) showed that this increased fat intake might negatively affect meal frequency. In their study of mice, subjects were freely fed either a high fat diet (20% fat, 4.54kcal/g) or a chow diet (5% fat, 3.46 kcal/g). Each group maintained their respective diets for nine days. The rats that consumed the high fat diet consumed more calories, ate larger meals and ate less frequently. Furthermore, the increased meal size and caloric load of a high fat diet also correlated positively with greater adiposity (27). Meal skippers are known to exhibit these same behaviors: a high fat diet, with the majority of calories consumed in one part of the day (28,29).

Another contributing factor to adolescent meal skipping is the purposeful desire to restrict calories to achieve weight loss. In a Canadian study of 1,826 9^{th} and 10^{th} graders, teens were asked to answer questions on body image and dietary habits. Twenty-eight percent of the participants surveyed admitted to worrying about high body weight and about 20% admitted to currently being on a diet (30). One way of restricting calories as reported by this population was to skip meals. Approximately 27%, 14%, and 7% (n = 129) of the participants skipped breakfast, lunch, and dinner, respectively. Though the tendency toward skipping meals in this study was the same across dieting and non-dieting populations, the researchers did find that meal skipping correlated with a poorer diet

quality as evidenced by low Healthy Eating Index-Canada (HEI-C) scores (30). This low HEI-C score may, in turn, make a child more at risk for becoming overweight or obese as one British study indicates. This study, which examined the dietary habits of 1,700 9-10 year olds, found an inverse relationship between diet quality scores and BMI ($P = 0.014$), BMI, waist circumference (WC), and percent body fat (31). These findings further support the idea that the poor dietary choices related to meal skipping may place children at a higher risk for excess adiposity.

Adolescents who restrict calories and skip meals (also referred to restrained eating) for the purpose of weight loss or management are also at a higher risk of developing anorexia nervosa and bulimia. Restrained eating is also associated with overweight and obese individuals with disordered eating (32). In obese subjects, restrained eaters often eat more than they need to stay in an energy balance, thus they are more likely to gain weight than unrestrained eaters. Markowitz et al. (2008) were interested investigating this phenomenon further. Their study consisted of 66 college females with no prior histories of eating disorders. These females were asked to complete 2 questionnaires (33). The first was, the Restraint Scale, which is a self-reported measure that determines the presence of dieting and weight concerns. The second questionnaire was a perceived deprivation questionnaire, which assessed the participants' satisfaction with the amount and types of food they ate (33). Their study revealed a significant correlation between perceived deprivation and restraint ($r = 0.28$, $p = 0.03$). Furthermore, they concluded that restrained eaters were prone to limiting what kinds of foods they allow themselves to eat, but not the amounts of food they eat (33).

Guerrieri et al. (2009) looked at restrained eating and its relationship to self-control.

In this study 66 female undergraduate students were recruited into the study under the belief that they were taking part in a study that involved, “cognitive performance and taste perception” (32). In the first stage of the study, participants completed a hunger scale questionnaire. Next they performed the Stop Signal Task, which was composed of two phases: first an impulsivity phase (IMP) followed by an inhibitory phase (INP) (32). In the IMP participants were asked to push a button as quickly as possible in response to a tone. In the INP, they were asked to inhibit their response when they heard a tone. Therefore, in both phases was response was the same, to hit a button in response to a tone. However, in the IMP the participants were told that reacting as fast as possible was more important than inhibiting their reaction. In the INP the participants were told that inhibiting their reaction was more important than reacting as fast as possible. After the Stop Signal Tasks, participants performed a bogus taste test, which was actually a measure of the amount of food a person would consume following the IMP and INP phases. Once the data were collected, the participants were separated into 3 groups, current dieters’ (CDs; n = 15), low restrained non-dieters’’ (LRNDs; n = 25), highly restrained non-dieters (HRNDs; n = 26). The CDs showed the most restraint on the IMP as opposed to the INP, and consistently overate in the taste test. This behavior is explained according to the Ego-Strength Model of Self-Regulation, which indicates that self-control is limited. The CDs only had so much self control after the IMP, that they were unable to hold back in the INP and by the time the reached to taste test their self-control was lost (32). This behavior is characteristic of restrained eaters and can explain why their tendency to become increasingly less inhibited about dietary restrictions after consumption of a diet-prohibited food (33). This limited self-control could explain meal

skippers' tendency to overeat - after exhibiting long periods of self-restraint, their self-control reaches its tipping point and the end result is uninhibited, overconsumption of calories. If this behavior becomes habitual, the result could be an overweight or obesity problem.

The tendency to overeat after a period of fasting is demonstrated in the Bakhotmah et al. (2011) study of Saudi Muslim's during Ramadan. During Ramadan, Muslims abstain from all food and water from dawn to dusk (8). In their study 173 Saudi families were interviewed about their changes in energy expenditure, lifestyle, meal frequency, dietary habits, food consumption and preferences, during Ramadan. Fifty-nine participants reported gaining weight instead of losing weight during this time (8). A significant weight gain was considered to be 3 kg or more. Researchers discovered that not only did physical activity decline (a plausible cause for weight gain), but there was also an increased intake of fatty, carbohydrate rich foods (8). Therefore, the traditional foods for this time of year appeared to compensate for the loss of midday meals by increasing the energy composition of their meals in the morning and at nighttime (8).

The establishment of favorable dietary habits are important in childhood because, as several longitudinal studies have observed the dietary habits of children often become progressively worse with increasing age. Smith et al. (2010) found that children who skip breakfast often grow into adults who skip breakfast, and the same dietary problems that exist in childhood carry into adulthood. These adults were found to be less likely to meet the dietary recommendations for fruit and dairy and more likely to eat restaurant food once or more times per week (7). Niemeier et al. (2006) found that children who consumed fast food 2 times a week, increased to 2.5 times a week by adulthood. It was

also found that those who consumed breakfast 4 to 5 times a week were eating breakfast only 3 times a week by adulthood (34).

The Effects of Energy Balance Deviations on Hormones and Appetite

The studies of meal frequency and insulin response yield conflicting results. Alexander et al. (2009), examined glucose dynamics in a group of overweight Latino adolescents at risk for type 2 diabetes, meaning a BMI≥85% and a family history of diabetes. They performed fasting oral glucose tolerance tests, insulin sensitivity index (ISI), acute insulin response (AIR), and disposition index (DI). DI is the product of AIR and ISI, and indicates the measure of β-cell function (13). The researchers of this study were unable to find any significant difference in insulin function between the breakfast skippers and the non-breakfast skippers (13). Farshchi et al. (2004) worked with a lean population, and found that consumption of a full day's calories in one meal as opposed to a normal diet, resulted in a significantly higher peak insulin response ($P<0.01$) following the test meal (14). This increased insulin response may be due to a decrease in insulin sensitivity following a period of fasting (14,35). One reason for a discrepancy between the groups is that in Alexander et al.'s (2009) study, individuals with impaired glucose tolerance were not excluded from the study. Therefore, insulin function was already compromised in portion of their study participants and their results could have skewed the data (13).

One study examined fasting and postprandial glucose reactions in a population of 9 healthy, lean females aged 18–42 years. This study was a 14-day randomized crossover trial that assessed the effects of eating breakfast (EB) as opposed to omitting breakfast (OB) on insulin control (9). Their results showed no significant difference in fasting

glucose profiles between the two periods of OB and EB, but there was a significant (P=0.07) increase in serum insulin in the OB period (9).

Stein et al. (1996) used rats to measure insulin sensitivity after a period of fasting. Their study showed a decline in insulin sensitivity (≈80%) after a period of 18-24 hour fast in rats. Besides documenting insulin's response to fasting, the researchers in this study were also interested in investigating the mechanism behind this response (36). After several trials, they concluded that the presence of free fatty acids (FFA) were essential to β-cell sensitivity when transitioning from a fasted to fed state. In a fed state, glucose metabolism leads to the production of malonyl-CoA [essential for fatty acid-CoA (FA-CoA) synthesis] and suppresses the production of CPT-1 (responsible for catalyzing the first step in long chain fatty-acid oxidation) (36). This FA-CoA might be involved in signaling insulin secretion from β-cells. In a starved state, Acetyl-CoA carboxylase, essential for the production of FA-CoA, is down-regulated in the liver and insulinoma cells thus compromising malonyl-CoA synthesis (36). Therefore, if the cell were unable to elevate its cytosolic FA-CoA concentration in response to glucose, there would be inefficient insulin secretion. However, if a high external concentration of FFA is available (as in a fasted state), then FA-CoA should again be available and insulin sensitivity reinstated (36). On the other hand, if one function of FA-CoA were to provide phospholipids to β-cells, glycerol-3-phosphate (typically derived from glucose) would also be needed to complete the cell membrane. This explains why in a re-feeding phase both FFAs and glucose are needed for efficient insulin secretion (36). However, as these experiments found, when glucose is reintroduced, FFA release is ablated leading to an

insufficient marriage of FA-CoA and glycerol-3-phosphate, thus compromised β-cell function (36).

Zawalich et al. (2000) used fasted mice and further examined the mechanism behind compromised insulin function after a period of fasting. Their mice were fasted for a period of 24 hours then fed varying amounts of glucose. Researchers then compared the islet activity of fed mice to the fasted mice and discovered a 90% reduction in the insulin secretory responses to 8 mM glucose in the fasted mice (37). When 11 or 27.5 mM of glucose was administered, the result was enhanced insulin secretion from the islets of fasted animals. Therefore, researchers discovered that the insulin content of the islets was not compromised by fasting, but islet sensitivity was (37).

In another experiment by Farshchi et al. (2004), researchers were able to show that irregular meal patterns also have an effect on insulin sensitivity. In this study, 9 lean healthy women ages 18–42 were asked to maintain a regular meal pattern (6 eating occasions a day) for 14 days followed by an irregular meal pattern (3-9 eating occasions, alternating) (14). Diets across both phases were isocaloric. At the end of the experiment, researchers discovered that the irregular meal pattern resulted in reduced insulin sensitivity and a greater insulin response to a test meal compared with the regular meal pattern (14).

