ABSTRACT

Measured Resting Energy Expenditure Using a Fixed Function Indirect Calorimeter in the Clinical Setting as a Predictor of Success with Weight Change in An Obese Pediatric Population

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The American Dietetic Association (ADA) standard of care for obese adults utilizes indirect calorimetry for calculating caloric targets for weight loss (1). Even though rates appear to be leveling off (2), childhood obesity is one of the major public health concerns of our time and much attention is currently being given to understanding the obese state. Resting energy expenditure (REE) makes up 60-70% of total energy expenditure and plays a major role in determining an individuals' daily energy needs and metabolism. In the clinical setting, indirect calorimetry is often unavailable, thus predictive equations are typically used to help set caloric goals for weight loss.

The first objective was to compare measured resting energy expenditure (MREE) using a portable indirect calorimeter with five predictive equations used to determine energy needs for children participating in the East Carolina University's Healthy Weight Clinic. The investigators also wanted to determine which of these equations are best to use in an obese pediatric population in the clinical setting. Results indicate that there is a significant ($p < 0.05$) and strong correlation between MREE and these five predictive equations; however, there are also

significant discrepancies. Overall, the Harris Benedict equation demonstrates the lowest mean calorie difference when compared to MREE.

Secondly, it was hypothesized that those subjects with a higher baseline MREE would be more successful with weight loss, and that metabolic factors such as leptin may contribute to weight change in an obese pediatric population. It was also proposed that there may be validity in adjusting MREE to body weight and/or body composition to account for confounders such as age and gender. MREE does not appear to predict success with weight change in obese youth aged 7-18 years. In older obese youth (Tanner Stage 5) it appears that those with a lower baseline fat mass and higher adjusted MREE to fat mass, may have more success with decline in BMI z score. Also, leptin and fat mass significantly ($p < 0.05$) and negatively correlated with BMI z score change in older youth.

MEASURED RESTING ENERGY EXPENDITURE USING A FIXED FUNCTION INDIRECT CALORIMETER IN THE CLINICAL SETTING AS A PREDICTOR OF SUCCESS WITH WEIGHT CHANGE IN AN OBESE PEDIATRIC POPULATION

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DEDICATION

I dedicate this project to Connie Bales and Gloria Henes.

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LIST OF ABBREVIATIONS

AA	African American
AAP	American Academy of Pediatrics
ADA	American Dietetic Association
ACO	acyl-CoA oxidase
AMPK	adenosine monophosphate-activated protein kinase
BMI	body mass index
BSA	body surface area
C	Caucasian
CPT1	carnitine palmitoyltransferase 1
DEXA	dual energy X-ray absorptiometry
ECU	East Carolina University
FAS	fatty acid synthase
FOXC2	forkhead transcription factor
FFA	free fatty acid
FFM	fat free mass
FM	fat mass
HOMA	homeostatic model assessment
HWC	healthy weight clinic
IOM	Institutes of Medicine
Kcal	kilocalorie (calorie)
Kg	kilogram

LBM	lean body mass
LM	lean mass
LnHOMA	log of HOMA (homeostatic model)
MREE	measured resting energy expenditure
PA	physical activity
PHWRTC	Pediatric Healthy Weight Research and Treatment Center
PPAR	peroxisome proliferator-activated receptor
PREE	predicted resting energy expenditure
RD	Registered Dietitian
REE	resting energy expenditure
RQ	respiratory quotient
SE	standard error
STP	standard temperature and pressure
TAG	triacylglycerol
TBW	total body weight
TDEE	total daily energy expenditure
TEE	total energy expenditure
TEF	thermic effect of food
WHO	World Health Organization

CHAPTER 1-REVIEW OF LITERATURE

WHY STUDY CHILDHOOD OBESITY?

As one of the greatest public health concerns of our time, at least 17% of today's youth are considered obese – or above the 95th percentile BMI for age and gender (6). Despite reports that rates are leveling off (2), childhood obesity is still a great concern in the medical and nutrition communities as it is associated with co morbidities such as hypertension, hyperlipidemia and type 2 diabetes.(7). Two decades ago, type 2 diabetes was almost unheard of in children, and today, 30-50% of all new childhood diabetes cases are Type 2. In 95% cases of newly diagnosed type 2 diabetics, these children are obese (8). In addition, if an adolescent is obese in his/her teen years, there is a 70-80% chance that the same youth will be an obese adult. (9). Currently, overweight and obesity in the United States accounts for over $78 billion dollars in medical health care costs. (10)

There remains much debate as to the causes of childhood obesity in terms of 'nature vs. nurture"- when in fact both may attribute to the development of the disease (11). Some studies have shown that there is a 30-40% link between childhood obesity and genetic influences as inherited from parents- such as body composition, resting energy expenditure, (REE), and hormonal influences on metabolism such as thyroid function (5). However this indicates as much as 50-60% of childhood obesity may be explained by environmental influences. Thus it is seems that there is an interaction between genetic and environmental factors that impact the development of obesity in our youth.

Summary: *Childhood obesity is a public health concern with detrimental health consequences for our youth and further development of obesity into adulthood. Much interest and debate surrounds the causes of this disease as related to its genetic and environmental influences.*

The obese state is defined as an imbalance between energy intake and energy expenditure; whether it be food intake is too high, energy expenditure is too low (through either low REE and/or physical activity), or a mixture of both (11) This project focuses on the energy expenditure aspect of obesity, particularly REE. REE accounts for approximately 60-70% of total energy expenditure (12). In the clinical setting, REE is typically calculated using predictive equations so as to help determine caloric targets for weight loss in an obese patient. Thus while utilizing equations developed to account for height, weight, age, and physical activity factors, the clinician can then address the other side of the "balance" equation and recommend appropriate energy intake for weight maintenance or weight loss. The American Dietetic Association (ADA) promotes measuring REE via indirect calorimetry in the obese adult population as the "gold standard" for determining goals for energy intake for weight loss.(1) **A central hypothesis of this project is that MREE is a predictor of weight loss in an obese pediatric population.** Many studies have established the relationship between measured REE (MREE) and predicting REE (PREE) with various equations, in both obese adults and children (13). However, particularly in the obese pediatric population, there is still debate as to what equation predicts with most accuracy. (14, 15, 16) Our preliminary data using an obese pediatric population indicates a correlation between MREE and a commonly used predictive equation- the Harris Benedict equation (Henes S et al, 2008 unpublished). However, as ours and other data suggest,

there is much variability between MREE and PREE. (17, 18, 19). Even small under or over-estimations in energy needs can greatly impact weight loss efforts, especially in children. The current literature has begun to investigate hand held calorimetry compared to predictive equations, however only published results have been shown in obese women (20**).** This particular study concluded that there was a significant difference between MREE and predictive equations and that further research was needed in utilizing hand held indirect calorimetry. Thus one aspect of this project is to demonstrate that using a fixed function indirect calorimeter is a necessary clinical tool in accurately determining caloric targets for weight loss in an obese pediatric population.

Summary: *Resting energy expenditure (REE) is a major component in the energy balance equation. It is often utilized in the clinical setting- whether estimated with equations or measured via indirect calorimetry- to help determine the appropriate energy intake goals for weight loss. There is great variability between MREE and PREE, especially in an obese pediatric population.*

WHEN INDIRECT CALORIMETRY IS NOT AVAILABLE, WHAT IS THE MOST APPROPRIATE PREDICTIVE EQUATION TO USE IN A MORBIDLY OBESE PEDIATRIC POPULATION?

As noted previously, there are various predictive equations that are utilized in the clinical setting to estimate REE and determine caloric targets for weight loss with obese patients. Although there have been several studies investigating and comparing these various equations used in obese children, there still remains controversy. Predictive equations such as the Shofield have been developed using a pediatric population and some studies indicate this as a valid

equation for use in an obese pediatric population (21) The WHO equations are also often utilized to determine energy needs of children.(22)

Other investigators (14,23) have developed equations using an obese pediatric population. Currently, the ADA recommends using the Institute of Medicine (IOM) equation for determining energy needs in an obese pediatric population (1). One of the most recent studies compared 43 predictive equations in 121 obese adolescents and noted the most commonly used Shofield equation significantly over-estimated REE, the Lazzer equation provided a fair prediction of REE (71%), and the best predictive equation appeared to be the Molnar equation when compared to MREE (24). As noted, various populations yield various results. As many authors also state, it may be appropriate to use different equations based on gender and racial differences.

In healthy adults, the Harris Benedict has long been considered the 'gold standard' in determining energy needs, while the more current Mifflin-St Jeor equation has been developed for use in obese adults and is recommended by the ADA for use in this population. One study using adolescent (age 12-17) obese Brazilian boys (25) concluded that the most commonly used equations overestimate REE, and that the Harris Benedict equation was one that showed no significant difference between measured and predicted REE. Thus this project takes the approach of using fixed function indirect calorimeter to measure REE in the clinical setting and comparing this with 5 commonly used predictive equations: the ADA recommended IOM, the Shofield, the WHO, the Harris Benedict and the Mifflin St Jeor.

Summary: *Although several studies have investigated many predictive equations commonly used in pediatrics and with obese children, there remains no consensus as to the 'best' one to use. Most studies do agree that using indirect calorimetry is a more accurate*

measurement of REE than any predictive equation. To our knowledge, no studies have investigated the use of a fixed function calorimeter in an obese pediatric population and compared this measured REE to commonly used predictive equations, particularly the ADA recommended IOM equations.

DOES MREE AFFECT RATES OF WEIGHT LOSS IN AN OBESE PEDIATRIC POPULATION RECEIVING TREATMENT AT A PEDIATRIC HEALTHY WEIGHT CLINIC?

In addition to body weight, body composition has been shown to be a determinant of REE in obese children. (26). Body composition is comprised of fat mass (FM) and fat free mass (FFM). FFM includes lean body mass (LBM) and bone mass. Typically, FFM is more metabolically active than FM due to LBM. Studies have shown that FFM is a major determinant of REE in adults and children (21) as well as in a mixed population of obese and non-obese children and adolescents (13). Others (18) have demonstrated that LBM alone is a best predictor of REE in obese children. Butte et al (26) have also shown in addition to body weight, FFM, FM, gender and Tanner stage (pubertal status) also have significant influences on REE. Our preliminary data demonstrates obese children with similar body weights may have different MREE. *Can this difference be explained by differences in body composition?*

An interesting study by Delaney and associates (27) investigated predictors of change in body fatness over a 2 yr time period in children, lean and obese, aged 9-11 yrs (n= 114). A significant predictor of fat gain in these children was lower REE (r^2 = .217, P<.00001) in addition to total daily energy expenditure (TDEE), the thermic effect of food (TEF) and Respiratory Quotient (RQ). Another study investigated predictors of long term weight

maintenance in adults (28). Ninety-two overweight men and women were studied over a 2 yr time. Ninety-two overweight men and women were studied over a 2 yr time period after a weight loss program. There was a negative correlation with baseline MREE and percent weight regain ($r = -0.38$, $p= 0.01$), and baseline fat mass and percent weight regain ($r= -0.24$, $p = 0.05$). It was determined that a higher baseline MREE was one of the best predictors of success with weight loss.

Summary: *We hypothesize that those obese children with a lower MREE will have less success with weight loss. Differences in MREE may be accounted for by body composition.*

SHOULD MREE BE 'NORMALIZED' IN OBESE CHILDREN TO MAKE COMPARISONS AND TO TEST THE HYPOTHESIS THAT MREE IS A PREDICTOR OF WEIGHT LOSS IN THIS POPULATION?

MREE via indirect calorimetry provides the number of calories per day (kcal/day) an individual needs at rest. To determine the individual's total energy needs (TEE), an activity factor takes into account the typical daily activity of the individual. This is important in various disease states- such as obesity to help determine an individual's energy needs for weight loss.