Insulin has many roles in appetite regulation, such as, ghrelin suppression. More specifically, when insulin is released, ghrelin hormone is suppressed, leading to a decrease in appetite (38). Some researchers believe that prolonged fasting disrupts ghrelin (an appetite stimulating hormone) function. Solomon et al. (2008) investigated the effects of ghrelin release after a period of fasting. Their crossover study included 5 lean, male

volunteers, aged 24 years (SEM 2 years), with a body mass index of 75.7 kg (SEM 3.2 kg) and a BMI of 23.8 kg/m2 (SEM 0.8 kg/m^2) (16). Participants underwent three 8-hour feeding regimens: 8 hours of fasting (FAST); a low-frequency meal ingestion (2 meals in 8 hours) (LOFREQMEAL); a high-frequency meal ingestion (12 meals in 8 hours) (HIFREQMEAL). Their results revealed an inverse relationship between serum insulin and plasma ghrelin during the FAST and LOFREQMEAL trials ($P<0.05$) (16). In the postprandial period of the LOFREQMEAL trial, there was a time delay, approximately 20 minutes, between insulin responses and successive ghrelin responses. This relationship was not observed during the HIFREQMEAL trial ($P<0.05$) (16). Though the exact mechanism of this delay is unclear, the researchers believe that it may be the result of a disconnect in hormone – hormone interaction or insulin does not fluctuate sufficiently to influence ghrelin (16). A study by Anderwald et al. supports the latter explanation. In this study a group of diabetic (n=6) and non-diabetic (n=6) subjects aged 56-68 years were injected with increasing doses of glucose to study the effects of insulin release on ghrelin (15). The insulin insensitive diabetics did not experience the same decline in plasma ghrelin as the non-diabetic subjects following the glucose treatments. Therefore, it is believed that the insulin release is directly related to ghrelin suppression, thus suppressed appetite (15).

Another hormone that may be disrupted by meal skipping is leptin. Leptin's function is opposite of ghrelin; leptin is an appetite suppressing hormone that also increases thermogenesis and has also been called the "anti-obesity" hormone (17). Kolaczynski et al. (1996) observed leptin responses to short-term fasting and re-feeding in humans. In their experiment, subjects began with a 36-hour baseline period, followed

by a 36-hour fast and then subsequent re-feeding (18). By the experiment endpoint, the researchers were able to conclude that habitual fasting and re-feeding creates an adaptive response in humans, leading to lower levels of serum leptin. Therefore, if a person is constantly experiencing long periods of starvation, the body turns off the mechanisms that would suppress appetite as a mode of survival, because it is uncertain when the next meal may be coming (18).

De Castro (2004) analyzed the intakes of 375 male and 492 female free-living adults for the purpose of observing how circadian and diurnal rhythms influence energy intake and appetite. He also recorded continuous satiety levels of these individuals as they progressed through their day (39). He found that those who ate the majority of their calories in the evening hours had greater total energy intakes and reported more hunger throughout their day than those who had more balanced intakes. De Castro (2004) theorized that hunger increases from morning to evening, independent of caloric intake, as a mode of natural circadian rhythm (39). Therefore, skipping meals in the early part of the day only compounds an individual's hunger and leads to overcompensation in the evening meal. On the other hand, eating sufficient amounts in the morning counterbalances natural circadian rhythms and decreases the likelihood to overeat in subsequent meals (39).

Astbury et al. (2011) looked at breakfast skipping and its effects on appetite, insulin release and ghrelin levels. In this randomized crossover trial, 12 healthy male participants who reported regularly consuming breakfast (aged 23.4 ± 7.3 years; BMI 23.5 ± 1.7 kg/m^2) were given a liquid preload 150 min after consuming a standardized breakfast (B) or no breakfast (NB). Lunch and dinner did not vary between trials (40). A

dorsal hand vein cannula was inserted to monitor blood values and participants' lab values continuously. In addition, appetite ratings were also continuously monitored. In response to the preload, serum insulin responses were higher in the NB condition ($P < 0.05$). In regards to appetite, hunger ratings were lower and fullness ratings were higher in the B trial as opposed to the NB trial immediately following the pre-preload ($P < 0.05$). There were no significant differences in plasma ghrelin levels after meals or preloads; however, the minimum ghrelin concentrations were lower in the B trial than the NB trial ($P < 0.05$), which might explain the higher hunger ratings in the NB trial (40). Energy intake at lunchtime after the B trial was reduced by 17% and participants reported feeling fuller and less hungry in response to the preload (40).

Cortisol is another hormone disrupted by meal skipping. Under conditions of mild stress, such as dietary restriction, this catabolic hormone experiences enhanced release in order to make energy more readily available for the body to use. It does this through mobilizing fat and other bodily tissues in order to synthesize glucose for energy (41).

The Effects of Energy Balance Deviations on Body Composition

A number of studies have found that decreased meal frequency correlates with greater daily energy consumption. This increased energy consumption may be a result of an up-regulation of appetite or tendency toward increased fat intake (3,7). If this increase in energy consumption is not matched by increased activity, the result is often excess adiposity. In a study of 2,379 African American and Caucasian girls aged 9 -10 years, this exact result was found (42). Even after adjusting for television watching and physical activity, BMI-for-age z-scores for the girls who ate 3 meals on most days were

significantly ($P < 0.001$) lower than the girls who skipped meals on most days (42). Berkey et al. (2003) also looked at BMI and meal frequency, but this study did not show the same effects of meal skipping on BMI. They discovered that overweight children who habitually skipped breakfast over a period of one year, actually had a decrease in BMI (43). The 16,882 participants were children of Nurses' Health Study II participants. All anthropometric measures and food frequency questionnaires were self-reported and collected via mail. Researchers attributed their weight loss to decreased caloric intake in the meal skippers (43). However, an interesting paradox is, that the majority of the participants who skipped breakfast were overweight. Also, the breakfast skippers had lower diet qualities and were less likely to engage in physical activity, both of which are risk factors for developing into an overweight or obese adult (43).

Another trend among meal skippers is an increased waist circumference (WC). Smith et al. (2010) followed a large cohort of adolescents through adulthood for the purpose of monitoring their breakfast eating habits and the cardiometabolic risk factors that accompanied these habits. The researchers discovered that the participants who skipped breakfast only as adults had a significantly larger WC and BMI than those who did not skip (P=0.008) (7). Those who skipped breakfast as children and then again as adults had even larger waist circumferences and BMIs than those who did not skip breakfast at all or who only skipped as adults, showing perhaps a cumulative effect of breakfast skipping over time. These results also correlated with a higher amount of cardiometabolic risk factors, such as higher HDL and decreased insulin sensitivity (7). Isacco et al. (2010) also looked at breakfast skipping WC, skin fold measurements, and BMI z-scores. In their study of 278 healthy French children, study participants were

divided into 3 separate groups: breakfast eaters, occasional breakfast skippers, and breakfast skippers. The two breakfast skipping groups had significantly ($P < 0.001$) higher WC, skinfold measurements, and BMI z-scores than the group that never skipped breakfast (44). This study demonstrated that even occasional breakfast skipping correlates with increased weight.

Alexander et al. (2009) were also interested in abdominal obesity and breakfast skipping, and measured the amounts of intra-abdominal adipose tissue (IAAT) in a group of overweight Latino youth using dual-energy X-ray absorptiometry (DEXA) (13). Independent of age, Tanner Stage (stage of maturation), gender, total body fat, total body lean tissue mass, and daily energy intake, the children who skipped breakfast had significantly higher IAAT (47.2 ± 3.8 vs. 32.1 ± 2.7; $P = 0.004$). These results are significant because higher amounts of IAAT often correlate with an increased risk for dyslipidemia and glucose intolerance (13).

Some researchers theorize that meal timing, in addition to frequency has an effect on body composition. In one study, nocturnal mice were separated into two groups. The first group was fed a high fat diet only in a 12-hour light period and the second group was fed the same diet only in a 12-hour dark period (29). The mice that were fed in the light period gained significantly ($P < 0.004$) more weight, despite similar caloric intake and physical activity levels. The researchers of this study believe that feeding in the light phase feeding may have caused a decrease in dark phase body temperature, which could have contributed to the increased fat storage and weight gain (29). This could explain why humans, a diurnal species, who eat the majority of their calories in the nighttime

hours (a habit of meal skippers) have a tendency to gain more weight than those who space their calories out more evenly throughout the day (39).

The Effects of Energy Balance Deviations on Blood Pressure and Lipids

Increased blood pressure and cholesterol levels are often associated with overweight and obesity. As the following literature indicates, they are also associated with meal skipping. According to the National Health and Nutrition Survey of Japan, people who skip breakfast have higher mean levels of blood pressure, serum total cholesterol and are less likely to be physically active (10). All of these habits are risk factors for developing heart disease. Umemura et al. (2005) found these characteristics to be true in their study of 421 healthy college students aged 18–20 living in Tokyo (10). In addition to a low consumption of fruits and vegetables, their study showed that the students who skipped breakfast had higher systolic and diastolic blood pressures levels (10). Kollias et al. (2009) found similar effects on blood pressure in a cohort of 558 Greek adolescents aged 12-17 years. The researchers gathered data on their subjects at the beginning of the study and then again after 3 years (11). At each visit the subjects were given a brief questionnaire on dietary and physical activity habits. Anthropometric data (BMI) were also collected and blood pressures were assessed. While their study was looking at multiple risk factors, they were able to specifically draw a significant correlation between increased systolic blood pressure and skipping meals (11).

Skipping meals also often correlates with hyperlipidemia. Farschi et al. (2005) also demonstrated that in their study of breakfast eaters and breakfast omitters. The participants of this study were subjected to two phases, two weeks where breakfast was

eaten and two weeks where breakfast was omitted (9). No significant effects to cholesterol were seen after the two weeks of EB, however, after two weeks of OB, participants experienced a significant increase in total plasma cholesterol (P=0.02). Furthermore, it was the LDL cholesterol (P=0.001) that increased not the HDL cholesterol (9). In Smith et al.'s (2010) previously mentioned study on breakfast skipping and cardiometabolic risk factors, the participants who skipped breakfast both as children and adults had significant high total cholesterol levels (P=0.005). While there were no statistically significant differences in the incidence of total high cholesterol amongst the study groups, those that skipped breakfast at both time points did have a higher incidence of high LDL cholesterol (P=0.037) (7).

In a controlled crossover trial, 32 healthy, normal-weight, middle-aged adults were placed on a 3-meal a day diet regime for 8 weeks, followed by a 1-meal a day regime without caloric restriction for another 8 weeks (12). While the participants did experience weight loss (1.4 kg) following the 1-meal a day trial, there was also a significant increase in diastolic and systolic blood pressures ≈ 6% (P<0.05) and total cholesterol (P=0.001). The researchers admitted to a 65-calorie deficit in the 1-meal a day trial, which may have contributed to the weight loss (12). Edelstein et al. (1992) were also interested in observing the effects of meal frequency and cholesterol and blood pressure levels. In their study, 2,034 Caucasian men and women aged 50-89 were divided into two categories, those that ate 4 or more meals a day and those that ate 1-2 meals a day (45). Those that ate 4 or more meals a day had 0.23mmol/L lower total cholesterol (P=0.01) and 0.16mmol/L lower LDL cholesterol (P=0.06). In addition the more frequent meal eaters had significantly lower systolic blood pressure by 5 mm HG (P=0.004) (44).

Not all studies found this same inverse correlation between meal frequency and poor cholesterol outcomes. Maislos et al. (1993) observed the plasma cholesterol levels of 24 healthy subjects just prior to ending their Ramadan fast and then again a month prior. Despite the higher fat/carbohydrate diets and decrease in physical exercise typical of the time if year, plasma VLDL and LDL-C did not change. However, plasma HDL-C levels ($P<0.005$) were significantly higher, but there was no significant change in BMI or bodyweight (46). The mechanism behind this change was not determined in this study, but the researchers hypothesized that it was most likely a component of their diets. Unfortunately, a major weakness of the study was that no dietary recalls were gathered (46).

Health Concerns of Overweight and Obese Adolescents

Being overweight or obese increases one's risk for a variety of medical conditions, including non-alcoholic fatty liver disease (NAFLD), asthma, and heart disease. One of the most thoroughly researched risks for overweight adolescents is heart disease. The Bogalusa Heart Study ran from 1973-1994, and is one of the longest and most comprehensive of its kind. The purpose of the study was to gain a better understanding of the origins of coronary artery disease and hypertension (23). In regards to overweight children ($\geq 85^{th}$ percentile), they were 2.4 times more likely to have elevated cholesterol levels, 7.1 times more likely to have elevated triglycerides and 12.6 times more likely to have elevated fasting insulin levels. Furthermore, the number of risk factors increased as body weight increased, placing these children at an even higher risk for adverse cardiovascular events in the long term (23).

Overweight children are at a higher risk for elevated fasting insulin levels, which is often an indication of type 2 diabetes. According to the Centers for Disease Control and Prevention (CDC), about 3,600 youth are newly diagnosed with type 2 diabetes every year. The disease is also more difficult to detect in children and adolescents, because children tend to have either no or very mild symptoms and a blood test is needed to diagnose (22). Therefore, the true incidence of the disease in children may be much higher. These rates are concerning, because this disease is typically only seen in people 40 years and older (22). Diagnosis at an early age can mean the complications of diabetes, such as, kidney disease, heart disease or blindness may come at a much earlier age. In addition, a person's risk of death from diabetes is twice that of a healthy person of the same age (47).

About 2.6% to 9.8% of children and adolescents and up to 74% among obese individuals are affected by NAFLD, which is reversible if found early. When children with elevated liver enzymes and fatty livers lost only 10% of their excess body weight, their alanine aminotransferase (ALT) and aspartate aminotransferase (AST) values dropped to normal levels and an ultrasound revealed decreased fatty infiltration of the liver (24). However, NAFLD is often hard to detect, because most people who have the disease feel well. Therefore, many individuals go undiagnosed until they develop cirrhosis, or worst-case scenario, need a liver transplant (48).

It is estimated that 10 to 25% of obese children in the United States have elevated serum aminotransferases and are at risk for developing NAFLD (49). This statistic is significant, not only due to the liver complications related to NAFLD, but also because this disease is attributable to cardiovascular deaths. Studies indicate that these

cardiovascular deaths are related to NAFLD's strong connection with metabolic syndrome (MetS) (49). MetS is defined by possessing three or more of the following conditions: abdominal obesity, hyperlipidemia, hypertension, insulin resistance and impaired glucose tolerance. MetS is closely related to the development of type 2 diabetes and atherosclerosis (50). Chui et al. (2011) studied a group of 550 adults (aged 53.8 ± 10.2 years) who had been referred by a doctor to screen for cardiovascular disease. Of the 550,209 were classified as having MetS and of the 209 with MetS, 74% exhibited with coronary atherosclerosis compared to 46% in the adults without MetS (51). In young adults (mean age 32 years) the incidence of subclinical atherosclerosis (as determined by carotid wall thickness), correlated with a 2.5-fold increased likelihood of MetS (52).

The evidence connecting childhood obesity to asthma is often conflicting. While much of the epidemiological evidence points to a strong connection, many scientific studies have failed to determine a mechanism behind the connection. Two large epidemiological studies, one out of Alexandroupolis, Greece and the other out of the Ashkelon region of Israel indicate that a high BMI is a strong independent risk factor for asthma (P=0.009, $P < 0.001$, respectively) (20, 21). In one of the largest prospective studies of asthma (135,000 participants) healthy asthma-free subjects were followed for a period of 21 years. At the end of the 21 years, this study revealed that every one unit increase in BMI correlated with a 10% increased rate of asthma in men and 7% increased rate in women (53). Some researchers hypothesize that asthma may be a result of elevated inflammatory cytokines. For instance, leptin is often elevated in obese populations. Leptin is also a member of the IL-6 inflammatory cytokine family and thus a factor in the acquisition of obesity-induced asthma. Others believe that asthma is not a

direct result of internal factors, such as inflammatory cytokines, by an indirect consequence of obesity-related co-morbidities, such as gastroesophageal reflux disease (GERD) (53). No matter what the mechanism, it is clear from a large body of epidemiological research that obesity increases the risk for asthma.

Summary

Those who skip meals are putting themselves at a higher risk for a number of health issues. It is unclear whether these health issues are a consequence of corresponding poor dietary and lifestyle habits or metabolic disturbances. However, strong epidemiological evidence does show a strong correlation with skipping breakfast and a higher incidence of overweight and obesity. Furthermore, those that are skipping meals, are more likely to have a higher WC, increased blood pressure, increased cholesterol and compromised insulin function following long periods of fasting. Over time, the increased weight gain and cardiometabolic disorders that accompany meal skipping could lead to a number of health issues, including type 2 diabetes, NAFLD, MetS or asthma. Typically, these health issues are seen only in adults. However, as incidence of childhood obesity continues to grow, children are also contracting them. Therefore, more attention needs to be paid to not only the quality of children's diets, but to the dietary patterns and habits that put them at a higher risk for long-term illness.

CHAPTER III

METHODS

This study assessed the relationship between meal frequency, energy balance, and adiposity in a group of children between the ages of 9 and 14. Subjects in this cross sectional study volunteered via convenience sampling and data collection occurred at two locations. The first location was the Laboratory for Elite Athlete Performance at Georgia State University. The second location was an Atlanta area sports and exercise facility. Researchers set up tables with the study materials and equipment and children and parents were seen by appointment. Using an IRB-approved protocol, both parental permission and child assent were required before admission into the study.

Data were collected on 16 subjects. Each subject had at least one parent or guardian present during the collection of the data. Three subjects were excluded from the study due to incomplete consent forms and a forth was eliminated for incomplete anthropometric data, leaving a total of 12 subjects (6 males/6 females).

The first part of the interview involved a single 24-hour recall of all dietary intakes and activities of a typical school day. Subjects were asked to approximate portion sizes to the best of their ability. If portion sizes were not provided, they were estimated via Food Guide Pyramid suggested serving sizes. Subjects were asked to rate the intensity of their activity according to the chart provided (Appendix E). Activity ranged from sedentary to intense exercise.

Anthropometric measures were taken on each subject using a Tanita BC-418 Segmental Body Composition Analyzer. Scale reported on weight, lean mass, fat mass, BMI and segmental body composition. Height was verified using a stadiometer. Shoesand socks were removed before stepping on the scale; all other clothes remained on during bioelectrical impedance analysis. A default of 2 pounds was subtracted from all subjects' weights to account for the extra weight of clothes.

Anthropometric data and 24-hour dietary recalls were entered into the NutriTiming® (NutriTiming LLC, 2011) software. Energy needs were calculated using an age and gender appropriate pediatric BMR. Data for nutritional content of the foods were obtained through NutriTiming®. The nutrient content of food items that were not available on NutriTiming® was entered manually. The subjects' identities were protected through the designation of subject numbers instead of names. Participants were assured that no personal indicators would be mentioned in study reports or data analysis.

Data were analyzed using SPSS 18.0, using $P \leq 0.05$ to determine statistical significance. Descriptive statistics, t-test, and Spearman's correlation were used to assess the relationships between anthropometric values and with-in day energy balance values. A linear regression analysis was performed to determine if any of the energy balance values explained a significant proportion of the variance in predicted body fat percent.

CHAPTER IV

RESULTS

Data were included on a total of 12 children (6 males/6 females). The age range of the subjects was 9-14 years and the mean age was 11.41 ± 1.5 years. The mean weight of the subjects was 48.64 ± 9.75 kg and the mean height was 150.29 ± 10.46 cm. The mean BMI was 20.77 ± 3.16.