REE is 30-40% genetically determined and 60-70% influenced by factors within the individual- such as body composition, age, gender and ethnicity (29). It is difficult to "tease" out the various influences on an individuals' REE (nature vs. nurture); however the individual variances, particularly body composition has been the focus of much research. As early as the 1920s experiments have determined a relationship between REE and body mass of an individual. Later studies then indicated that indexing REE to fat free mass (FFM),which includes skeletal

muscle, was a more accurate method to determine intergender differences in REE (i.e. men have more FFM than women). (30) This concept of "normalizing" MREE to some factor has become important in research so as to account for confounders such as gender, age, and ethnicity. Ways to normalize MREE would include kcal/ kg FFM, kcal/kg body weight (and/or BMI), and kcal/kg fat mass. Thus the research question becomes: **Is it important to normalize MREE so as to make comparisons in obese children and to test the hypothesis that MREE is a predictor of weight loss in these obese children?** The literature suggests that since skeletal muscle comprises approximately 40-50% body mass of most individuals and contributes to about 18-20% total REE, FFM is often the most "conventional" way to normalize MREE (30,31). Most of these studies utilize the adult population. There is no consensus however in the literature as to the best way to normalize MREE in children, let alone in obese children. Tershakovek A and associates (32) studied 203 obese African American and white children aged 5-17yrs. The average age was 10 years old, and pubertal status was divided as Tanner Stage 1 = prepubertal; Tanner Stage >2= peripubertal-pubertal. REE was measured via indirect calorimetry with metabolic cart and body composition was assessed using Dual Energy X-Ray Absorptiometry (DEXA). FFM was defined as lean tissue mass and bone mass, and lean tissue mass was equal to skeletal muscle and organ tissue. The study investigated the following as potential independent factors predicting REE: age, gender, ethnicity, FFM- (whole body, trunk region, limbs), and fat mass (FM). The authors separately investigated trunk FFM to better assess the potential effect of more metabolically active organ tissues (located in the trunk region) vs. skeletal muscle (less metabolically active) on REE. The rationale for investigating limb FFM was to "tease out" gender and ethnic differences. Various studies have shown that males typically have a higher MREE than females and whites have higher MREE than African

Americans (AA). (26,33). The investigators postulated that longer limbs (more skeletal muscle) and a shorter trunk region (less mass in metabolically active tissue) in AA may partially account for racial differences in MREE. Multiple regression was used in three predictive models: each included age, ethnicity, gender, fat mass as independent variables for MREE, while separately looking at whole body FFM, trunk FFM and limb FFM. An important finding in this study was that fat mass was an independent predictor of MREE ($p<0.0001$). Another interesting finding is that MREE decreased with age in children 5-17 years of age. The authors postulated that this may be due to the changing composition of FFM through growth with a smaller proportion of metabolically active tissues compared to skeletal muscle, as well as bone growth. Lastly , the study confirmed that independent of body composition, MREE was higher in boys than girls. In a recent review , Muller M et al. (34) discuss the body of literature that show how up to 80% of the variance in energy intake and energy expenditure is explained by body composition- to include both FFM and FM. This paper also discusses a 'multicomponent model' whereby individual organ masses (i.e. liver, heart, brain, and kidney) account for approximately 85% of the variance in REE. (2009). Thus, convention, particularly in an adult population has been to normalize MREE to FFM; however, data suggests that in an obese population, to include children, the effect of fat mass in predicting MREE is something to be considered.

Summary: *Convention often dictates that MREE be normalized to FFM; however, this data is mostly in adults. Recent data suggests that particularly in an obese pediatric population, taking into account fat mass while normalizing MREE may be important.*

IS THERE A GENDER BIAS IN SUCCESS WITH WEIGHT LOSS IN AN OBESE PEDIATRIC POPULATION AS RELATED TO MREE?

Butte and associates (26) demonstrate that gender is an independent factor affecting REE, where obese Hispanic boys aged 5-19 have higher MREE than their female counterparts. It is also known that boys and males in general, have a greater proportion of fat free mass than females, and accounts for higher energy expenditure and requirements. (35). Not many studies have investigated specific gender differences in weight loss in an obese pediatric population. Lazzer and colleagues (36) reported significant weight loss in obese adolescents, aged 12-16 (n= 26) after an intensive 9-month dietary and physical activity program. The authors note that exercise preserved fat free mass but not metabolic rate (MREE) in these children. Interestingly, the boys had a higher baseline MREE than the girls, and less of a decrease in MREE after the weight loss program. Also, the boys had similar baseline fat-free mass as the girls, as but with less FFM loss and more total weight loss than the girls. However, the focus on the study was not gender differences, as much as it was on changes in MREE after weight loss. Other studies suggest that a higher baseline MREE and less loss of FFM predict less weight regain after weight loss in adults (28, 37). A recent review by Sweeting (35) on childhood obesity suggests that there are gender differences in the way boys and girls respond to societal influences in an obesogenic environment, to consequences of being obese, and responses to interventions and treatments. The author suggests the current literature does not focus specifically on gender differences, but should be an area of further study in addressing childhood obesity. Thus, considering biological and psychosocial differences between boys and girls, interventions for obese children can be tailored based on gender to promote more favorable responses.

Summary: *We suspect that there are gender differences as related to MREE which will predict success of weight loss in an obese pediatric population receiving treatment at a pediatric Healthy Weight Clinic.*

IS ETHNICITY A PREDICTOR OF SUCCESS WITH WEIGHT LOSS IN AN OBESE PEDIATRIC POPULATION AS RELATED TO MREE?

The National Health and Nutrition Examination Survey (NHANES) 1999-2004 establishes that in the US there is a higher prevalence of obesity in African-American (AA) women than in white women (2). The literature indicates that AA individuals have a lower MREE than whites and this difference in energy expenditure has been an area of research in terms of explaining weight gain in this population (38). The general consensus is that a lower MREE in the AA adult population is most likely due to a smaller mass of less metabolically active organs and does not explain the prevalence of obesity in this racial group; however the literature is less clear in explaining racial differences in childhood obesity. Sun and associates (2001) studied 92 AA and white children, boys and girls (mean age 8 yrs.) over a 2 yr time period. The objectives were to investigate changes in FM, FFM, REE and Tanner Stage in these 2 populations of children, and to determine if there were racial differences in MREE. The authors determined that after adjusting for age, Tanner stage, FM, and lean mass (LM), these AA children had a significantly lower MREE compared to the white children, and this difference was not fully explained by LM distribution. Others have also shown that independent of LM, AA girls aged 6-16 had a lower MREE than white girls of similar ages. (39). When looking at obese and non obese prepubertal AA and white children (n= 34; mean age 9yrs), Kaplan et al (40) determined that MREE is significantly lower in black vs. white children, and that both FFM and ethnicity were determinants of MREE in this population, after adjusting for body size and composition. Delaney and associates (27) found similar results when looking at a mixed population of obese and non-obese AA and white children (mean age 10 yrs). Their results combined the obese and non-obese children and determined after adjusting for FFM, that the AA

children had a lower MREE and that these AA children expended less energy than the whites in physical activity. The most recent study to explore racial differences in MREE in healthy weight youth was done by Lee S et al. (41). The authors found that in the 50 AA and 51 white boys and girls, (aged 9-13), not only was MREE lower in the AA girls vs. white girls, but fat oxidation was also lower in this population. They did not observe the same results with the AA and white boys studied. The investigators concluded that this combination of lower MREE and fat oxidation may be a "metabolic risk phenotype" for a predisposition of young AA girls to gain weight and experience obesity.

Note that most of these studies investigate healthy weight children, or a mix of obese and lean children. Tershakovek and colleagues (32) are one of the few to investigate only obese AA and white children and adolescents, aged 5-17 yrs.(n= 203). They demonstrated that MREE was lower in AA vs. whites, in girls vs. boys (regardless of race), and that MREE declined with age. A conclusion of this study was that a higher bone density in these obese AA children partially explained differences in MREE. Another conclusion was that the lower REE of AA children may contribute to difficulty in weight management in this population.

Summary: *Many studies have determined that MREE is lower in AA children vs. white children. Studies are mixed in that some explain this racial difference in part by differences in FFM and bone density; others do not find this difference. Not many studies have investigated differences between obese AA and white children and adolescents. We would hypothesize that racial differences in MREE in an obese pediatric population will be a predictor of weight loss success during treatment at a pediatric Healthy Weight Clinic.*

IS THERE A RELATIONSHIP BETWEEN PLASMA LEPTIN LEVELS AND MREE IN OBESE CHILDREN PARTICIPATING IN A PEDIATRIC HEALTHY WEIGHT CLINIC?

Leptin has been well established in the literature as a long-term regulator of energy balance (42). The hormone is secreted primarily by adipose tissue and with normal functioning indicates a state of positive energy balance and regulates appetite to decrease intake. Leptin also increases activity of the sympathetic nervous system thus increasing energy expenditure of adipose tissue. In the obese state, leptin appears to lose its ability to decrease energy intake and increase energy expenditure. This is often termed 'leptin resistance', whereby serum concentrations of leptin are elevated without the common effects of decreased appetite and limiting food intake. Much of the literature agrees that elevated leptin levels correlates to the increase in fat mass present in obesity; however, this only partly explains the variability in leptin concentrations. (43). Research has been dedicated to explain possible relationships of leptin to other metabolic factors such as central vs. peripheral adiposity, energy expenditure and metabolic markers for cardiovascular disease. Liuzzi and associates (43) investigated this using 400 male and female obese adult patients with BMI 30-82. The results indicated that women had significantly higher plasma leptin levels even when adjusting for kg of fat mass. Leptin was positively correlated with BMI ($r = 0.55$, $P=0,01$)), although the authors noted intersubject variability. Thus patients with leptin values of 100ng/ml or higher may have BMI ranging from 35 to 82. Gender, BMI and percentage of fat mass accounted for approximately 55% of variability in leptin concentrations. Interestingly, independent of fat mass and gender, leptin concentrations were inversely correlated with MREE when expressed as an absolute value

(r= -0.69, P=0.001) but not when expressed as a ratio to FFM (r= - 0.17, ns). Niskanen et al (1997) also found this inverse relationship between leptin levels in obese subjects and MREE (and RQ as well) after adjusting for fat mass, age and gender.

There are limited studies of plasma leptin and its relation to childhood obesity. One investigation by Savoye M et al (45) however, helps determine a relationship between plasma leptin levels and predicting future weight gain in obese children aged 7-18 yrs. The researchers measured baseline fasting leptin and insulin levels in 68 obese children- male, female , Caucasion and African American. BMI and BMI z score were also calculated. All measurements were again determined after a 2 ½ yr follow-up period. Gender specific multiple linear regression was used to determine the longitudinal relationship between changes in BMI z-score, insulin and leptin levels. A strong positive correlation was found between plasma leptin levels and BMI in both obese boys ($p<0.0001$) and obese girls ($p < 0.0002$). An important finding was that basal leptin levels were indicative of greater positive changes in BMI z-score in girls. Further, basal leptin measurements explained 18% of the increase in BMI over a 2 ½ yr period. (p, 0.0006) . Recently (46), Reinehr R and associates postulated that leptin levels were negatively associated with degree of weight loss in obese children participating in a lifestyle intervention. The investigators studied 248 obese subjects 8-14 yrs, who participated in an 8 week intervention program based on physical activity, nutrition education and behavior therapy. The findings indicated that baseline leptin concentrations were significantly correlated with BMI, pubertal stage, gender, waist circumference, and insulin. There was a significant negative relationship between degree of weight loss and baseline leptin concentrations.

Summary: *The literature establishes a relationship between leptin levels and MREE ; however mostly in an adult population and with conflicting results. Further research is needed to determine a relationship in obese children.*

WHAT ARE THE ACTIONS OF LEPTIN ON PERIPHERAL TISSUES WHICH MAY AFFECT MREE IN OBESE CHILDREN?

It is well know that leptin acts centrally to decrease food intake and to increase energy expenditure, most directly via the hypothalamus. More recent studies, however, indicate that leptin receptors exist on peripheral tissues to include the liver, skeletal muscle, and adipocytes such that a direct effect has been observed. (47,48,). Both Muoio et al (49,50) and Minokoshi Y et al (51) have shown that leptin stimulates fatty acid oxidation in skeletal muscle. Another important point relevant to leptin and its action is the interplay between this hormone and others such as insulin. Muoio et al (50) found that leptin's action opposed the affects of insulin in triacylglycerol accumulation in both lean and ob/ob mouse skeletal muscle. Given skeletal muscle comprises approximately 40% of total body weight, leptin's peripheral action in skeletal muscle metabolism may have an overall effect on fuel homeostasis and weight regulation.. It is not unreasonable then to propose that leptin-stimulated free fatty acid regulation may then provide a peripheral mechanism by which MREE is affected by plasma leptin levels. Given the interplay between insulin action on skeletal muscle and leptin, perhaps both insulin resistance and leptin resistance, which are common states in obesity, may have a deleterious affect on skeletal muscle metabolism, thus MREE. Elevated insulin levels may promote the uptake of TAG and FFA into skeletal muscle; however inhibits the utilization of these as fuel sources. Leptin's action may serve to promote utilization of these as fuel; however as the state of leptin

resistance increases in obesity, its function to essentially increase energy expenditure in skeletal muscle metabolism may then begin to falter. The issue becomes more complex in attempting to tease out the effects of one hormonal function on the other. That is to question how elevated leptin levels as marked by increased adipose tissue affect insulin resistance; and how may elevated insulin levels as marked in the state of obesity contribute to leptin's action or dysfunction in attenuating the storage of fuels in skeletal muscle. More research is needed in this area to investigate the interactions of these two hormones in regulation of fatty acid oxidation which may ultimately help elucidate the development of the obese state.