Table 1. Descriptive Data

Descriptive Statistics	**Total Subjects N=12 Mean ± SD**	**Min**	**Max**	**Males n=6 Mean ± SD**	**Females n=6 Mean ± SD**
Age (yr)	11.41 ± 1.51	9	14	11 ± 1.67	11.83 ± 1.32
Weight (kg)	48.64 ± 9.75	33.28	68.55	50 ± 12.4	46.96 ± 6.97
Height (cm)	150.29 ± 10.46	125.73	162.56	153.04 ± 7.85	147.52 ± 12.7
BMI	20.77 ± 3.16	15.8	27.60	21.67 ± 4.19	20.67 ± 1.96

Subjects consumed a mean of 1985.25 ± 510.33 kilocalories and expended a mean of 1689.83 ± 351.79 kilocalories. Kilocalories expended (kcal expended) referred to all activities including kilocalories burned while sleeping, performing activities of daily living, and exercise. The children in this study spent the majority of their day in negative energy balance (Hours < 0 EB) at 13.33 ± 6.51 hours, therefore, had a greater anabolic to catabolic ratio 2.57 ± 3.06. The rest of their day was spent in positive energy balance () was 10.67 ± 6.51 hours. At the end of the day, the mean energy balance of all subjects was 294.42 ± 593.34 kilocalories, indicating that despite spending most of their time in a kilocalorie deficit, most subjects were in positive energy balance by the end of the day.

These children received most of their nutrition from carbohydrates (57.50% ± 6.05), thenfat (27.67% ± 5.87) and protein (15.2% ± 55.10). Therefore, as a whole, the group was receiving a balanced distribution of nutrients.

Table 2. Within Day Energy Balance Descriptive Statistics

Variables	Total Subjects N=12 Mean(SD)	Minimum	Maximum
Kcal Consumed	1985.25(510.33)	1374	3227
Kcal Expended	1689.83(351.79)	1163	2231
Kcal consumed/kg	41.93(10.86)	21.20	55.90
Energy Balance	294.42(593.34)	-778	1156
Hours + or- 400kcal	18.00(3.44)	14.00	23.00
Hours>400Kcal	4.50(4.10)	-	10.00
Hours<400Kcal	1.50(2.91)	-	9.00
Ratio of Surplus to deficit	0.58(1.73)	-	6.00
Hours < 0 EB	13.33(6.51)	1	22
Hours > 0 EB	10.67(6.51)	2	23
Ratio Anabolic to Catabolic	2.57(3.06)	0.04	11.00
Highest EB Surplus	738.92(447.16)	73.00	1406.00
Lowest EB Deficit	-364.83(315.38)	-995.00	-35.00
% Kcal from CHO	57.50(6.05)	47.00	70.00
% Kcal from PRO	15.25(5.10)	9.00	26.00
% Kcal from FAT	27.67(5.87)	17.00	40.00

Neither independent group t-test nor Mann-Whitney U-test revealed any significant variances in anthropometric or energy balance values between the genders.

Table 3. Independent Group T-tests comparing males and females on anthropometric and body fat percent values

	Males (n=6) Mean(SD)	Females (n=6) Mean(SD)	Significance (*P*)
Weight (kg)	50.32 (12.41)	46.95 (6.97)	0.575
Height (cm)	153.04 (7.85)	147.52 (12.70)	0.388
BMI	21.27 (4.19)	20.27 (1.96)	0.608
Body Fat %	24.31 (6.75)	26.35 (1.99)	0.496

Neither independent group t-test nor Mann-Whitney U-test revealed any significant differences in energy balance values between the genders, with the exception of BMR

(*P*=0.025).

Table 4. Independent Group T-tests comparing males and females on energy balance values

	Males (n=6) Mean(SD)	**Females (n=6) Mean(SD)**	**Significance (*P*)**
BMR	1073.21(203.33)	1130.77(255.14)	**0.025***
Kcal consumed	1848.00(334.02)	2120.50(645.35)	0.380
Kcal expended	1660.17(403.03)	1719.50(328.21)	0.785
Kcal consumed/kg	38.85 (11.32)	45.00 (10.42)	0.351
Energy Balance	187.83 (604.65)	401.00 (617.82)	0.559
Hours + or- 400kcal	19.83(3.31)	19.67(3.43)	0.258
Hours>400Kcal	4.50(4.96)	4.50(3.51)	1.000
Hours<400Kcal	2.67(3.88)	0.33(0.52)	0.175
Ratio of Surplus to deficit	-	1.67(2.40)	0.262
Hours < 0 EB	10.66 (8.52)	10.66 (4.54)	1.000
Hours > 0 EB	13.33 (8.52)	13.33 (4.54)	1.000
Ratio Anabolic to Catabolic	3.45(4.14)	1.69(1.30)	0.344
Highest EB Surplus	599.50(442.13)	878.33(444.72)	0.302

Neither independent group t-test nor Mann-Whitney U-test revealed any significant variances in the distribution of energy substrates between the genders. Therefore, both males and females experienced the same variation across the majority of data included in this study.

Table 5. Independent group t-tests comparing males and females on distribution of energy substrates

	Males (n=6) Mean(SD)	**Females (n=6) Mean(SD)**	**Significance (*P*)**
% Kcal from CHO	57.5(7.61)	57.5(4.76)	1.000
% Kcal from PRO	17.17(4.58)	13.33(5.24)	0.207
% Kcal from FAT	25.67(6.06)	29.67(5.43)	0.256

When examining the effects of within day energy balance on body composition, 3 measures were targeted, body fat percentage (body fat %), fat free mass, and trunk body

fat percentage (trunk BF %). These measures were tested for correlations with total daily kilocalories consumed (kcal consumed), kcal expended, end of day energy balance, kilocalories consumed per kilogram of body weight (kcal consumed/kg), hours spent in an energy balance above or below 400 kcal (hours + or- 400kcal), hours spent in an energy balance above 400 kcal (hours>400Kcal), hours spent in an energy balance below 400 kcal (hours<400Kcal), ratio of energy balance surplus to energy balance deficit, ratio of energy balance surplus to deficit, hours spent in an a negative energy balance (Hours < 0 EB), hours spent in an a positive energy balance (hours > 0 EB), ratio of hours spent in an anabolic state to hours spent in a catabolic state (anabolic to catabolic), highest energy balance surplus (highest EB surplus), lowest energy balance deficit (lowest EB deficit), percent of kilocalories from carbohydrates (% kcal from CHO), percent of kilocalories from protein (% kcal from PRO), and percent of kilocalories fat (% kcal from FAT). Data were analyzed for total group then for males and females separately.

In the total population the statistically significant correlations with body fat % were kcal expended (0.706; P=0.010), hours<400Kcal (0.732; P=0.007), hours<EB (0.914; P<0.001), hours>EB (-0.914; P<0.001), ratio anabolic to catabolic (-0.914; P<0.001), and lowest EB deficit (-0.832; P=0.001). These data indicate that children who are spending most of their day in energy balance deficits have a tendency toward greater adiposity than those children who did not. Therefore, we were able to reject the first null hypothesis. However, these data also show a negative correlation between time spent in an energy balance surplus and body fat %. Therefore, we were not able to reject our second null hypothesis. Also important to note is that total daily intake did not have a significant correlation to body fat %. This finding further supports the belief that end of

day energy balance is not a reliable predictor of adiposity. Finally, as one would expect, those spending most of their time in an anabolic state had a tendency toward lower body fat percentages.

The statistically significant correlations with body fat % in the male population were: kcal expended (0.866; *P*=0.019), energy balance (-0.829; *P*=0.042), kcal consumed/kg (-0.829; *P*=0.042), hours<400kcal (0.941; *P*=0.005), lowest EB deficit (-0.812; *P*=0.050), and % kcal from FAT (0.829; *P*=0.042). The results of these data indicate that the boys who are the most active spend most of their day in energy balance deficits and also have higher body fat %. Also from this data it would appear that the children consuming more kcal/kg and spending more time in energy balance surpluses are leaner. These data may have been a consequence of inaccurate dietary recalls. Finally, in the males, a higher % of dietary fat intake was associated with a greater body fat %.

The only statistically significant correlation with body fat % for the females was with % kcal from CHO. According to this statistic, a higher proportion of dietary carbohydrate intake was the greatest predictor of increased adiposity.

Table 6. Spearman Correlation coefficients (ρ) showing the association between body fat % and within day energy balance statistics of all subjects (n=12)

	Total ---------- ρ (***P***)	**Male** ---------- ρ (***P***)	**Female** ---------- ρ (***P***)
Body Fat % Kcal Consumed	-0.028(0.931)	-0.429(0.397)	0.657(0.156)
Body Fat % Kcal Expended	**0.706(0.010)***	**0.866(0.019)***	0.657(0.156)
Body Fat % Energy Balance	-0.322(0.308)	**-0.829(0.042)***	0.543(0.266)
Body Fat % Kcal consumed/kg	-0.231(0.471)	**-0.829(0.042)***	0.657(0.156)
Body Fat % Hours + or- 400kcal	0.085(0.794)	0.145(0.784)	-0.429(0.397)
Body Fat % Hours>400Kcal	-0.552(0.063)	-0.698(0.123)	0.353(0.492)
Body Fat % Hours<400Kcal	**0.732(0.007)***	**0.941(0.005)***	0.414(0.414)
Body Fat % Ratio of Surplus to deficit	0.274(0.389)	-	0.507(0.305)
Body Fat % Hours < 0 EB	**0.914(<0.001)***	**1.00**	0.580(0.228)
Body Fat % Hours > 0 EB	**-0.914(<0.001)***	**-1.00**	-0.580(0.228)
Body Fat % Ratio Anabolic to Catabolic	**-0.914(<0.001)***	**-1.00**	-0.580(0.228)
Body Fat % Highest EB Surplus	-0.350(0.265)	**-0.829(0.042)***	0.314(0.544)
Body Fat % Lowest EB Deficit	**-0.832(0.001)***	**-1.00**	-0.314(0.544)
Body Fat % % Kcal from CHO	-0.025(0.939)	-0.174(0.742)	**0.812(0.050)***
Body Fat % % Kcal from PRO	-0.221(0.490)	-0.319(0.538)	-0.88(0.868)
Body Fat % % Kcal from FAT	0.504(0.094)	**0.829(0.042)***	-0.273(0.600)

* Statistically significant

Fat free mass in the total population correlated significantly with the following statistics: kcal expended (0.834; *P*=0.001), hours<400kcal (0.591; *P*=0.043), Hours < 0 EB (0.883; *P*<0.001), hours > 0 EB (-0.883; *P*<0.001), and lowest EB deficit (-0.599;

$P \leq 0.040$). According to these statistics the more active children had larger amounts of fat free mass. Also, the children who spent more time in an energy balance surplus, tended to have lower fat free mass and the children who spent the most time in an energy balance deficit had more fat free mass.