In terms of leptin action on adipocytes, Wang et al (52) used zucker rats to investigate this phenomenon. The investigators isolated adipocytes from both lean and obese rats, cultured with recombinant leptin and performed glycerol and free fatty acid (ffa) assays. The ob/ob adipocytes were used as a control since these contain defective leptin receptors, and as expected, there was no effect on FFA or glycerol release in this model. However, in the 'healthy' adipocytes, leptin significantly lowered the mRNA of leptin, and fatty acid synthase (FAS), and upregulated the mRNA of PPAR-alpha, CPT-1 and ACO- all known to up-regulate FFA oxidation. Interestingly, glycerol was released from these adipocytes but without a proportional release of FFA. This supports the idea that FFA are oxidized within the adipocyte rather than released to the liver. As the authors noted, further investigation is needed to shed light on this potential mechanism of lipolysis.

Summary: *Investigators have shown peripheral action of leptin in skeletal muscle. Given that skeletal muscle contributes to a large percentage of resting energy expenditure leptin may perhaps act peripherally as well as centrally in helping to regulate MREE with particular*

consequence in the obese state. There is also some evidence that leptin may act directly on the adipocyte to promote upregulation of FFA oxidation.

RESTATEMENT OF HYPOTHESIS

The primary hypothesis is that measured resting energy expenditure (MREE), as determined by a fixed function calorimeter in the primary care setting, is a predictor of weight loss in an obese pediatric population. Additionally, we propose taking into account fat mass as well as FFM when normalizing MREE. We further hypothesize that rates of weight loss will be determined based on gender and racial differences, independent of body composition. Our last hypothesis is that metabolic markers of obesity, such as leptin, will factor into rates of weight loss in an obese pediatric population while peripheral actions on tissues such as skeletal muscle may affect metabolic homeostasis and energy expenditure.

The central hypothesis of this proposal is that measured REE (MREE) is a predictor of weight loss in an obese pediatric population and a necessary measurement in the clinical setting to determine accurate caloric needs in this population.

Specific Aim 1: The use of a portable calorimeter as a clinical tool for measuring REE has been compared to research standards of using a metabolic cart (3, 4). Results indicate that measurements are comparable, and the fixed function calorimeter is less complex, less expensive and more practical to use in the clinical setting. Our preliminary data suggests that although there is a strong correlation between PREE and MREE, there is great variability between actual and estimated REE, and among various predictive equations (Henes, 2008, unpublished data).

The following questions will be investigated in Aim 1:

a. How does MREE using the KORR ReeVue calorimeter compare with PREE equations such as the Harris Benedict, Shoefield , Mifflin St Jeor , WHO and the IOM equations in obese children

b. In comparison to MREE- what is the best predictive equation to use in an obese pediatric population when indirect calorimetry is not available in the clinical setting?

Specific Aim 2: Preliminary data suggests a positive correlation between MREE and body weight (kg). The overarching hypothesis is that MREE is a predictor of success of weight loss in obese children whereby those with a higher baseline MREE will be more successful with weight loss as defined by change in BMI z score. There are various well known contributors to MREE to include age, race, gender, body weight and body composition. (5) Lean body mass- particularly skeletal muscle- is purported to be a main contributor to 'metabolism' and REE. In adult studies, REE has often been 'normalized' to fat free mass (FFM); however no current studies, to our knowledge, address this issue in children; particularly obese children. Thus Aim 2 explores the rationale for 'normalizing'MREE to factors such as body weight, FFM, and fat mass (FM). The following questions will be investigated in Aim 2:

a. Is MREE a predictor of weight change in an obese pediatric population undergoing treatment in a 3-6 month healthy weight program?

b. Is it important to 'normalize' MREE to account for body composition? If so, should total body weight, FFM and or FM be considered?

c. Are there other metabolic factors- such as leptin- that contribute to weight change in an obese pediatric population?

METHODOLOGICAL CONSIDERATIONS

USE OF INDIRECT CALORIMETRY IN THE CLINICAL SETTING:

Total energy expenditure (TEE) is the amount of energy required to carry out metabolic processes within the body, both synthesis and breakdown, and to the cellular level. There are three main components that make up TEE: REE (60-70%), thermic effect of food (10%), activity expenditure (20-25%). As noted, REE is the largest component of TEE and is the energy required to maintain normal regulatory balance and body functions at rest (11). REE is often considered the 'metabolism' of an individual- which is scientifically defined as the heat generated for cellular processes needed for the body. This unit of measure is called the "calorie" which is equivalent to the amount of heat needed to raise the temperature of 1 gram of water 1 degree Celsius. This is normally expressed as a Kilocalorie (1000 calories) and how we refer to metabolic processes in the human body. There are two main ways to determine this energy in humans- direct and indirect calorimetry. Direct calorimetry utilizes a specially constructed chamber that measures the amount of heat liberated from the body. Air temperature remains constant and heat from the subject's body warms an existing water bath equal to rate of heat generated from the body. This is difficult to perform and is not practical in the day to day living of human subjects. Indirect calorimetry utilizes the science that 95% of energy expended in the body is derived from reactions with oxygen. Typically, regardless of type of food ingested, the energy liberated per liter of oxygen used in the body is about 4.825 kilocalories (or Calories).

The Weir Equation is the basis for using indirect calorimetry- in the research setting with a metabolic cart, and in the clinical setting using a portable indirect calorimeter.

Wier Equation: Kcal/minute= Expired minute ventilation (volume of air breathed in each minute, corrected for STPD conditions) X (1.044-0.0499 x percent expired oxygen). This then provides the calculated calories/day (kcal/day) per individual-also known as REE. The metabolic cart used in the research setting, is a 30-45 minute test, whereas a hand held; or portable calorimeter can be conducted in 10-15 minutes. There are several studies that have validated such portable calorimeters to a research standard such as a metabolic cart (3, 4, 20,53).

In this project, the KORR ReeVue indirect calorimeter was used in the clinical setting. The criteria included patients referred to the East Carolina University's Pediatric Healthy Weight Clinic aged 7-18 yrs. The specifications were that the patient was fasting for 12 hrs, and was able to sit calm and still for the duration of the procedure (10-15 minutes). Exclusion criteria included any patient not fasting, not able to complete the test, unable to sit still (i.e. fidgeting), and on any medications that could potentially alter metabolism (to include psychotropic medications, ADHD medications).

The procedure took place in a quiet room, where the indirect calorimeter had been set up, and calibrated to STDP. The Healthy Weight Registered Dietitian (RD) conducted the procedure with explanation to the patient. Briefly, the patient wore nose clips, and breathed through the disposable tubing while keeping a tight seal around the mouthpiece. The patient sat comfortably in a chair and remained calm and still during the procedure. The patient acclimated to testing during the first three minutes, and then display calories were written down by the RD from 3-10 minutes. The final reading at 10 minutes (the average of minute 3 through minute 10) was used as the MREE, and utilized in making appropriate caloric targets for weight loss, and according to ADA and AAP recommendations for age.

CALCULATION OF BODY SURFACE AREA AND BODY COMPOSITION

As a project undertaken to reflect the clinical applications of measuring REE in 'real time' at a Healthy Weight Clinic, measurement of body composition was not a component of the patient's typical first visit. One aspect of this project is to utilize clinical approaches in answering our research question, which is: does MREE predict success of weight loss in an obese pediatric population. Outright, accounting for body composition is not a necessity. However, as the project evolved, there appeared a strong relationship between MREE and body weight (as expected) and the question of normalizing MREE to body weight came into play whereby addressing body composition became more of a factor. Since at the outset, body composition measurements were not performed in the patients studied, a clinical tool was utilized to estimate each subject's body composition utilizing an equation for body surface area (BSA). In pharmacology, equations such as the Mostellar Equation are often used to determine BSA in dosing medications, and based on being fat or water soluble. (54). A report by ThanhVu (55), Pharmacologist with the Cross Cancer Institute recommends the use of the Mostellar equation (compared to others), and discusses with references utilization of this equation in various populations- to include children and the obese. Thus, an online calculator : http://www.halls.md/body-surface-area/bsa.htm (56) which used the Mostellar equation was utilized to input each patient's height, weight, age and gender , and determine BSA and lean body weight (LBW). It is from this calculation that fat mass and percent body fat was calculated as such. LBW (kg) was subtracted from total body weight (TBW, kg) to give fat mass. This was then divided by TBW to give percent fat of each patient. Others (57) have also determined fat mass via this method/calculation, while determining LBW with bioelectrical impedance.

CHAPTER 2: MEASURED RESTING ENERGY EXPENDITURE IN OBESE YOUTH USING THE REEVUE FIXED FUNCTION CALORIMETER IN A PEDIATRIC HEALTHY WEIGHT CLINIC COMPARED WITH 5 COMMONLY USED PREDICTIVE EQUATIONS

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Abstract: The American Dietetic Association (ADA) standard of care for obese adults utilizes indirect calorimetry for calculating caloric targets for weight loss (1). Even though rates appear to be leveling off (2), childhood obesity continues to be one of the major public health concerns of our time and such organizations as the American Academy of Pediatrics (AAP) and the ADA have continued to address this disease state with evidence-based recommendations (1,58). Currently, the ADA recommends using the Institutes of Medicine (IOM) equations to estimate caloric needs for obese children when indirect calorimetry is not available. The purpose of this paper is to compare measured resting energy expenditure (MREE) of obese youth with the use of a portable indirect calorimeter in the clinical setting with five commonly used predictive equations.

Resting energy expenditure was measured via a portable indirect calorimeter. MREE was determined in obese youth aged 7-18 years who were referred to the East Carolina University Healthy Weight Clinic and who were greater than the 85th percentile BMI for age and gender.

Results indicate that there is a significant ($p< 0.05$) and strong correlation between MREE and these predictive equations; however, there are also significant discrepancies. While there appears to be no significant racial differences between MREE and PREE equations, there is more variability between MREE and these equations among non- Caucasian . There is a significant difference ($p <0.05$) in gender whereby, there is a greater mean difference between the predictive equations and MREE in the boys. Thus, these equations tended to give more of an over (or under) prediction of REE in boys vs. with girls.

Overall, the Harris Benedict equation demonstrates the lowest mean difference in energy expenditure (kcal/day) when compared to MREE. These data add to a growing body of literature that suggests indirect calorimetry is a more accurate means to determining caloric targets for weight loss in an obese population when compared to commonly used predictive equations. To our knowledge, this investigation is the first to report differences between these commonly used predictive equations, particularly the IOM equations, and measured energy needs using portable indirect calorimetry in the clinical setting.

INTRODUCTION: Childhood obesity is one of the most dire public health concerns of our time. Particularly over the past 5 years, evidence based research has led to recommendations from the AAP and ADA in prevention and treatment of this epidemic (1,58). Currently, the ADA recommends using indirect calorimetry to determine resting energy expenditure (REE) in an obese population; however, particularly in the pediatric clinical setting this measurement is not widely available. As REE is the basis for determining caloric needs for weight loss in obesity, most clinicians use a variety of predictive equations. Many predictive equations have been developed for use with children in the clinical setting (14, 15, 16, 24, 25); however there remains lack of consensus of what is best to use in an obese pediatric population; particularly in morbidly obese children(>99thpercentile BMI for age and gender). There is some data comparing commonly used predictive equations in an adult population with measured resting energy expenditure (MREE) using a hand held indirect calorimeter in the clinical setting (20). However, to our knowledge there is no data comparing predictive equations to MREE using a portable indirect calorimeter in the clinical setting and an obese pediatric population. In Spears and associates' study, the subjects were women older than 25 years of age, and with a BMI > 25, or overweight. The findings indicated the usefulness of a portable indirect calorimeter in determining MREE in this population and that there was a significant discrepancy between predicted and measured REE. The investigators concluded that further research was needed to investigate measured REE with the use of such devices and compare with predictive equations. Thus, the present study investigates the relationship and differences between REE measured in the clinical setting using the ReeVue portable calorimeter and five commonly used predictive equations: the Institutes of Medicine for children 3-18 yrs (IOM) , the Shofield, the World Health Organization (WHO), the Mifflin St Joer, and the Harris Benedict equations. The current

study aspires to add to a growing body of literature that indicates the usefulness of measuring energy expenditure in the clinical setting , specifically in an obese pediatric population. Additionally, the investigators hope to contribute data to help determine most appropriate predictive equation(s) to use with this population when indirect calorimetry is unavailable in the clinical setting.

METHODS:

SUBJECTS: Youth aged 7-18 referred to the East Carolina Pediatric Healthy Weight Clinic, who were > the 85th percentile BMI for age and gender, who were fasting, and who were able to perform indirect calorimetry. Patients who fidgeted during testing, were unable to complete the test, or who were taking medications that could potentially alter metabolic rate were excluded.

INDIRECT CALORIMETRY: The KORR ReeVue portable indirect calorimeter. (http://www.korr.com/products/reevue.htm).(59) was set up in a clinic exam room designated for the sole purpose of testing. Calibrated to room standard, temperature, pressure, dry STDP. Each patient was provided a new and separate disposable tubing, mouthpiece and nose clip. Testing was performed for ten minutes. The patient sat calmly and still in the quiet room, breathing as they would normally breathe into a mouthpiece, with a nose clip in place to prevent air escaping through the nose. Readings were recorded by the pediatric registered dietitian (RD) at every 30 second interval, beginning at 3 minutes. The final reading was taken at 10 minutes, which was the average from minute 3 to minute 10 (the first three minutes of testing were discarded).