In the males and females separately, fat free mass significantly correlated with kcal expended [(0.829; P=0.042) (0.943; P=0.005), respectively]. In other words, as we concluded earlier, the more active children have more fat free mass. In males, the macronutrient significantly associated with fat free mass was protein (-0.812; P=0.050) and in females it was carbohydrates (0.812; P=0.050). Increased protein intake in males lead to less fat free mass and in females increased carbohydrates lead to greater fat free mass.

Table 7. Spearman Correlation coefficients (ρ) showing the association between fat free mass and within day energy balance statistics of all subjects (n=12)

	Total ---------- ρ (***P***)	**Male** ---------- ρ (***P***)	**Female** ---------- ρ (***P***)
Fat Free Mass Kcal Consumed	0.161(0.617)	-0.029(0.957)	0.371(0.468)
Fat Free Mass Kcal Expended	**0.834(0.001)***	**0.829(0.042)***	**0.943(0.005)***
Fat Free Mass Energy Balance	-0.109(0.737)	-0.314(0.544)	0.257(0.623)
Fat Free Mass Kcal consumed/kg	-0.322(0.307)	-0.714(0.111)	0.371(0.468)
Fat Free Mass Hours + or- 400kcal	0.206(0.520)	-0.058(0.913)	0.371(0.468)
Fat Free Mass Hours>400Kcal	-0.541(0.069)	-0.395(0.439)	-0.441(0.381)
Fat Free Mass Hours<400Kcal	**0.591(0.043)***	0.577(0.231)	0.621(0.188)
Fat Free Mass Ratio of Surplus to deficit	0.215(0.501)	-	0.676(0.140)
Fat Free Mass Hours < 0 EB	**0.883(<0.001)***	0.600(0.208)	0.667(0.148)
Fat Free Mass Hours > 0 EB	**-0.883(<0.001)***	-0.600(0.208)	-0.667(0.148)
Fat Free Mass Ratio Anabolic to Catabolic	-0.509(0.091)	-0.600(0.208)	0.029(0.957)
Fat Free Mass Highest EB Surplus	-0.312(0.324)	-0.314(0.544)	0.771(0.072)
Fat Free Mass Lowest EB Deficit	**-0.599(0.040)***	-0.600(0.208)	-0.086(0.872)
Fat Free Mass % Kcal from CHO	0.550(0.064)	0.551(0.257)	**0.812(0.050)***
Fat Free Mass % Kcal from PRO	-0.291(0.359)	**-0.812(0.050)***	-0.088(0.868)
Fat Free Mass % Kcal from FAT	-0.148(0.645)	0.086(0.872)	0.030(0.954)

* Statistically significant

In the total population, trunk BF % correlated significantly with kcal expended (0.741; *P*=0.006), hours>400kcal (-0.704; *P*=0.011), hours<400kcal (0.881; *P*<0.001),

Hours < 0 EB (0.890; P<0.001), hours > 0 EB (-0.890; P<0.001), ratio anabolic to catabolic

(-0.890; P<0.001), and lowest EB deficit (-0.937; P<0.001). These results are similar to the body fat % statistics, which seem to suggest energy deficits put one at greater risk of adiposity. Therefore, it is no surprise that one would also tend to have increased central adiposity.

In the males, hours<400kcal (0.880; P=0.021), hours<EB (-0.943; P=0.005), hours>EB (-0.943; P=0.005), ratio anabolic to catabolic (-0.943; P=0.005), and % kcal from PRO (0.886; P=0.019). Again, these correlations are similar to the body fat % statistics, with the exception of % kcal from PRO. Though not significant, % kcal from PRO in the body fat % correlation statistics indicated a negative trend for total body fat (see table 4), whereas, in regards to trunk BF % the correlation was positive.

In the females hours<400kcal (0.845(0.034) and ratio of surplus to deficit (0.845; P=0.034) were significantly correlated to trunk BF %. These statistics were not seen in the previous two comparisons, indicating, that caloric surpluses and time spent in energy balance deficits may not be a good indicator of total body fat % in females, but instead on fat distribution. In this case, a greater energy balance surplus and time spent in an energy balance deficit indicates a trend toward greater central adiposity.

Table 8. Spearman Correlation coefficients (ρ) showing the association between trunk body fat % and within day energy balance statistics of all subjects (n=12)

	Total ---------- ρ (*P*)	**Male** ---------- ρ (*P*)	**Female** ---------- ρ (*P*)
Trunk BF % Kcal Consumed	-0.077(0.812)	-0.314(0.544)	0.486(0.329)
Trunk BF % Kcal Expended	**0.741(0.006)***	0.771(0.072)	0.657(0.156)
Trunk BF % Energy Balance	-0.378(0.226)	-0.771(0.072)	0.257(0.623)
Trunk BF % Kcal consumed/kg	-0.322(0.308)	-0.771(0.072)	0.486(0.329)
Trunk BF % Hours + or- 400kcal	0.243(0.447)	0.290(0.577)	-0.257(0.623)
Trunk BF % Hours>400Kcal	**-0.704(0.011)***	-0.698(0.123)	0.088(0.868)
Trunk BF % Hours<400Kcal	**0.881(≤0.001)***	**0.880(0.021)***	**0.828(0.042)***
Trunk BF % Ratio of Surplus to deficit	0.333(0.290)	-	**0.845(0.034)***
Trunk BF % Hours < 0 EB	**0.890(≤0.001)***	**0.943(0.005)***	0.580(0.228)
Trunk BF % Hours > 0 EB	**-0.890(≤0.001)***	**-0.943(0.005)***	-0.580(0.228)
Trunk BF % Ratio Anabolic to Catabolic	**-0.890(≤0.001)***	**-0.943(0.005)***	-0.580(0.228)
Trunk BF % Highest EB Surplus	-0.441(0.152)	-0.771(0.072)	0.257(0.623)
Trunk BF % Lowest EB Deficit	**-0.937(≤0.001)***	**-0.943(0.005)***	-0.600(0.208)
Trunk BF % % Kcal from CHO	-0.082(0.800)	-0.319(0.538)	0.551(0.257)
Trunk BF % % Kcal from PRO	-0.039(0.904)	**0.886(0.019)***	0.117(0.738)
Trunk BF % % Kcal from FAT	0.434(0.159)	0.293(0.573)	-0.516(0.295)

* Statistically significant

Also from these data, we were able to create two equations to predict body fat % using either an 'enter' or 'stepwise' linear regression procedure. Using the 'enter' procedure to include all variables selected to explain variance in body fat percent, included the following independent variables: age, height, weight, hours ± 400,

hours<400kcal, Hours < 0 EB, highest EB surplus, lowest EB deficit and excluding hours>400kcal and hours>EB. The unstandardized β coefficients were then used to formulate the following equation:

Body fat% = age(-0.334) + height (-0.110) + weight (0.123) + hours ± 400 (-0.119) + hours<400kcal(-1.247) + Hours < 0 EB (0.397) + highest EB surplus (0.003) + lowest EB deficit (-0.017) + 31.227

This equation predicted body fat % with a SEE= 2.9, R^2=0.903 and an adjusted R^2=0.645. However, the *P* value was not significant, so the process was repeated using a different independent variable.

Table 9. Enter procedure linear regression unstandardized β coefficients with hours>400kcal and hours>EB excluded due to collinearity, and with body fat % as the dependent variable

	β	*P*
Constant	31.225	0.207
Age	-0.334	0.829
Height	-0.344	0.415
Weight	0.123	0.689
Hours ± 400kcal	-0.119	0.898
Hours<400kcal	-1.247	0.610
Hours<EB	0.397	0.502
Highest EB Surplus	0.003	0.603
Lowest EB Deficit	-0.017	0.576

The second equation was determine by was determined using a stepwise procedure, whereby the procedure kept only the variables that explained a significant proportion of the variance in body fat percent, and included the independent variable hours>EB and excluding: age, height, weight, hours ± 400, hours<400kcal, Hours < 0 EB, highest EB surplus, lowest EB deficit, and hours>400kcal. The unstandardized β coefficients were

then used to formulate the following equation:

$$\text{Body fat \%} = \quad \text{hours} > 0\ \text{EB}\ (0.659) + 34.114$$

This equation predicted body fat % with a SEE= 2.407, R^2=0.777, an adjusted R^2=0.755 with a P<0.001.

Table 10. Stepwise linear regression unstandardized β coefficients with age, height, weight, hours ± 400, hours<400kcal, hours > 0 EB, highest EB surplus, lowest EB deficit and hours>400kcal were excluded and body fat % as the dependent variable.

	β	*P*
Constant	34.114	<0.001
Hours > 0 EB	-0.659	<0.001

Summary of Significant Findings

All in all, more time spent in energy balance deficits in this population indicate a tendency toward increased total and trunk BF %. More time spent in energy balance surpluses indicate the opposite, a tendency toward less total and trunk BF %, except in the females. Females who had a greater ratio of energy surpluses to deficits were more prone to increased trunk BF %. Fat free mass increased with a greater increase in kcal expended and decreased with hours spent in kcal surpluses. In regards to macronutrient intake, a higher % kcal from FAT in males indicates an increase in body fat % and in females it is a higher % kcal from CHO. Also, as % kcal from PRO increases in males fat free mass decreases, whereas in reference to trunk BF %, % kcal from PRO increases along with trunk BF %.

CHAPTER V

DISCUSSION

The results of this study revealed that body fat in children increased with the greater amount of hours spent in negative energy balance. These findings were in accordance with our first hypothesis and are supported by the research of Metzner et al. (1997). In their study, 24-hour dietary recalls of 2,000 men and women aged 35-69 were gathered to assess the relationship between eating frequency and adiposity. After adjusting for food level intakes, their analysis revealed an inverse relationship between the number of meals consumed and adiposity (54).