PREDICTIVE EQUATIONS: (60)

Harris Benedict: (gold standard for adults)

Male: REE (kcal/day) = 66.47 + 13.75 (wt in kg) + 5 (ht in cm) – 6.8 (age in yrs)

Female: REE (kcal/day) = 665 + 9.6 (wt in kg) + 1.8 (ht in cm) – 4.7 (age in yrs)

Mifflin-St Jeor: (ADA recommended for Obese Adults)

Males: REE (kcal/day) = 10 (wt in kg) + 6.3 (ht in cm) – 5 x age + 5

Females: REE (kcal/day) = 10 (wt in kg) + 6.3 (ht in cm) – 5 x age – 161

Shofield HW: (an acceptable method for children)

Males (3-10yrs): REE (Kcal/day) = 19.6 (wt in kg) + 1.033 (ht in cm) + 414.9

Females (3-10 yrs): REE (Kcal/day)= 16.8 (wt in kg) + 1.618 (ht in cm) + 371.3

Males (10-18 yrs): REE (Kcal/day) = 16.25 (wt in kg) + 1.373 (ht in cm) + 515.5

Females (10-18): REE (Kcal/day) = 8.37 (wt in kg) + 4.65 (ht in cm) + 200

WHO equation (an acceptable method for children)

Males (0-3yrs): REE(kcal/day)= 60.9(wt in kg) - 54

Females (0-3yrs): REE(Kcal/day)= 61(wt in kg) - 51

Males (3-10yrs): REE (Kcal/day)= 22.7(wt in kg) + 495

Females (3-10yrs): REE (Kcal/day)= 22.5 (wt in kg) + 499

Male (10-18yrs): REE(Kcal/day)= 17.5 (wt in kg) + 651

Females (10-18yrs): REE (Kcal/day) = 12.2 (wt in kg)+ 746

IOM equation (ADA recommended for obese children)

Males (includes activity factor (PA) to give Total Energy Expenditure (TEE):

TEE= 114- (50.9 x age(yr) + PA x (19.5x wt (kg) +1161.4 x ht (m))

Females (includes PA to give TEE)

TEE = 389 – (41.2 x age (yrs) + PA x (15 x wt (kg) + 701.6 x ht (m))

Note: to compare equations the activity factor calculated was 1.0 or sedentary and was reflective of the pediatric population studied.

STATISTICAL ANALYSES: Data are presented as mean ± SE. Statistical analyses were performed using SPSS (61) using Pearson correlation for comparing MREE to each predictive equation. A 2-tailed significance paired sample T-test was also used to compare means.

RESULTS: Total sample size was 80 (46 female, 57 non-white) children with a mean age of 12.18 yrs, and mean BMI 37.39 (SE ± 1.07), > 99th Percentile BMI for age and gender. The mean average MREE (kcal/day) was 1977.13 (SE ± 49.97). Table 1 presents descriptive data as well as the mean average for each predictive equation. There was a strong and significant ($p<0.001$) correlation between MREE and all predictive equations. There was also a statistically significant difference ($p<0.001$) between the absolute means of these predictive equations and MREE (Figure 1). The smallest and only non-significant absolute mean difference was seen between MREE and the Harris Benedict equation (197.93 kcal/day SE ± 23.26) and the largest difference between MREE and the IOM equation (763.75 SE ± 76.62). When comparing MREE to all predictive equations by gender, there was a significantly ($p<0.05$) greater absolute

mean difference between MREE and predictive equations in boys vs. girls. This difference was not seen when comparing by race (Figures 2 and 3).

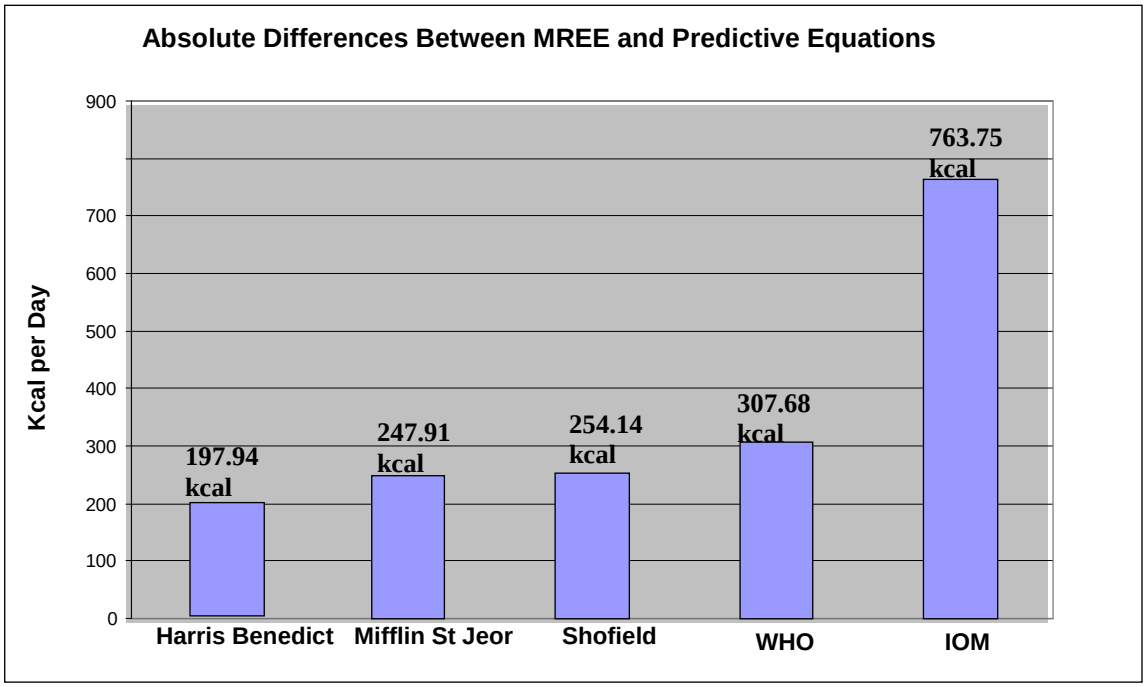
Table 1 Descriptive Data (n=80)

	Mean (Kcal/day)	Std Error (SE)	Pearson Correlation (with MREE)
Age	12.18	0.31	
BMI	37.39	1.07	
MREE	1977.13	49.97	
Harris Benedict	1950.41	50.22	0.795*
Mifflin	1800.64**	42.83	0.811*
Shofield	2057.41**	60.45	0.746*
WHO	2204.70**	61.70	0.739*
IOM	2738.93**	106.23	0.742*

*p-value <0.05

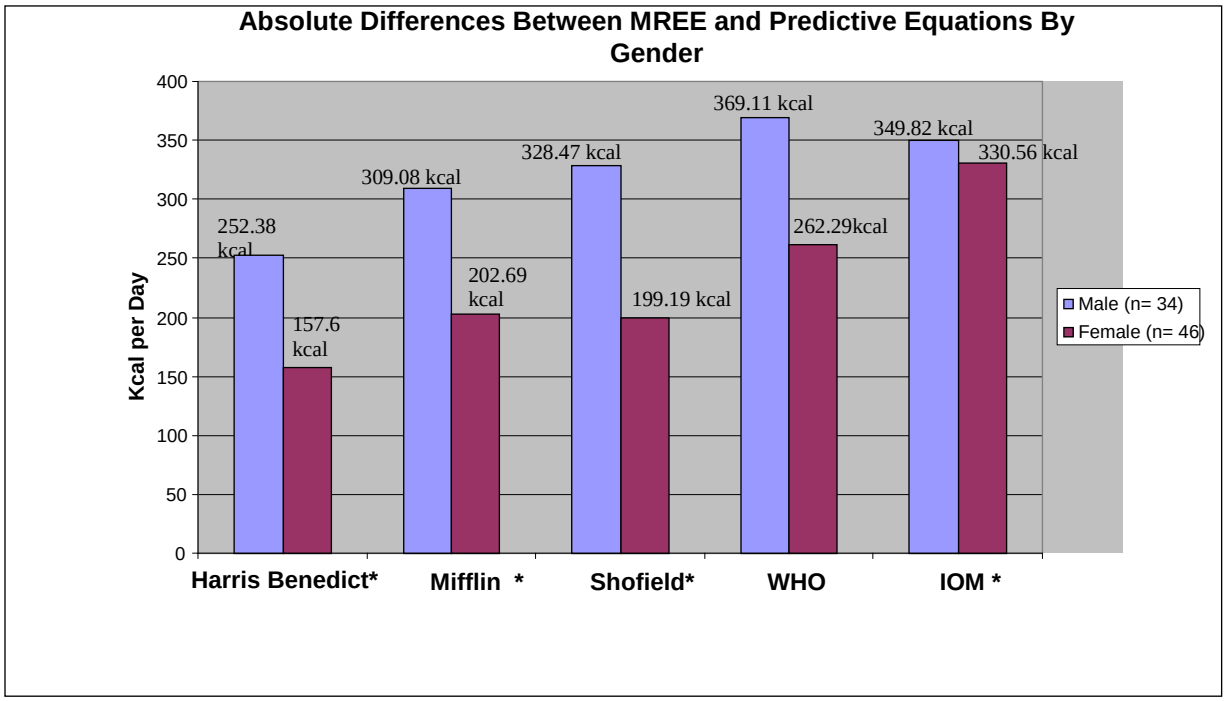
** p< less than 0.05; significant difference between the mean compared to MREE

Figure 1: Absolute Differences Between MREE and Predictive Equations: Harris Benedict, Mifflin St Jeor, Shofield, WHO, and IOM. (N= 80)



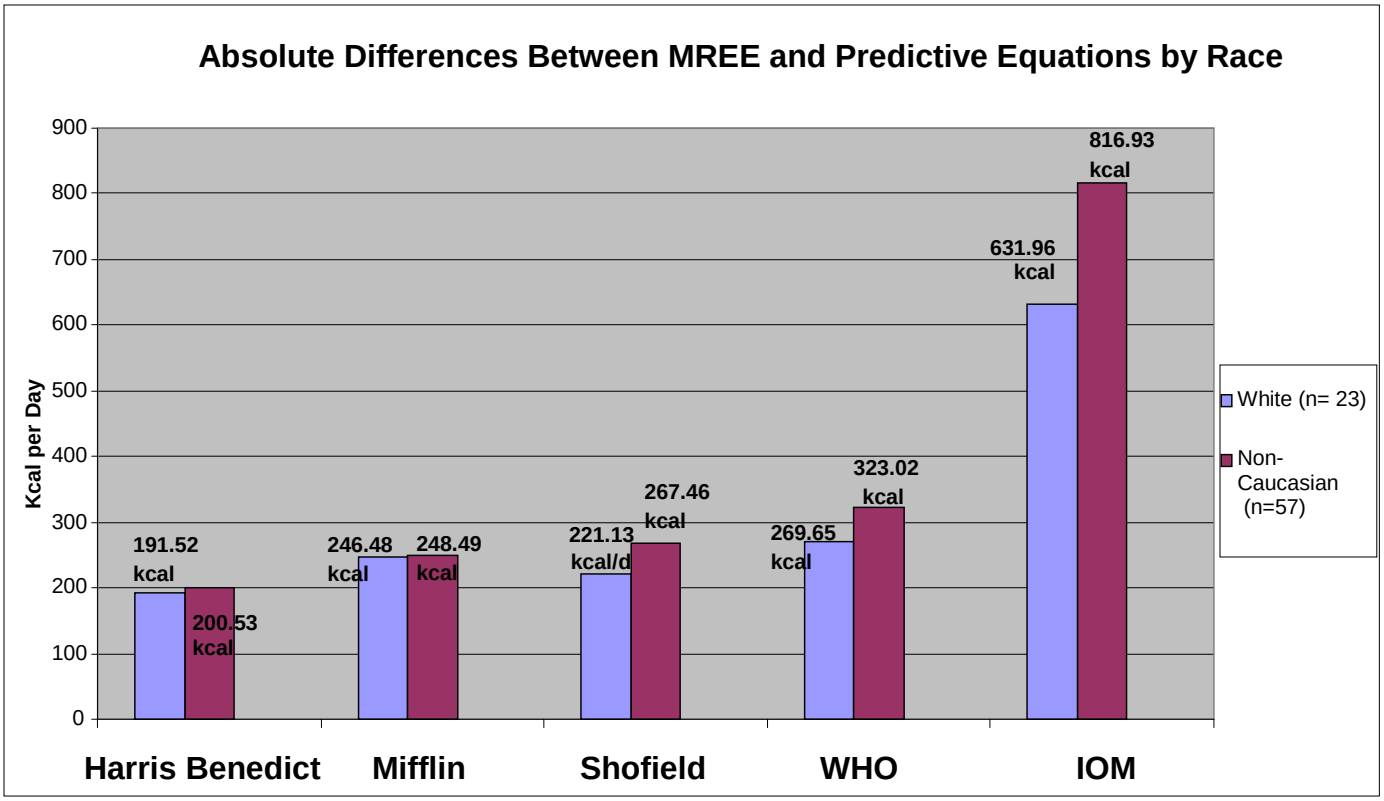
All Subjects: n = 80

Figure 2: Absolute Differences Between MREE and Predictive Equations by Gender: Harris Benedict, Mifflin St Jeor, Shofield, WHO, IOM. (N=80)



* Significant difference between genders ($p<0.05$)

Figure 3: Absolute Differences Between MREE and Predictive Equations by Race: Harris Benedict, Mifflin St Jeor, Shofield, WHO, IOM (n=80)



No Significant Differences between Races.

DISCUSSION: REE is the basis for determining energy needs for weight loss in an obese population. In the present study, REE was measured in obese youth and in the clinical setting with a portable indirect calorimeter. This measured REE was then compared to several commonly used predictive equations. To our knowledge, this is the first data to explore these relationships while utilizing indirect calorimetry in the clinical setting with an obese pediatric population, and in particular, comparing measured REE with the ADA recommended IOM equation. As other studies have shown, our data agrees that while there is a strong positive relationship between MREE and various predictive equations, there is also a significant discrepancy. In the clinical setting, even a few hundred calorie difference can make a significant impact in terms of setting caloric targets for weight loss. Our data indicates the smallest mean difference between MREE and predictive REE was with the Harris Benedict equation (197 kcal); where as the largest absolute difference between MREE and REE was using the IOM equation (763kcal). It is also important to note that when comparing mean differences between MREE and all predictive equations used, the Harris Benedict was the only equation to *not* be significantly different from MREE. Using a Caucasian female 16 yr old patient, the following provides a practical example of differences between MREE and predictive equations.

MREE: 1987kcal/day-

Harris Benedict: 1980kcal/day

Mifflin: 1964kcal/day

Shofield: 1945kcal/day

WHO: 2127kcal/day

IOM: 2633kcal/day

Recommended TEE for 2#/wk wt loss (Activity factor 1 for Sedentary, - 1000kcal/day)

MREE: 1200kcal/day (can not go below)

Harris Benedict: 1200kcal/day

Mifflin: 1200kcal/day

Shofield: 1200kcal/day

WHO: 1200kcal/day

IOM: 1600kcal/day

In this clinical example, if one were to use the IOM equation, there would be an approximate 400 kcal/day difference between measured energy expenditure and predicted energy expenditure. This could potentially indicate *weight gain* for an individual over time. Thus, this simple example indicates not only the usefulness of measuring REE in the clinical setting, it demonstrates the various discrepancies, and a gross over- estimation of energy needs in predictive equations.

While some investigators (21) have found that the Shofield equations are suitable for use with a mixed non-obese and obese pediatric population, others (25) have found that in obese adolescent boys, the Harris Benedict equation most closely predicts energy expenditure when compared to indirect calorimetry. Regardless, much of the literature is mixed and most agree that in general predictive equations typically over-estimate energy needs in an obese pediatric population. It is important to note that the WHO, IOM and Shofield equations have been developed based on healthy weight children (24). Given that the proportion of LBW to fat mass in lean children would be considerably higher, this may contribute to an overestimation of energy needs in an obese pediatric population. Some investigators such as Lazzer (23) have begun to try to develop equations based on an obese pediatric population; however, these are not

widely known or used at this point. There is a need to further investigate and develop predictive equations using an obese pediatric population. Thus consensus has not been reached in terms of the 'best' predictive equation to use and the general recommendation is becoming to utilize indirect calorimetry in the clinical setting. (15, 24)

The current study finds that there are significant differences by gender when comparing MREE to various predictive equations. On one hand it is known that MREE is generally higher in boys than in girls due to body composition (14, 18,); however, this is typically taken into account within each predictive equation. These data indicate that in general, predictive equations tend to over-estimate energy needs in obese boys more consistently than when compared with girls (i.e. 250-350kcal). However , there appears more variability between predictive equations and MREE in girls (150-350kcal/day). As stated earlier, variability in MREE could be due to various factors to include body composition, genetic influences, and race; however that discussion goes beyond the scope of this paper. When looking at differences in MREE and predictive equations by race, there does not appear to be a significant difference between Caucasian and African American (non white) subjects. This is somewhat surprising as several investigators have indicated that African American healthy and overweight children have a lower MREE than their Caucasian counterparts (27, 32) . Although data here do not compare MREE between races, given that most predictive equations have been developed using a healthy weight Caucasian population, one might expect to see these differences reflected when comparing MREE to these predictive equations. While some investigators have developed predictive equations to account for fat free mass (FFM), a major factor in determining REE, (14,16); recent findings state that they fair no better when compared to indirect calorimetry (24). Further

investigation may be warranted to assess if predictive equations should be utilized to account for race.

In general it appears that with this obese pediatric population, the Harris Benedict equation best predicts energy needs when compared to a portable indirect calorimeter for use in the clinical setting. Although, this may be surprising, as the equation was developed using a healthy weight, Caucasian, adult population; the equation does account for age, gender, height and weight. Most pediatric equations have been developed to also account for the growth needs within a specific age population (i.e. 3-10 yrs of age, 10-18 yrs of age). Perhaps in an obese pediatric population, and in this case a mostly morbidly obese population, energy needs for growth may be counter-productive in determining caloric targets for weight loss. Hence there is a propensity for the studied predictive equations to over-estimate energy needs. The Harris Benedict equation does not account for needs for growth, so perhaps is a logical predictive equation to use with obese children and adolescents when weight loss is often the goal.

To our knowledge, these are the first data to utilize a portable indirect calorimeter in the clinical setting with an obese pediatric population and compare this measured resting energy expenditure with the specific predictive equations named, particularly the IOM. The findings indicate that although a strong correlation with MREE, predictive equations tend to over-estimate energy needs in an obese pediatric population. These data support recommendations to utilize portable indirect calorimetry in the clinical setting to determine energy needs, and adds to a growing body of literature needed to demonstrate the usefulness of this clinical tool.

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CHAPTER 3: MEASURED RESTING ENERGY EXPENDITURE USING A HAND HELD INDIRECT CALORIMETER IN THE CLINICAL SETTING AS A PREDICTOR OF SUCCESS WITH WEIGHT LOSS IN AN OBESE PEDIATRIC POPULATION AFTER 3-6 MONTH FOLLOWUP AT A HEALTHY WEIGHT CLINIC.

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ABSTRACT: The literature has established measured resting energy expenditure (MREE) as a predictor of weight gain in adults (62) whereby a lower adjusted REE predicts greater weight gain over time in obese individuals. It is also known that heavier individuals tend to lose more weight and have a higher MREE. Body weight and body composition, particularly fat free mass (FFM) and skeletal muscle are major determinants of REE (31); however, recent literature suggests that fat mass is also a contributor to REE particularly in obese subjects (34). Very few studies have investigated MREE as a predictor of weight loss, and none have studied this in an obese pediatric population. The purpose of the current study was to test the hypothesis that a higher baseline MREE in obese youth will lead to greater success with weight change as

determined by BMI z score. The investigators also explored the relationship between metabolic factors such as leptin with MREE and if a relationship exists how this may affect weight status change.

Resting energy expenditure was measured via a portable indirect calorimeter. MREE was determined in obese youth aged 7-18 years who were referred to the East Carolina University Healthy Weight Clinic and who were greater than the 85th percentile BMI for age and gender.

The results indicated that MREE was not a significant predictor of success with weight change in youth aged 7-18yrs of age. Tanner stage and lean body were significant ($p < 0.05$) predictors in this age category. The results also suggest that when divided into two groups, those with a higher baseline HOMA (6.01 ± 0.92) tended to decline in BMI z score when compared to those with a lower HOMA (3.85 ± 0.65) ($p=0.081$). Data analyzed for Tanner Stage 5 youth as a group (n=17) indicate a strong, negative and significant ($p=0.03$) correlation between baseline fat mass and change in BMI z score (Pearson correlation -0.527) (Table 3) There was also a strong, negative correlation between baseline percent fat and change in BMI z score (Pearson correlation -0.525) (Table 3). When divided into groups, there was a significant difference between groups ($p<0.05$) in baseline leptin, fat mass and MREE normalized to fat mass. Those that decreased BMI z score (n=9) had the following baseline values: leptin (ug/L, n=6) 19.8 ± 4.76 (n=6), fat mass (kg) 41.23 ± 4.84, and MREE/fat mass (kcal/kg) 54.7 ± 6.28. Those with BMI z score incline (n=8) had the following baseline values: leptin (ug/L) 51.58 ± 13.67(n=5); fat mass (kg) 77.48 ± 15.94 , and MREE/fat mass (kcal/kg) 34.70 ± 5.60.

The main conclusions were that in youth aged 7-18 , MREE did not predict success with weight change status. It appears that in Tanner Stage 5 baseline leptin, fat mass, and percent fat

predicted change in BMI z score. Those in this pubertal category and who had a lower baseline fat mass and lower leptin concentration declined in BMI z score. Also, those Tanner stage youth who had a higher energy expenditure (kcal/kg), when MREE was normalized to fat mass, tended to decline in BMI z score. These data suggest there needs to be further exploration of the role of baseline leptin and its relationship to fat mass and MREE normalized to fat mass as predictors of change in weight status in this pubertal stage.

INTRODUCTION: MREE via indirect calorimetry provides the number of calories per day (kcal/day) an individual needs at rest to maintain body mass. To determine the individual's total energy needs (TEE), an activity factor takes into account the typical daily activity of the individual. This is important in various disease states- such as obesity to help determine an individual's energy needs for weight loss. REE is 30-40% genetically determined and 60-70% influenced by factors within the individual- such as body composition, age, gender and ethnicity. (29). It is difficult to "tease" out the various influences on an individuals' REE (nature vs. nurture); however the individual variances, particularly body composition has been the focus of much research. As early as the 1920s experiments have determined a relationship between REE and body mass of an individual. Later studies then indicated that indexing REE to fat free mass (FFM),which includes skeletal muscle, was a more accurate method to determine intergender differences in REE (i.e. men have more FFM than women). (30) This concept of "normalizing" MREE to some factor has become important in research so as to account for confounders such as gender, age, and ethnicity. Ways to normalize MREE would include kcal/ kg FFM, kcal/kg body weight (and/or BMI), and kcal/kg fat mass. The literature suggests that since skeletal muscle comprises approximately 40-50% body mass of most individuals and contributes to about 18-

20% total REE, FFM is often the most "conventional" way to normalize MREE (30,31). Most of these studies utilize the adult population. Very few studies have explored the relevance of adjusting REE in children, let alone obese children. Tershakovek A and associates (32) found that fat mass was an independent contributor to REE in obese children, and a recent review by Muller et al (34) indicates that the variability in REE as contributed by fat mass increases with increased adiposity. The authors also state that the effect of fat mass on REE is determined by fat depots, fat cell size and liquid droplet structure (34). and indicate that more research is needed in this area. Another study (63) explores a statistical model for normalizing REE to body mass and composition in children; but this has not been utilized in practical terms and in the clinical setting.

The importance and independent contribution of fat mass to REE in obese subjects and children has been postulated (32, 34); but not explored in the clinical setting. The mechanisms by which fat mass exerts its effects on metabolism is still unknown. However the hormone leptin has been proposed as a potential feedback signal from fat to skeletal muscle whereby exerting a thermic effect via uncoupling proteins (52, 64) and increased substrate cycling between de novo lipogenesis and mitochondrial lipid oxidation (65,66).

The purpose of this study is to: 1. test the hypothesis that MREE is a predictor of success with weight loss in an obese pediatric population undergoing treatment in the clinical setting; 2. to help determine the relevance of adjusting MREE to body weight and composition in terms of predicting outcomes in change in BMI z score and change in body weight after 3-6 month follow-up in a healthy weight clinic; and 3. to determine the relationship between baseline plasma leptin, fat mass and MREE and potential effects on success with weight loss as related to MREE in an obese pediatric population.

METHODS:

SUBJECTS: Youth aged 7-18 referred to the East Carolina Pediatric Healthy Weight Clinic, who were > the 85th percentile BMI for age and gender, who were fasting, and who were able to perform indirect calorimetry. Patients who fidgeted during testing, were unable to complete the test, or who were taking medications that could potentially alter metabolic rate were excluded. Baseline and follow up (3-6 months) height, weight, BMI and BMI z score were measured and calculated for each patient. Tanner stage was determined by the clinic MD. All measurements were recorded in the patient's electronic medical record.

LABORATORY TESTING: Blood was drawn at the Healthy Weight Clinic lab or a nearby affiliate lab from fasting subjects. Baseline leptin, insulin, cholesterol, glucose and thyroid function were measured at Pitt County Memorial Hospital (PCMH) Clinical Laboratories Greenville , NC using standardized procedures.