When the results of males and females were examined separately, the correlation between hours in an energy deficit and body fat percentage remained significant only for the males. This discrepancy may have been a consequence of the smaller sample size, or as Drummond et al. (1998) discovered, caused by a difference in the manner males and females moderate caloric intake. In their study, 48 men and 47 women were recruited and followed for a period of 7 days (55). During these 7 days, dietary intake and timing along with all physical activity were recorded. At the end of the study they found a significant relationship between males eating frequency and weight (inversely related), but not the women (55). What they discovered is that the men who ate more frequency tended to consume fewer kilocalories per meal, whereas the women did not. In other words, the

women who ate fewer meals ate approximately the same amount of calories per meal than the women who ate more frequently and therefore had higher daily intakes (55). Ifwomen tend to eat the same amount per meal no matter what the frequency, it would be logical to assume that those expending more kilocalories would be leaner.

Duval et al. (1998) also investigated the effects of eating frequency on body composition in women. More specifically, they were measuring body fat percentages (56). The researchers of his study did not find a significant correlation between body fat percentage and eating frequency. Furthermore, they found a positive correlation between eating frequency and caloric intake. What they concluded was that physical activity energy expenditure was a greater predictor of lower body fat percentages than eating frequency (56). Again, these results are to be expected in women if eating frequency significantly increases caloric intake.

While there was no significant correlation between hours<400Kcal and body fat % in females, there was a significant correlation between hours<400Kcal and trunk BF %. This finding may explain why Berkey et al. (2003) found no difference between the adiposity risk of children who skipped breakfast and those who did not. As a matter of fact, those who skipped breakfast actually reported losing weight. However, WC was not assessed in this study (42). Smith et al. (2010) and Isacco et al. (2010), on the other hand, observed a significant increase in WC ($P<0.008$ and $P<0.001$, respectively) in those who habitually skipped breakfast compared to those that did not (7,43). Therefore, from the results of our study and previous epidemiological evidence, it is important to include WC (or other measures of central adiposity) in any study that assesses the effects of energy balance on adiposity.

Another important finding of our study, was that there was no significant relationship between total calories consumed and body fat percentage in the males. This lack of correlation may be explained by many factors. For instance, Keski Rahkonen et al. (2003) discovered that skipping meals accompanies and myriad of other unhealthy behaviors, including a tendency toward decreased physical activity. Exercise may neccesitate more frequent eating occasions due to increased hunger and energy needs (56). Therefore, an increased energy intake would not lead to greater adiposty because the body is expending the extra intake.

In our study, energy expended positively correlated with body fat percentage in the males. One explanation for this couterintuitive correlation, is the fact that the more mass one has, the more energy they expend. Ekkakis and Lind (2005) explained this relationship in their study of 16 overweight (BMI $\geq$ 25kg/m2) and 9 normal-weight women (BMI < 25kg/m2) between the ages of 35 and 53 years. All women reported having been physically inactive for at least 1 year before entering the study (58). All women were underwent 3 sessions with at least 48 hours between. The first session involved assessing peak oxygen uptake and peak heart rate (58). The second session involved a 20-min on a treadmill at a self-selected speed. The third session involved another 20-min treadmill exercise during where the speed was imposed by the experimenters and adjusted to be 10% higher than the self-selected level (58). Even though, self-selected speeds and imposed speeds did not differ between the groups, the overweight group had to utilize a higher percentage of their peak aerobic capacity to perform the same task. Furthermore, the overweight group did report higher rates of perceived exertion (58). Because exercise intensity was self-reported it is also possible

that the calculated energy expenditures were skewed by the fact that heavier individuals tend to perceive greater intensity, thus greater energy expenditure with their activity than the leaner individuals.

Our second hypothesis dealt with body fat percentage as it related to energy balance surpluses. More specifically, we believed that more time spent in energy balance surpluses would lead to greater adiposity. Our findings indicated exactly the opposite. To date there is no research that observes the effects of within day energy balance surpluses in children, therefore it is difficult to speculate on these results. It is possible that children, due to their increased energy needs, are less sensitive to energy balance surpluses throughout the day than adults. Another possibilty is the underreporting of caloric intake in the more overweight subjects, which could have made it appear as though the leaner subjects ate proportionately more. Singh et al. (2011) observed this tendency to overreport caloric intake in their study of 34 overweight adolescents (14 girls; 20 boys), aged 12–15 years. Subjects took part in a summer research camp and resided in campus residence halls, receiving energy-controlled diets that met their estimated individualized energy needs (58). They were also asked to record their intakes for 6 days of the camp and for 3 days between the 2 sessions of the camp. At the end of the camp, they were asked submit their self-recorded dietary recalls for researchers to compare with actual intakes. After examination of recalls, reseachers noted a 35% intake bias in their subjects (59). Furthermore, the intake biases increased as weight increased, indicating that the tendency to underreport intakes were greater in the more overweight subjects (r^2=0.15, P<0.05). Also, the higher the intake of dietary fat, the more inaccurate the reporting (r^2=0.59, P<0.05) (59).

Another important finding of this research is the lack of significance between end of day energy balance and adiposity. These findings contradict what most of the scientific community hold to be true that weight gain is a direct result of excess energy intake alone. The CDC states that weight maintenance is achieved through 'balancing the number of calories you consume with the number of calories your body uses or "burns off"'(60). While this may be true to a certain extent, our research suggests that within day energy balance is also important. Bachman et al. (2011) analyzed the diets of 257 women aged 21-65 years (81 overweight, 80 normal weight, and 96 normal weight who were once overweight or obese), to determine a relationship between eating frequency and weight maintenance (61). This study showed an inverse relationship between eating frequency and BMI. It also showed that those who ate more frequently consumed more calories and tended to be more physically active. This study did not report on positive or negative energy balance statistics, but the finding of increased caloric intake negatively correlating with BMI does support the need to revisit current belief systems (61). In other words, there is sufficient evidence, through our research and the research cited in our study, to challenge the "calories in must equal calories out" approach to weight loss and maintenance.

A weakness of this study was the small sample size (n=12). Also, because it was a covenience sample, a wide variety of sample demographics could not be guarenteed. A strength of the this study was the original nature of the data. To our knowledge there has not been a study of within day energy balance in this population. That being said, further research needs to be done to further validate the significance of our findings.

Conclusions

The incidence of childhood obesity is on the rise. While erratic daily energy balance is only one of the many factors that may come into play in this epidemic, it is important that it is not ignored. Fewer energy deficits are associated with more physical activity, better dietary choices and habits, and as this study has shown, less adiposity. More research needs to be done to more clearly define the mechanisms behind these connections, however the strong epidemiological evidence of more favorable health outcomes should be enough to necessitate greater public awareness.

IV. REFERENCES

1. Statistics National Center for Health Studies. Special Feature on Medical Technology, United States, 2009: 2010:26.
2. Koletzko B. Meal Patterns and Frequencies : Do They Affect Body Weight in Children and Adolescents? *Critical Reviews in Food Science and Nutrition.* 2010;50:100-105.
3. Dongen M, de Graaf C, Siebelink E, Kok F, Dongen V-v. Hidden Fat Facilitates Passive Overconsumption. *The Journal of Nutrition.* 2008;139:394-399.
4. Giovannini M, Agostoni C, Shamir R. Symposium overview: Do we all eat breakfast and is it important? *Critical Reviews In Food Science And Nutrition.* 2010;50:97-99.
5. Heber D. An integrative view of obesity. *Am J Clin Nutr.* 2010;91:280S-283.
6. Moreno L, Rodriguez G, Fleta J, Bueno-Lozano M, Lazaro A, Bueno G. Trends of dietary habits in adolescents. *Critical Reviews in Food Science and Nutrition.* 2010;50:106-112.
7. Smith K, Gall S, McNaughton S, Blizzard L, Dwyer T. Skipping breakfast longitudinal associations with cardiometabolic risk factors in the Childhood Determinants of Adult Health Study. *The American journal of clinical nutrition.* 2010;92:1316-1325.
8. Bakhotmah B. The puzzle of self-reported weight gain in a month of fasting (Ramadan) among a cohort of Saudi families in Jeddah, Western Saudi Arabia. *Nutrition Journal.* 2011;10:84.
9. Farshchi HR, Taylor MA, Macdonald IA. Deleterious effects of omitting breakfast on insulin sensitivity and fasting lipid profiles in healthy lean women. *The American journal of clinical nutrition.* 2005;81:388-396.
10. Umemura U, Ishimori M, Kobayashi T, Tamura Y, Koike K, Shimamoto T, Iso H. Possible effects of diets on serum lipids, fatty acids and blood pressure levels in male and female Japanese university students. *Environmental Health and Preventive Medicine.* 2005;10:42-47.
11. Kollias A, Antonodimitrakis P, Grammatikos E, Chatziantonakis N, Grammatikos EE, Stergiou GS. Trends in high blood pressure prevalence in Greek adolescents. *Journal of human hypertension.* 2009;23:385-390.
12. Stote KS, Baer DJ, Spears K, Paul DR, Harris GK, Rumpler WV, Strycula P, Najjar SS, Ferrucci L, Ingram DK, Longo DL, Mattson MP. A controlled trial of reduced meal frequency without caloric restriction in healthy, normal-weight, middle-aged adults. *American Journal of Clinical Nutrition.* 2007;85:981-988.
13. Alexander K, Ventura E, Spruijt Metz D, Weigensberg M, Goran M. Association of Breakfast Skipping With Visceral Fat and Insulin Indices in Overweight Latino Youth. *Obesity.* 2009;17:1528-1533.