TANNER STAGE: Tanner staging, or pubertal stage, was determined by the MD. See Appendix 2 for categorization of Tanner Stage. (See Appendix 2)

INDIRECT CALORIMETRY: The KORR ReeVue portable indirect calorimeter. (http://www.korr.com/products/reevue.htm) (59) The device was set up in a clinic exam room designated for the sole purpose of testing. Calibrated to room STDP. Each patient was provided a new and separate disposable tubing, mouthpiece and nose clip. Testing was performed for ten minutes, or until accurate reading provided. The patient sat calmly and still in the quiet room, breathing as would normally with nose clip and mouthpiece. MREE (kcal/day) was indicated at the final reading taken at 10 minutes, which was the average of minute 3 to minute 10 , whereby the first 3 minutes of testing were discarded.

CALCULATION OF BODY SURFACE AREA AND BODY COMPOSITION: A clinical tool was utilized to estimate each subject's body composition utilizing an equation for body surface area (BSA). In pharmacology, equations such as the Mostellar Equation are often used to determine BSA in dosing medications, and based on being fat or water soluble. (54). A report by ThanhVu (55), Pharmacologist with the Cross Cancer Institute recommends the use of the Mostellar equation (compared to others), and discusses with references utilization of this equation in various populations- to include children and the obese. Thus, an online calculator: http://www.halls.md/body-surface-area/bsa.htm (56) which used the Mostellar equation was utilized to input each patient's height, weight, age and gender , and determine BSA and lean body weight (LBW). It is from this calculation that fat mass and percent body fat was calculated as such: LBW (kg) was subtracted from total body weight (TBW, kg) to give fat mass. This was then divided by TBW to give percent fat of each patient. Others (57) have also determined fat mass via this method/calculation.

THE PATIENT EXPERIENCE AT ECU'S HEALTHY WEIGHT CLINIC: At the initial visit, the fasting patient performed indirect calorimetry and had blood drawn from the clinic laboratory. The total initial visit (total 2-3hours) consisted of approximately an hour with the clinic MD addressing medical concerns, approximately an hour with a Registered Dietitian (RD) discussion nutrition history and goals, and as needed, addressing psychosocial aspects of the family with Medical Family therapy (MFT). The RD counseled subjects and their families utilizing the Medical Nutrition Therapy (MNT) Protocol developed by Pitt County, NC dietitians for helping children achieve a healthy weight. A detailed dietary intake and nutrition history was obtained and the counseling style was modeled after motivational interviewing. Nutrition goals were developed and tailored to the families' individual needs and stage of readiness to change.

Additionally, the initial visit typically focused on decreasing sugar sweetened beverages, using a 'stoplight' approach for incorporating healthier foods and grocery shopping, as well as utilizing the AAP's expert guidelines in a "Top 10 Tips for Helping Families and Children Achieve a Healthy Weight" handout. The MD not only addressed the medical concerns of the family, he/she promoted health behavior changes by targeting reduction in TV/sedentary time, and promoting physical activity with a pedometer given to the patient (and family members as appropriate).

The follow-up visit was performed by the same RD, was often 30-45 minutes, and reinforced goals previously made and reset new nutrition goals as appropriate. If the subject and family were at a stage of readiness, a focus of the follow-up visit was portion control in utilizing a divided plate approach/handout, and instruction on writing up a 2-3 food journal. At this follow-up visit, the MD also 'checked in' with the subject and family about lifestyle changes such as with increased physical activity, step counts with the pedometer, and decreased TV/screen time.

STATISTICAL ANALYSIS: Data are presented as mean ± SE. Statistical analyses were performed using SPSS (61.) Linear regression was performed for outcome measures BMI z score and weight change over time. Independent samples t-test was performed to compare mean values. Pearson correlation was performed to explore relationships between continuous variables. Significance was $p < 0.05$

RESULTS: When analyzing all subjects (n=80), there was a significant (p=0.001) positive relationship between MREE and body weight, lean body mass (LBM) and fat mass (FM) .

MREE and adjusted MREE (to kg body weight, LMB, FM) were not significant predictors of weight change as measured by BMI z score and change in weight in kg from baseline to follow-up. Tanner Stage and lean body weight were significant predictors of change in BMI z score (p= 0.044; 0.024) and weight change (kg) (p= 0.048; 0.008) from baseline to follow-up. HOMA was an independent predictor of weight change (kg) from baseline to follow up (p= 0.042).

Differences between groups regarding those whose BMI z score increased vs. those whose BMI z score declined were significant with respect to mean LnHOMA (p=0.039) and approached significance with mean HOMA (p= 0.096). Gainers had a lower mean HOMA and LnHOMA vs. those that declined in BMI z score (see Table 1) There were no significant differences in body weight (kg) between BMI z score losers or gainers.

Although not quite statistically significant (p= 0.061), there was a lower mean MREE in those who gained weight (1921.27 calories/day ± 54.06) and those who lost weight (2134.0 calories/day ± 109.99). There was also a difference (p= 0.047) when normalizing MREE to kg body weight (See Table 1)

When exploring gender differences, there was a difference (p=0.003) in mean LBW between boys (58.05 kg LBW ± 2.51) and girls (49.6kg LBW ± 1.43) but no differences in mean body weight and fat mass. There was also a significant (p=0.001) higher difference in baseline MREE between boys (2171 calories/day ± 87.44) and girls (1833 calories/day ± 48.94). There were no gender differences with regard to baseline mean leptin or mean HOMA concentrations or in mean change in BMI z-score or mean change in weight (kg). (see Table 2)

In terms of racial differences, there were no significant differences in mean LBW; however, there was a significant difference (p=0.046) in baseline mean body wt between non-Caucasian (103.15kg ± 4.40) and whites (87.83 kg ± 5.43). There was also a significant

difference (p=0.030) in mean fat mass between non-Caucasian (49.42 kg fat mass ± 4.36.) and Caucasian (33.72 kg fat mass ± 3.21). There were no racial differences in baseline mean MREE, mean HOMA, or in mean change in BMI z-score and mean change in weight (kg). Although not statistically significant there was difference (0.097) in baseline mean leptin between non-white (33.90 ± 3.35) and white (23.60 ± 4.46). (See Table 2)

Figure 1 demonstrates the difference ($p < 0.05$) between Tanner Stages 1-4 and Tanner Stage 5 whereby pubertal stages 1-4 increased in BMI z score and Stage 5 was the only group to decline in BMI z score.

When analyzing data for children in Tanner Stage 5 (n= 17) there was a negative correlation between change in BMI z score and fat mass (p= 0.030) as well as percent fat (p=0.030), and a negative correlation with baseline leptin concentration (p= 0.08; Table 3). There was not a difference between those that declined BMI z score (n= 6) and those that increased in BMI z score (n= 5) in terms of MREE, or MREE normalized to LBW. There was, however, a significant difference (p=0.033) between groups. The mean MREE/fat mass of those that increased BMI z score was 34.70 ± 5.60 calories/ kg fat mass while those that decreased BMI z score had a mean MREE/fat mass of 54.72 ± 6.28 calories/ kg (See Table 4). There was also a significant difference (p= 0.037) in baseline mean fat mass: those that had increased in BMI z score = 77.48 ± 15.94 FM (kg) vs. those that had decreased in BMI z score (41.23 ± 4.84 kg fat mass). Mean percent fat was also different (p=0.014) between the groups (BMI z score increase: 56.5 ± 6.13) vs. (BMI z score decreased: 38.72 ± 2.53) . A significant difference in baseline leptin (p=0.041) was evident between those that declined in BMI score who had a lower baseline leptin concentration (19.8 ± 4.76 ug/L) compared to those that increased BMI z score (leptin =51.58 ± 13.6 ug/L) (Table 4).

Table 1: Differences in Baseline MREE, Body Composition, and HOMA between Gainers and Losers: BMI z Score and Weight (kg)

	BMI z score	Mean Values (± SE)	Weight Change (kg)	Mean Values (± SE)
Body Weight (kg)	Gainer: 30 Loser: 50	93.83± 6.41 97.67± 4.07	Gainer: 59 Loser: 21	91.56 ± 4.01***** 109.33± 6.29
Lean Body Weight (kg)	Gainer: 30 Loser: 50	50.73± 2.24 54.21± 1.76	Gainer: 59 Loser: 21	49.83 ± 2.90***** 61.73 ± 1.37
MREE (kcal/day)	Gainer: 30 Loser: 50	1967.13± 81.70 1983.12± 63.79	Gainer: 59 Loser: 21	1921.27 ± 54.06 2134.05 ± 109.99 **(p = 0.061)**
MREE/body weight (kg)	Gainer: 30 Loser: 50	22.24 ± 0.82 21.08± 0.58	Gainer: 59 Loser: 21	22.07 ± 0.58* 19.9 ± 0.73
MREE/Fat Mass (kg)	Gainer: 30 Loser: 50	63.48± 5.40 54.84± 3.58	Gainer: 59 Loser: 21	61.07 ± 3.76 48.95 ± 4.05 **(p= 0.075)**
HOMA	Gainer: 26 Loser: 43	3.85 ± 0.65 6.01 ± 0.92 **(p= 0.081)**	Gainer: 51 Loser: 18	4.70 ± 0.67 6.32± 1.49
LnHOMA	Gainer: 26 Loser: 43	1.09 ± 0.14***** 1.48 ±0.11	Gainer: 51 Loser: 18	1.28 ± 0.10 1.55± 0.19

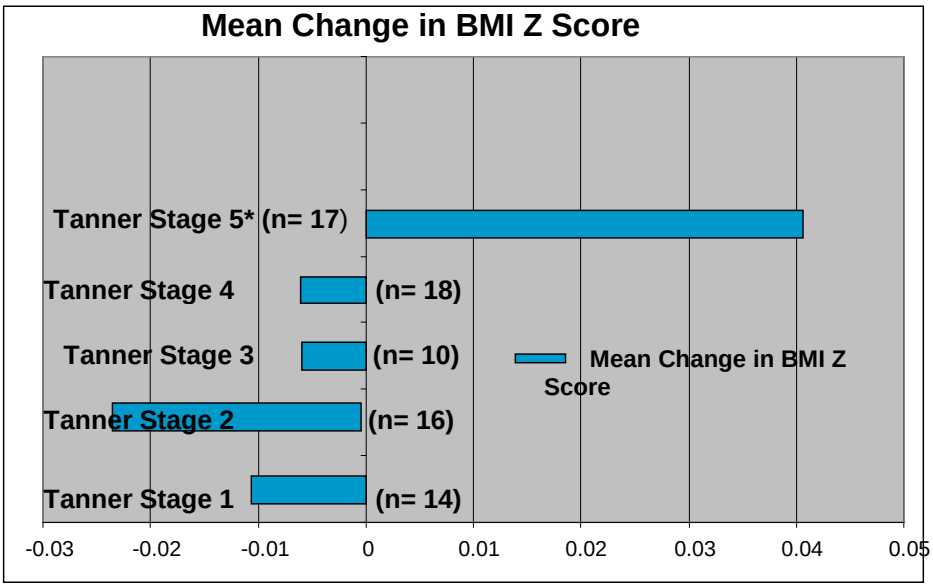
*** p= < 0.05; All Subjects (n= 80)**

Table 2: Gender and Racial Differences in Weight, Body Composition and Leptin –All Subjects (n= 80)

	Gender	Mean Values (± SE)	Race	Mean Values (± SE)
Body Weight (kg)	Boys: 34 Girls: 46	102.57± 6.17 91.54± 3.90	Non-Caucasian: 52 Caucasian: 23	103.15 ± 4.40***** 87.83 ± 5.43
Lean Body Weight (kg)	Boys: 34 Girls: 46	58.05 ± 2.51***** 49.6 ± 1.43	Non-Caucasian: 52 Caucasian: 23	53.92± 1.62 52.58± 2.98
MREE (kcal/day)	Boys: 34 Girls: 46	2171 ± 87.44***** 1833 ± 48.94	Non- Caucasian:52 Caucasian:23	2052.52± 57.61 1885.17±105.76
Fat Mass (kg)	Boys: 34 Girls: 46	45.64± 6.39 41.82± 3.31	Non-Caucasian:52 Caucasian: 23	49.42 ± 4.36***** 33.72 ± 3.21
Leptin	Boys: 23 Girls: 29	25.34± 4.05 32.66± 3.39	Non-Caucasian: 34 Caucasian: 14	33.90 ± 3.35 23.60 ± 4.46 **(p=0.09)**

*** p= < 0.05**

Figure 1: Differences in Change in BMI z Score by Tanner Stage (n=80)



***Significant Difference Between Tanner Stages 1-4 and Tanner 5 (p=0.013)**

Table 3: Correlations: Baseline Body Composition and Leptin with Change in BMI z score- Tanner 5 (n=17)

Parameter	Pearson Correlation	p -Value
Fat Mass (kg)	-0.527	0.030*
Percent fat	-0.525	0.030*
Leptin(ug/L)	-0.549	**0.080**

Table 4: Differences in Baseline Leptin, MREE, and Body Composition By Group: Decrease or Increase BMI z Score (Tanner Stage 5)

Parameter	Decrease (n=9)	Increase (n= 8)
Leptin* (ug/L)	19.8 ± 4.76 (n=6)	51.58 ± 13.67 (n=5)
Fat Mass (kg)*	41.23 ± 4.84	77.48 ± 15.94
Percent Fat*	38.72 ± 2.53	56.5 ± 6.13
MREE (kcal/kg fat mass)*	54.7 ± 6.28	34.70 ± 5.60

***p <0.05**

Table 5: Differences in Baseline MREE and Body Composition by Group: Decrease or Increase in BMI Z Score (Tanner Stage 5)

Parameter	Decrease (n=9) (SE)	Increase (n= 8) (SE)
MREE (kcal/day)	2083 ± 181.70	2214± 113.75
Weight (kg)	104.41 ± 7.78	131.76 ± 12.33
LBW (kg)	63.18 ± 4.35	52.5 ± 6.00
BMI*	34.52 ± 1.59	48.31 ± 5.56

***p <0.05**

DISCUSSION: The authors of this study investigated MREE in an obese pediatric population as a predictor of success with weight loss while undergoing treatment at a healthy weight clinic. It explores the hypothesis that a higher baseline MREE will contribute to more success with weight loss over time as measured by BMI z score and change in weight. In this study, MREE was not a predictor of weight change in obese youth aged 7-18 after 3-6 months of treatment at a healthy weight clinic. Predictors of change in BMI z-score were tanner stage and pre-intervention lean body weight (LBW) whereby on average, those in Tanner Stage 5 lost z score; while the majority in Tanner stages 1-4 gained z score (See Figure 1). Also, those with a higher LBW were more likely to lose BMI z-score and body weight. BMI z-score is a measurement that normalizes for age and gender, such that comparisons can be made among children. It is not surprising that LBW, which includes skeletal muscle mass, bone mass and organ mass, is a predictor of success of BMI z-score and weight change. Others (67) have found that LBW accounts for 60-70% of the variance MREE. In the current study, there was a significant ($p<0.05$) relationship between MREE and body weight, LBW and Fat mass (FM). In a linear regression model, each variable was a significant ($p <0.05$) contributor to MREE; however in a multiple regression model body weight remained the only significant predictor ($p=0.036$) of MREE.

In agreement with others (26, 67), the present study demonstrates gender differences such that boys have a significantly higher baseline MREE and LBW than girls. An interesting finding with this investigation is that, while African American (AA) obese children had a significantly higher baseline body weight compared to their Caucasian (C) counterparts, there were no racial differences in MREE. These AA children also had higher baseline fat mass and leptin levels, perhaps suggesting more leptin resistance within in this population. Similar baseline MREE is a finding that does not agree with others (32, 40), who have reported that AA obese children have

a lower MREE compared to obese white children. One explanation for the similar baseline MREE in our group may be that in this particular sample size, and in ages ranged from 7-18 yrs, there was no significant difference in LBW between the races (See Table 2).

Another interesting finding is that there was a close to significant (p=0.081) relationship between BMI z-score gainers and losers and HOMA**.** When analyzing date from all subjects, it appears that the losers had a higher baseline HOMA when compared to the gainers (see Table 1). This is similar to the findings of Cummings D and associates (69). The mechanisms are unclear as to why those with a greater insulin resistance would be more successful with weight loss; however, there may be an interplay between transcription factors present in adipose tissue (i.e. FOXC2), and perhaps proteins in the insulin signaling cascade which may affect energy expenditure (69,70).

In exploring differences between Tanner Stages, Tanner Stage 5 subjects on average were the only group to decrease in BMI z score (see Figure 1). Tanner Stage 5 subjects were also the only one as a group to lose BMI z-score and body weight; the difference in MREE is significant such that this group had a higher baseline MREE than when compared to the other groups combined. Given the higher body weight, and older age of the group, this higher MREE is not surprising. What is surprising is that in normalizing MREE to body composition, it appears fat mass is more significantly related to change in BMI z score than LBW. (Tables 4 and 5). This was explored further when investigating Tanner Stage 5 subjects only.

When evaluating Tanner Stage 5 subjects, significant negative correlations ($p < 0.05$) were noted between change in BMI z score from baseline to follow-up and FM as well as percent body fat. A close to significant (p=0.09) negative correlation was found between change in BMI z-score and baseline leptin concentrations (Table 3). These data support the connection others

have found between fat mass and leptin; where by it has been shown that greater fat mass lends itself to greater leptin concentrations (48). This finding would also be in agreement with a recently published study by Reinehr T et al (46) which indicates that leptin concentration is a predictor of overweight reduction in obese children participating in a lifestyle intervention. The main findings of Reinehr's study was that baseline leptin concentrations were correlated with gender, degree of overweight, and pubertal stage in obese children and significantly and negatively correlated with degree of weight loss.

When investigating differences between BMI z score gainers and losers, there was not a significant difference between the two groups in MREE (kcal/day), baseline kg body weight, or baseline LBW. Those in Tanner Stage 5 who had a lower baseline BMI declined in BMI z-score ($p=0.05$) as compared with those that had a higher BMI (Table 5). There were, however, differences ($p<0.05$) between these two groups in FM (kg); percent fat, MREE/FM, and leptin (Table 5). Those children with a higher FM, percent fat, and leptin tended to be 'gainers' (Table 4).

It is interesting that LBW was not significantly different between the groups, nor did it seem to predict change in weight or BMI z score. This could perhaps be a 'growth effect' whereby in older children the proportion of LBW to total body mass is less than in younger children when growth is occurring. This is also perhaps the result of a larger proportion of fat mass in these older children. Others (67) have noted that with age, LBW decreases, as does MREE. Perhaps in Tanner Stage 5, and in obese children, it is the ratio of skeletal muscle mass to fat mass decreases, and with an increased fat mass, the overall ratio of LBW to total body mass is decreased. As noted in Table 1, when investigating the difference in MREE/kg fat mass in all subjects, there was not a significant difference between those that increased BMI score and

those that declined in BMI z score. It appears that it is in older youth, adjusting MREE to fat mass may be more important in helping to predict success with weight change status.

In the current study, it appears that in Tanner Stage 5 youth, those with a lower baseline leptin declined in BMI z Score as compared to those with a higher baseline leptin (Table 4) Leptin is a hormone that acts centrally and peripherally in the body. The main function of Leptin is to decrease appetite and increase REE via centrally mediated mechanisms (48). In recent years it has been found that leptin also acts peripherally on tissues such as adipose and skeletal muscle (34, 65). Leptin resistance is the concept that high levels of leptin do not have the expected appetite suppressant and thermic effect and if often considered a common metabolic dysfunction seen in the obese state. (48). It appears in the current study, in older children, higher levels of leptin are negatively associated with BMI z score and weight change. This is similar to what Reinehr and associates (46) found in their study with lifestyle intervention and obese adolescents. There are a few proposed mechanisms by which leptin may act to affect weight change in the obese. In line with the concept of leptin resistance, perhaps on the central level leptin may not perform normally on the hypothalamic-pituitary-adrenal axis in decreasing appetite,. Wang et al (52) reported that leptin also may act on adipose tissue. These researchers found that in healthy rat tissue, leptin stimulated lipolysis within adipose tissue by evidence of release of glycerol, and increased mRNA of enzymes such as CPT-1 and acyl CoA oxidase, while down regulating fatty acid synthase. When compared to the ob/ob zucker rat, known as a model for 'leptin resistance' where the leptin receptor is defective, this lipolysis in adipose did not occur. Thus, perhaps in human adipose, when leptin resistance is present, lipolysis within adipose tissue is blunted, which in turn may contribute to overall outcomes of less weight loss. Finally- Solinas et al (65) demonstrate that leptin has an effect on skeletal muscle thermogenesis.

This interesting study postures that leptin mediates a cycle between de novo lipogenesis and lipid oxidation in skeletal muscle which requires PI3 kinase and AMP kinase. The authors show a link between glucose and lipid metabolism, whereby leptin may in a sense 'protect' skeletal muscle from excessive fat storage. In the case of obesity, where leptin resistance may occur, perhaps there is dysfunction in this mechanism such that via defective leptin receptors, or PI3/AMPK signaling, skeletal muscle thermogenesis is geared more towards lipid storage; hence decreasing the thermogenic affect of muscle on REE.

Both a limitation and strength of this study was that it was intended to demonstrate MREE utilized in the clinical setting, in 'real time'. These data could be used as an indication that further study by randomized clinical trial may be needed to explore the use of indirect calorimetry in obese youth, particularly in those that are nearing adulthood. Also in older obese youth, further exploration of the importance of baseline leptin and body composition, particularly FM, and the relationship to MREE and metabolic dysfunction is also warranted

Taken together the literature and the data presented in this study suggest that while MREE may not be a predictor of weight change status in obese youth aged 7-18, it may be a predictor in older obese youth, Tanner Stage 5. While lean body weight may be important in affecting MREE and outcomes related to change in weight, the metabolic affects of fat mass in this population can not be ignored. As seen in Tanner Stage 5 youth in this study, higher proportions of fat mass in obese youth may contribute to increased resting energy expenditure and quite possibly to metabolic defects, which in turn affect success with weight change as measured by BMI z score and weight loss.

CHAPTER 4: INTEGRATED DISCUSSION

As discussed in Chapter 1, childhood obesity is considered one of the greatest public health concerns of our time (2). Particularly in recent years, such organizations as the American Academy of Pediatrics(AAP) and the American Dietetic Association (ADA) have turned much of their attention to developing evidence based guidelines for prevention and treatment of this disease state (1, 58). A call for both mechanistic and clinical research in the field of childhood obesity has been made in the scientific and medical communities. One of the main impetuses for the current research described in this dissertation was to contribute findings of clinical work with an obese pediatric population, particularly in the area of measured resting energy expenditure (MREE) and outcomes after participation in a healthy weight clinic.

Chapter 1 discusses the importance of MREE in determining caloric targets for weight loss in obese adults and children in clinical and research settings (26, 67). The ADA has proclimated that measuring REE with indirect calorimetry in the clinical setting is the gold standard in the adult population, and has also considered this 'ideal' in the obese pediatric population (1). Current literature is beginning to shed light on the usefulness of indirect calorimetry with obese adults in the clinical setting (20, 68); however, there is currently very limited literature with obese youth. To our knowledge there is no literature that utilizes indirect calorimetry in the clinical setting with obese youth to compare this measurement with predictive equations or to describe outcomes of MREE while undergoing treatment in a 'real world' setting such as a healthy weight clinic.

The studies discussed in Chapters 2 and 3 describe the clinical outcomes of utilizing MREE in obese youth and the potential mechanisms involved in the results shown. The first

investigation compares MREE determined using a portable indirect calorimeter to 5 commonly used predictive equations that estimate REE and TEE. Several studies have demonstrated that predictive equations used for obese youth may compare to indirect calorimetry in the research setting, yet there is a significant discrepancy between predictive and measured REE (18,24). The literature is also still inconclusive as to the 'best' equation to use in an obese pediatric population. No studies have addressed the use of a fixed function calorimeter in comparison to these equations used in the clinical setting. Chapter 2 describes the differences between MREE using a portable device and 5 equations often used to calculate TEE and to set caloric targets for weight loss in obese youth. One unique aspect of this study is that it compares the 'gold standard' of predictive equations for obese youth as recommended by the ADA to other commonly used equations and to MREE. Two adult equations were included for two reasons: First, others (25) have shown MREE in obese youth in a research setting closely compares to the Harris Benedict equation (an adult 'gold standard"). Furthermore, we wanted to explore the 'gold standard' for obese adults, the Mifflin St Jeor equation, with youth who were , as a whole, considered "morbidly obese' (as measured by being well above the 99th percentile BMI for age and gender). The average BMI for the youth investigated was 39 kg/m^2, which is considered morbidly obese in an adult population. Given that several studies provide testimony to the fact that often predictive equations over-estimate (and under- estimate) energy needs in obese youth (18,24), the goal of Chapter 2 was to explore this in the 'real world' clinical setting with true application of these equations, and to compare these equations to MREE determined using a portable calorimeter.