14. Farshchi HR, Taylor MA, Macdonald IA. Regular meal frequency creates more appropriate insulin sensitivity and lipid profiles compared with irregular meal frequency in healthy lean women. *European Journal of Clinical Nutrition.* 2004;58:1071-1077.
15. Anderwald C, Brabant G, Bernroider E, Horn Rd, Brehm A, Waldhusl W, Roden M. Insulin-Dependent Modulation of Plasma Ghrelin and Leptin Concentrations Is Less Pronounced in Type 2 Diabetic Patients. *Diabetes.* 2003;52:1792-1798.
16. Solomon TPJ, Chambers E, Jeukendrup A, Toogood A, Blannin A. The effect of feeding frequency on insulin and ghrelin responses in human subjects. *British Journal of Nutrition.* 2008;100:810-819.
17. JeQuier E. Leptin Signaling, Adiposity, and Energy Balance. *Annals of the New York Academy of Sciences.* 2002;967:379-388.
18. Kolaczynski JW, Considine RV, Ohannesian J, Marco C, Opentanova I, Nyce MR, Myint M, Caro JF. Responses of leptin to short-term fasting and refeeding in humans: a link with ketogenesis but not ketones themselves. *Diabetes.* 1996;45:1511-1515.
19. Nicklas TA, Baranowski T, Cullen KW, Berenson G. Eating Patterns, Dietary Quality and Obesity. *Journal of the American College of Nutrition.* 2001;20:599-608.
20. Bibi H, Shoseyov D, Feigenbaum D, Genis M, Friger M, Peled R, Sharff S. The relationship between asthma and obesity in children: is it real or a case of over diagnosis? *The Journal of Asthma.* 2004;41:403-410.
21. Spathopoulos D, Paraskakis E, Trypsianis G, Tsalkidis A, Arvanitidou V, Emporiadou M, Bouros D, Chatzimichael A. The effect of obesity on pulmonary lung function of school aged children in Greece. *Pediatric Pulmonology.* 2009;44:273-280.
22. Centers for Disease Control and Prevention. Diabetes Public Health Resource. Web site. http://www.cdc.gov/diabetes/projects/diab_children.htm. Accessed October 3, 2011.
23. Freedman DS, Dietz WH, Srinivasan SR, Berenson GS. The Relation of Overweight to Cardiovascular Risk Factors Among Children and Adolescents: The Bogalusa Heart Study. *Pediatrics.* 1999;103:1175.
24. Vajro P, Fontanella A, Perna C, Orso G, Tedesco M, De Vincenzo A. Persistent hyperaminotransferasemia resolving after weight reduction in obese children. *The Journal of Pediatrics.* 1994;125:239-241.
25. Deshmukh-Taskar P, Nicklas T, O'Neil C, Keast D, Radcliffe J, Cho S. The relationship of breakfast skipping and type of breakfast consumption with nutrient intake and weight status in children and adolescents: the National Health and Nutrition Examination Survey 1999-2006. *Journal of the American Dietetic Association.* 2010;110:869-878.
26. Pasman WJ, Blokdijk VM, Bertina FM, Hopman WPM, Hendriks HFJ. Effect of two breakfasts, different in carbohydrate composition, on hunger and satiety and mood in healthy men. *International Journal of Obesity & Related Metabolic Disorders.* 2003;27:663.

27. Melhorn S, Krause E, Scott K, Mooney M, Johnson J, Woods S, Sakai R. Acute exposure to a high-fat diet alters meal patterns and body composition. *Physiology & Behavior.* 2010;99:33-39.
28. Sierra-Johnson J, Unden A-L, Linestrand M, Rosell M, Sjogren P, Kolak M, De Faire U, Fisher RM, Hellenius M-L. Eating Meals Irregularly: A Novel Environmental Risk Factor for the Metabolic Syndrome. *Obesity.* 2008;16:1302-1307.
29. Arble DM, Bass J, Laposky AD, Vitaterna MH, Turek FW. Circadian Timing of Food Intake Contributes to Weight Gain. *Obesity.* 2009;17:2100-2102.
30. Woodruff SJ, Hanning RM, Lambraki I, Storey KE, McCargar L. Healthy Eating Index-C is compromised among adolescents with body weight concerns, weight loss dieting, and meal skipping. *Body Image.* 2008;5:404-408.
31. Jennings A, Welch A, van Sluijs EMF, Griffin S, Cassidy A. Diet quality is independently associated with weight status in children aged 9-10 years. *The Journal of Nutrition.* 2011;141:453-459.
32. Guerrieri R, Nederkoorn C, Schrooten M, Martijn C, Jansen A. Inducing impulsivity leads high and low restrained eaters into overeating, whereas current dieters stick to their diet. *Appetite.* 2009;53:93-100.
33. Markowitz J, Butryn M, Lowe M. Perceived deprivation, restrained eating and susceptibility to weight gain. *Appetite.* 2008;51:720-722.
34. Niemeier HM, Raynor HA, Lloyd-Richardson EE, Rogers ML, Wing RR. Fast Food Consumption and Breakfast Skipping: Predictors of Weight Gain from Adolescence to Adulthood in a Nationally Representative Sample. *Journal of Adolescent Health.* 2006;39:842-849.
35. Farshchi HR, Taylor MA, Macdonald IA. Beneficial metabolic effects of regular meal frequency on dietary thermogenesis, insulin sensitivity, and fasting lipid profiles in healthy obese women. *American Journal of Clinical Nutrition.* 2005;81:16-24.
36. Stein DT, Esser V, Stevenson BE, Lane KE, Whiteside JH, Daniels MB, Chen S, McGarry JD. Essentiality of circulating fatty acids for glucose-stimulated insulin secretion in the fasted rat. *The Journal of Clinical Investigation.* 1996;97:2728-2735.
37. Zawalich WS, Zawalich KC. Glucose-induced insulin secretion from islets of fasted rats: modulation by alternate fuel and neurohumoral agonists. *Journal of Endocrinology.* 2000;166:111-120.
38. Schwarz N RR, La Bounty P, Shelmadine B, and Bowden R. A review of weight control strategies and their effects on the regulation of hormonal balance. *Journal of Nutrition and Metabolism.*2011:1-15.
39. De Castro J. The time of day of food intake influences overall intake in Humans. *The Journal of Nutrition.* 2004;134:104-111.
40. Astbury N, Taylor M, Macdonald I. Breakfast consumption affects appetite, energy intake, and the metabolic and endocrine responses to foods consumed later in the day in male habitual breakfast eaters. *The Journal of Nutrition.* 2011;141:1381-1389.
41. Mattson MP. Energy intake, meal frequency, and health: a neurobiological perspective. *Annual Review of Nutrition.* 2005;25:237-260.

42. Franko DL, Striegel-Moore RH, Thompson D, Affenito SG, Schreiber GB, Daniels SR, Crawford PB. The relationship between meal frequency and body mass index in black and white adolescent girls: more is less. *International Journal of Obesity.* 2008;32:23-29.
43. Berkey CS, Rockett HRH, Gillman MW, Field AE, Colditz GA. Longitudinal study of skipping breakfast and weight change in adolescents. *International Journal of Obesity.* 2003;27:1258-1266.
44. Isacco L, Lazaar N, Ratel S, Thivel D, Aucouturier J. The impact of eating habits on anthropometric characteristics in French primary school children. *Child Care Health and Development.* 2010;36:835-842.
45. Edelstein SL, Barrett Connor EL, Wingard DL, Cohn BA. Increased meal frequency associated with decreased cholesterol concentrations; Rancho Bernardo, CA, 1984-1987. *The American Journal of Clinical Nutrition.* 1992;55:664-669.
46. Maislos M, Khamaysi N, Assali A, Abou Rabiah Y, Zvili I, Shany S. Marked increase in plasma high-density-lipoprotein cholesterol after prolonged fasting during Ramadan. *The American Journal of Clinical Nutrition.* 1993;57:640-642.
47. U.S Department of Health and Human Services. *National Diabetes Information Clearinghouse* Web site. http://diabetes.niddk.nih.gov/DM/PUBS/statistics/ - ddY20. Accessed October 3, 2011.
48. Mathur P, Das MK, Arora NK. Non-alcoholic fatty liver disease and childhood obesity. *Indian Journal of Pediatrics.* 2007;74:401-407.
49. Pacifico L, Nobili V, Anania C, Verdecchia P, Chiesa C. Pediatric nonalcoholic fatty liver disease, metabolic syndrome and cardiovascular risk. *World Journal of Gastroenterology.* 2011;17:3082-3091.
50. Hamaguchi M, Kojima T, Takeda N, Nagata C, Takeda J, Sarui H, Kawahito Y, Yoshida N, Suetsugu A, Kato T, Okuda J, Ida K, Yoshikawa T. Nonalcoholic fatty liver disease is a novel predictor of cardiovascular disease. *World Journal of Gastroenterology.* 2007;13:1579-1584.
51. Chiu T, Chen C, Chen S, Soon C, Chen J. Indicators associated with coronary atherosclerosis in metabolic syndrome. *Clinica Chimica Acta.*
52. Steinberger J, Daniels S, Eckel R, Hayman L, Lustig R, McCrindle B, Mietus Snyder M. Progress and challenges in metabolic syndrome in children and adolescents: a scientific statement from the American Heart Association Atherosclerosis, Hypertension, and Obesity in the Young Committee of the Council on Cardiovascular Disease in the Young; Council on Cardiovascular Nursing; and Council on Nutrition, Physical Activity, and Metabolism. *Circulation.* 2009;119:628-647.
53. Stephanie A S. Obesity and asthma: cause for concern. *Current Opinion in Pharmacology.* 2006;6:230-236.
54. Metzner H, Lamphiear D, Wheeler N, Larkin F. The relationship between frequency of eating and adiposity in adult men and women in the Tecumseh Community Health Study. *The American Journal of Clinical Nutrition.* 1977;30:712-715.
55. Drummond NC, Cursiter MC, Kirk TR. Evidence that eating frequency is inversely related to body weight status in male, but not female, non-obese adults

reporting valid dietary intakes. *International Journal of Obesity.* 1998;22:105 - 112.

56. Duval K, Strychar I, Cyr M, Prud'homme D, Rabasa-Lhoret R, Doucet V. Physical activity is a confounding factor of the relation between eating frequency and body composition. *The American Journal of Clinical Nutrition.* 2008;88:1200-1205.
57. Keski Rahkonen A, Kaprio J, Rissanen A, Virkkunen M, Rose RJ. Breakfast skipping and health-compromising behaviors in adolescents and adults. *European Journal of Clinical Nutrition.* 2003;57:842-853.
58. Ekkekakis P, Lind E. Exercise does not feel the same when you are overweight: the impact of self-selected and imposed intensity on affect and exertion. *International Journal of Obesity.* 2005;30:652-660.
59. Singh R, Martin B, Hickey Y, Teegarden D, Campbell W, Craig B, Schoeller D, Kerr D, Weaver C. Comparison of self-reported, measured, metabolizable energy intake with total energy expenditure in overweight teens. *The American Journal of Clinical Nutrition.* 2009;89:1744-1750.
60. Centers for Disease Control and Prevention. Overweight and Obesity. *Causes and Consequences.* http://www.cdc.gov/obesity/causes/index.html. Accessed November 29, 2011.
61. Bachman J, Phelan S, Wing R, Raynor H. Eating frequency is higher in weight loss maintainers and normal-weight individuals than in overweight individuals. *Journal of the American Dietetic Association.* 2011;111:1730-1734.

APPENDIX A

Georgia State University
Department of Health and Human Sciences
Parental Permission Form

Title: The Effect Of Meal Frequency And Energy Balance Fluctuations On Body Composition In Children And Adolescents

Principal Investigator: Dan Benardot, PhD, RD, LD, FACSM
Co-Principal Investigator: Anita Nucci, PhD, RD, LD
Student Principle Investigator: Laura Worsham

I. Purpose:

Your child has been invited to participate in a research study. The purpose of this study is to learn how regular meals and activity affect energy levels, weight and body fat. Your child is invited to join because he/she is between 8 and 16 years old. A total of 30 participants will be recruited for this study. The study will take approximately 1 hour.

II. Procedures:

If you and your child agree to participate he/she will be asked to complete diet and activity survey. You may assist with information when necessary. Next he/she will complete a 10-question questionnaire on dietary habits. Then he/she will have his/her height, weight, and body composition taken. This survey will track all food and beverages consumed. It will also track all activities performed within 24-hours. He/she will be asked to supply the information to the best of his/her abilities. Information will be collected during a 1-hour visit. The research will take place at Titus Sports Academy at Chastain Park in Atlanta, GA.

III. Risks:

In this study, your child will not have any more risks than he/she would in a normal day of life.

IV. Benefits:

Participation in this study may benefit you and your child. It will give you and your child a better idea of his/her calorie intake, body fat and dietary habits. These factors can help determine if your child's diet is appropriate for his/her activity level. Overall, we hope to gain information on meal regularity and energy balance. We would like to see if the two are related to a higher body fat percentage.

V. Voluntary Participation and Withdrawal:

Participation in research is voluntary. Your child does not have to be in this study. If you allow and your child decides to be in the study and either of you change your mind, you both the right to drop out at any time. Your child may skip questions or stop participating at any time. Whatever you and your child decide, your child will not lose any benefits, either real or perceived, to which he/she is otherwise entitled.

VI. Confidentiality:

Consent Form Approved by Georgia State University IRB October 19, 2011 - November 11, 2011

We will keep your child's records private to the extent allowed by law. Only the research investigators (Laura Worsham, Dan Benardot, and Anita Nucci) will have access to your information. Information may also be shared with those who make sure the study is done correctly (GSU Institutional Review Board, the Office for Human Research Protection (OHRP) and/or the Food and Drug Administration (FDA), and the sponsor). We will use a study number rather than your child's name on study records. The information you and your child provide will be stored on firewall-protected computers and password protected software. Your child's name and other identifying facts will not appear when we present this study or publish its results. The findings will be summarized and reported in group form. Your child will not be identified personally.

VII. Contact Persons:

Contact Laura Worsham at lworsham1@student.gsu.edu if you have questions about this study. If you have questions or concerns about your rights in this research study, contact Susan Vogtner in the Office of Research Integrity at 404-413-3513 or svogtner1@gsu.edu.

VIII. Copy of Consent Form to Subject:

We will give you a copy of this consent form to keep.

If you are willing to volunteer for this research, please sign below.

_____	_____
Participant	Date
_____	_____
Principal Investigator or Researcher Obtaining Consent	Date

APPENDIX B

THE EFFECT OF MEAL FREQUENCY AND ENERGY BALANCE FLUCUATIONS ON BODY COMPOSITION IN CHILDREN AND ADOLESCENTS

Student PI: Laura Worsham
PI: Dr. Dan Benardot

In this study we are trying to learn about food and exercise. We want to know how these things affect your weight. You have been asked to be here because you are between 8 and 16 years old. If you join this study, you will be weighed and have your body fat taken. Next you will explain what you ate and your activity the day before. Last you will be asked to answer some questions. The questions will ask how you eat at school and home. This study will be at Titus Sports Academy at Chastain Park in Atlanta, GA. The study should take about 1 hour. Your parent/guardian will be with you the entire time. You will not be asked to be alone. You will not be alone with anyone doing this study.

The researchers hope this study helps them learn how eating and exercise affects weight. The benefit to you is you might learn ways to be healthier.

You don't have to be in this study if you don't want to. You can quit the study at any time. If you don't like a question, you don't have to answer it. If you ask, your answers will not be used in the study. No one will get mad at you if you don't want to be in the study.

Other than the researchers, no one will know your answers. If you have any questions, ask the researcher.

This research study has been explained to me and I agree to be in this study.

_____ _____
Subject's Signature for Assent **Date**

Check which applies (to be completed by person conducting assent discussion):

The subject is capable of reading and understanding the assent form and has signed above as documentation of assent to take part in this study.

The subject is not capable of reading the assent form, however, the information was explained verbally to the subject who signed above to acknowledge the verbal explanation and his/her assent to take part in this study.

_____
Name of Person Obtaining Assent (Print)

_____ _____
Signature of Person Obtaining Assent **Date**

Consent Form Approved by Georgia State University IRB October 19, 2011 - November 11, 2011

APPENDIX C

THE EFFECT OF MEAL FREQUENCY ON ENERGY BALANCE FLUCTUATIONS IN OVERWEIGHT CHILDREN AND ADOLESCENTS

Student PI: Laura Worsham
PI: Dr. Dan Benardot

WEEKEND QUESTIONNAIRE

Subject/Client #: _____ Date: _____
mm/dd/yyyy

1. Typical eating habits on weekend

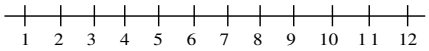
- Mostly eat at home
- Mostly go out to eat

2. Do you typically snack?

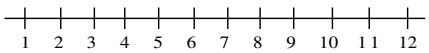
- Yes
- No

If yes, what time(s) of day do you snack? (circle all that apply)

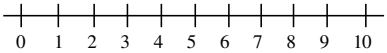
Morning:



Afternoon/Evening:



3. On average, how many regular meals do you eat on a weekend day? (Circle best answer)



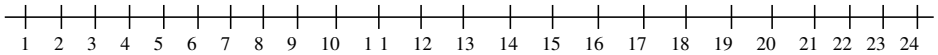
4. If skipping meals at home, main reason for skipping meals?

- Not hungry
- Dislike food that is available
- Not enough time to eat
- Embarrassed

5. Review the activity chart provided and describe to the best of your ability how your activity is divided throughout a weekend day (Sat. and Sun.). Total must equal 24 hrs.

How many hours are spent at level 1? ____ level 2? ____ level 3? ____ level 4? ____

If needed use the guide below to help track your hours. Each individual hash mark represents an hour in your day (not the time of day).



THE EFFECT OF MEAL FREQUENCY ON ENERGY BALANCE FLUCTUATIONS IN OVERWEIGHT CHILDREN AND ADOLESCENTS

Student PI: Laura Worsham
PI: Dr. Dan Benardot

Activity Chart	
1	At Rest: Sleeping, sitting, relaxing, watching T.V.
2	Light: Normal everday activity. Walking to class, household chores
3	Moderate: Walking fast, heart rate and breathing faster than normal
4	Heavy: Running, sweating, heart rate and breathing very fast.

THE EFFECT OF MEAL FREQUENCY ON ENERGY BALANCE FLUCTUATIONS IN OVERWEIGHT CHILDREN AND ADOLESCENTS

Student PI: Laura Worsham
PI: Dr. Dan Benardot

Subject/Client #: __12_____ Date: _____10/10/2010_____
mm/dd/yyyy

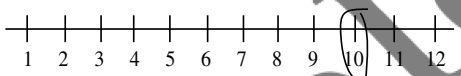
1. Typical eating habits on weekend

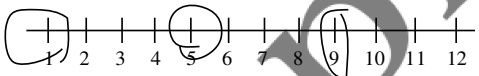
! Mostly eat at home
Mostly go out to eat

2. Do you typically snack?

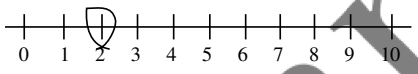
! Yes No

If yes, what time(s) of day do you snack? (circle all that apply)

Morning: 

Afternoon/Evening: 

3. On average, how many regular meals do you eat on a weekend day?
(Circle best answer)



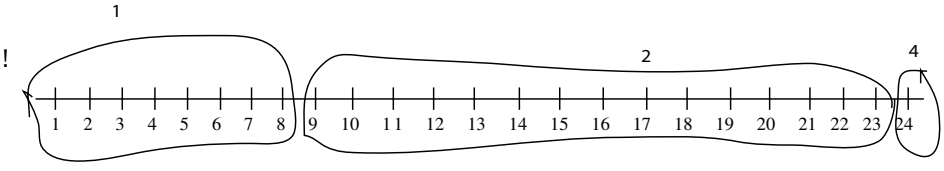
4. If skipping meals at home, main reason for skipping meals?

Not hungry Not enough time to eat
Dislike food that is available ! Embarrassed

5. Review the activity chart provided and describe to the best of your ability how your activity is divided throughout a weekend day (Sat. and Sun.). Total must equal 24 hrs.

How many hours are spent at level 1? _8_ level 2? _15_ level 3? _0_ level 4? _4_

If needed use the guide below to help track your hours. Each individual hash mark represents an hour in your day (not the time of day).



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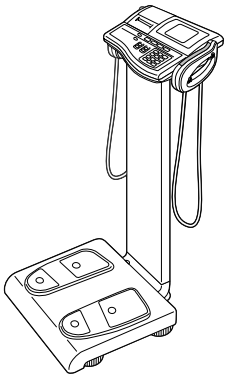
APPENDIX D

TANITA

BODY COMPOSITION ANALYZER
BC-418
INSTRUCTION MANUAL



Please read this Instruction Manual carefully and keep it handy for future reference.

APPENDIX E

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