Results as discussed in Chapter 2 indicate that indeed, all predictive equations, although significantly correlated with MREE, do also significantly deviate from MREE. The majority of

the predictive equations compared over-estimated the TEE of the obese pediatric population studied. In particular, the largest mean difference was seen between MREE and the IOM predictive equation for - boys and girls, with a mean absolute difference in energy expenditure of over 700 kcal/day. The overall smallest absolute mean difference was noted between MREE and the Harris Benedict equation (197kcal). Thus, in an obese pediatric population, the more 'accurate' predictive equation appears to be the Harris Benedict equation. However, the term 'accurate' is used loosely here, since even a 200 kcal difference in determining energy needs for weight loss can translate into a recommendation that leads to 1-2 pounds per month weight gain. As shown in the practical, real life example of comparing energy needs of a subject using all equations and MREE, there is room for error to either over- or under- predict energy needs. Particularly in obese youth it is very important to not over-predict needs- as shown with the use of the IOM equation, because that could potentially lead to a 1-2 pound weight gain over time. .

An important consideration when utilizing the common predictive equations for obese youth (WHO, Shofield, IOM) is that these equations were initially developed with healthy weight youth (24, 25)**.** Compared to their obese counterparts, lean youth may have a higher ratio of LBW to FM, such that an over-estimation of energy needs could occur when using these equations in a population that has a higher ratio of fat mass, which is metabolically less active compared to skeletal muscle. Another point to make is that the Harris Benedict equation was developed using healthy weight white males during the early 1900's (75). Although obese youth may have a higher ratio of fat mass than adult men, the Harris Benedict equation accounts for height, kg body weight age and gender. As the literature indicates, there is a strong correlation between kg body weight and REE (30). Perhaps obese youth have more similar total body

weights to adults, and more energy stores in fat mass such that energy needs for growth may not have to be taken into account

Other results presented in chapter 2 include gender differences such that there was more variability in girls when comparing predictive equations and MREE. The variability in differences between MREE and each predictive equation among females could be due to various factors to include body composition, genetic influences, and race. Interestingly, all equations tended to significantly overestimate REE in boys. Predictive equations account for age, gender, height and weight, with the assumption that boys have a different body composition than girls (i.e. more skeletal muscle); hence, the reasoning for separate equations for boys and girls. Perhaps in an obese population, the same 'assumptions' do not apply in that the 'ratio' of metabolic tissue is different in obese vs. morbidly obese vs. a healthier weight child/adolescent from which many of these equations have been developed. Another consideration is that predictive equations for children, in general, often account for the growth needs of development. The dichotomy in childhood obesity is that energy needs must consider meeting 'nutrient' needs for growing and developing youth, but our goal is actually for weight loss in obese individuals. Perhaps this is an explanation for the Harris Benedict, or an adult equation, more closely matching measured REE in this obese pediatric population. When looking at differences in MREE and predictive equations by race, there does not appear to be a significant difference between Caucasian and African American (non white) subjects. This is somewhat surprising as several investigates have indicated that African American healthy and overweight children have a lower MREE than their Caucasian counterparts (27,32). Although data here do not compare MREE between races, given that most predictive equations have been developed using a healthy

weight Caucasian population, one might expect to see these differences reflected when comparing MREE to these predictive equations.

The findings in Chapter 2 are unique in that not only does it demonstrate the usefulness of using indirect calorimetry in the clinical setting, it is the first to compare a clinical tool for measuring REE in obese youth to this specific subset of predictive equations: two commonly used pediatric equations (WHO and Shofield), two adult equations (Harris Benedict- gold standard for healthy adults; and Mifflin St Joer, ADA recommended for obese adults), and the ADA recommended IOM equations for youth (boys and girls). It is important to note also that although TEE is utilized to calculate caloric targets for weight loss: all equations, including the IOM which is based on TEE rather than REE, can be compared using the same activity factor. In this particular population an activity factor of "1" was used to denote the sedentary lifestyle of all participants. The IOM equation incorporates a physical activity factor in its calculation for TEE, while all other equations predict REE and then an activity factor is utilized to calculate TEE. Thus Chapter 2 demonstrates that when using the same activity factor to ultimately calculate TEE, the Harris Benedict equation most closely matches REE in obese youth as measured with indirect calorimetry. Most equations (WHO, Shoefield, IOM) over-estimated energy needs in this population; with the IOM equation grossly over-predicting. The Mifflin St Jeor, commonly used in an obese adult population, tended to slightly underestimate energy needs in this population in practice (see patient example in Chapter 2).

As more fully discussed in Chapter 3, variations in REE depend on factors such as age, gender, genetics, race, and body composition. While some investigators have developed predictive equations to account for fat free mass (FFM), a major factor in determining REE, (14,16), recent findings state that they fair no better when compared to indirect calorimetry (24).

Further investigation may be warranted to assess if predictive equations should be utilized to account for race. The study described in Chapter 2 contributes unique data regarding REE in an obese pediatric population as measured with a portable indirect calorimeter in the clinical setting. It also attempts to continue to elucidate the 'best' predictive equation to determine energy needs in this population. Suggestion for further investigation would be to continue to validate the use of indirect calorimetry in obese youth. While there is research to support the validity of portable indirect calorimetry in the clinical setting using both adults and youth (4, 53.), more research is needed, particularly in an obese and severely obese pediatric population. Additional research is also needed to build upon the literature of measuring REE in the clinical setting using a handheld portable device and comparing this to predictive equations. Both a strength and limitation of this current investigation is that data were collected in 'real time' and was intended as measurement of REE in the clinical setting rather than as a randomized clinical trial in a research setting where a metabolic cart would be used. As mentioned, further research will be needed in this area to strengthen the validity and reproducibility of indirect calorimetry use in the clinical setting(4).

Chapter 3 takes a more in depth look at outcomes related to MREE in obese youth undergoing treatment at a healthy weight clinic after 3-6 months follow-up. The overarching hypothesis was that MREE would be a predictor of success with weight change in this population, such that a higher baseline MREE would signify more weight loss after follow-up. It was also hypothesized that leptin, a metabolic marker related to fat mass in the obese would negatively correlate with MREE and with weight change status because of leptin resistance. It has been established in the literature that factors such as low MREE (62) and high leptin levels in obese children (45) are predictors of weight gain over time. What has not been investigated, is how baseline MREE, specifically in obese youth, contributes to success with

weight loss as measured by BMI z-score change and change in absolute weight. Adjusting MREE to body composition has been studied in adults, however, it has not been investigated in children, let alone in an obese pediatric population. Chapter 3 aims to answer the question whether a higher baseline MREE will lead to more success with change in BMI z score status in obese youth aged 7-18 yrs of age. We also wanted to answer the question if it was useful to adjust MREE to body weight and/or body composition. Finally, Chapter 3 explored a potential relationship between the metabolic marker of leptin with MREE and how this could potentially related to success with weight change status. The results indicate that, overall, MREE does not predict success with weight loss in obese youth aged 7-18 who participate in a healthy weight clinic. When analyzing the total sample of 80 subjects, the predictors of weight change as measured by BMI z score were lean body weight (LBW), and Tanner Stage. Older youth (Tanner Stage 5), and those youth with more LBW, had a tendency to lose BMI z-score. In some ways this is not surprising as those in Tanner Stage 5 also had higher baseline MREE and a higher LBW compared to the other Tanner Stage 1-4 youth. The results indicate that in terms of mean change in BMI z-score from baseline to follow-up, the Tanner Stage 5 group lost BMI z-score. One may assume, given a higher LBW in this group, that this would contribute to the higher MREE, since the literature indicates that LBW includes skeletal muscle which accounts for 50-60% of the variation in REE (31,67); however, although LBW significantly correlated with MREE, MREE alone was not an independent predictor of BMI z score change. This indicates that while skeletal muscle may account for a large variation in MREE, there is still 40-50% of REE that needs to be considered as attributed to other factors, such as fat mass. Particularly in an obese population, as others have indicated (32), fat mass can not be ignored and is an independent contributor to the variation of MREE.

When separating out BMI z-score losers and gainers, there was a difference (p= 0.09, although not statistically significant) in HOMA, such that BMI z score losers tended to have a higher baseline HOMA. There was a significant difference in LnHOMA between the two groups (Chapter 3, Table 1) This is in agreement with Cummings D et al (69). The investigators found that in a population of obese youth less than 18 years of age (n= 45) who received nutrition counseling on decreasing sweetened beverages, those who had a higher baseline HOMA , or were more insulin resistant responded better to dietary changes (i.e. decreased sugar sweetened beverages) in terms of loss in BMI z score. Although the mechanisms involved in this phenomenon are unclear, others (69,70) suggest that an upregulation of transcription factors present in adipose (i.e. FOXC2), may affect energy expenditure such that those with higher insulin resistance have greater weight loss after carbohydrate dietary modifications. It is important to note that all subjects in the present study did receive nutrition counseling at baseline on dietary modification such as decreased sweetened beverage intake. Perhaps this finding is more related to a behavior modification of decreased sweetened beverages. At baseline, many subjects were consuming a large amount of sugar beverages which may have resulted in elevated HOMA values. Perhaps, it is that after 3-6 months of decreased consumption of sweetened beverages contributed to a change in weight status, or a decline in BMI z score. This would need to be explored further, and further investigation would be needed to determine any relationship between dietary modification, baseline HOMA, and baseline MREE as related to change in BMI z score.

Since there appeared to be a significant difference in many variables with Tanner Stage 5 subjects (n= 17) compared to the other Tanner stages, further analysis was done in this group. One of the most interesting findings in this group, was that although there were no differences in

MREE between those that decreased vs. those that increased BMI z-score, there was a difference ($p < 0.05$) in MREE/FM. Another unexpected finding in this group , that is different when comparing to all subjects, is that FM and percent body fat were negatively correlated ($p < 0.05$) with change in BMI z score from baseline to follow-up. Furthermore, a close to significant negative relationship ($p=0.09$) appeared between BMI z-score change and baseline leptin., such that those with a lower baseline leptin lost BMI score. The significant variables when comparing BMI z-score gainers and losers were FM, percent body fat, MREE/FM, and baseline leptin ($p<0.05$ for all variables). It is important to note that there was not a significant difference in LBW between gainers and losers. This finding indicates that in Tanner Stage 5 youth, LBW does not appear to be predictor of weight change status; however, those with a lower baseline fat mass, and a higher calorie/kg fat mass ratio were more likely to decrease BMI z score.

The relationship between increased fat mass and higher baseline leptin levels in this group is not surprising given the established relationship between the two (71). What is striking, is that even 'lower' baseline leptin levels are much greater than expected levels for age: for example, a 'normal' average leptin for a 13 year old male is about 3.0 ng/ml (72), whereas, in this sample a 13 year old male had leptin levels of approximately 41 ng/ml. The 'reference range' (from 5^{th}->95^{th}) - for adult males is 0.7-5.3 ng/ml at a BMI of $22kg/m^2$ (74). The highest BMI on the adult leptin value chart is $35kg/m^2$, giving a range from 8.7-70.3 (5^{th}->95^{th}). Thus, the average Tanner Stage 5 youth not only had BMI's considered morbidly obese by adult standards (i.e. ave BMI 39), but had leptin values greater than the 50^{th} percentile for BMI, and over 10 times the 'normal' leptin value for a healthy weight youth.

Conclusions from analysis of the Tanner Stage 5 group indicate that fat mass is strongly negatively related to both BMI z score change and weight change, such that those with higher fat mass are less likely to have success with weight loss. A possible explanation is that with higher fat mass, leptin levels were also higher resulting from leptin resistance. Fat mass does indeed have its own resting metabolic effect (34). The 'thermic affect' of fat may be attributed to what is known as 'mass dependent secreting activity'- or the secretion of hormones such as leptin based on the amount of adipose tissue.(34) On one hand, investigators have demonstrated that those with higher fat mass also have higher MREE compared with lean counterparts (34). It is recognized that this higher REE is likely due to both a higher LBW and fat mass combined. In the present study it is noted that there is a significant difference in MREE between Tanner Stage 5 youth and those in other Tanner Stages. There is also a significant difference in body weight between these two groups (Tanner 1-4 vs. Tanner 5),; but the Tanner Stage 5 group also had a significantly higher LBW and FM. Thus it is feasible that both LBW and FM contribute to the higher MREE.

The 'metabolic effects' of adipose tissue, or perhaps better stated as 'dysfunction' of fat metabolism, may be better demonstrated when focusing on the Tanner Stage 5 youth. It is this group as a whole that tended to lose BMI z-score and weight. There may be several reasons for this observation. One may be that in Tanner Stages 1-4 there is a potential 'conflict' between the goals of a Healthy Weight program for weight loss in a population that is still 'growing 'in terms of bone, organ tissue and skeletal muscle. The 'natural metabolic drive' in the developing child is to gain weight in terms of body mass. Unfortunately, in the studied population these youth are already at a BMI with excess fat mass comparable to morbidly obese adults. Another explanation for more 'success' with weight loss (change in BMI z score) in the older group may also be

behavioral. In working with obese youth and lifestyle changes as defined by dietary and physical activity modification, the younger the child, the more dependent on the parent for environmental changes (i.e. access to high caloric foods). By the teen years, or Tanner Stage 5, there may be more 'choice' involved to make changes and these youth may be at a developmental stage to more independently make changes that contribute to weight changes.

As discussed in Chapter 3, when comparing BMI-z score, gainers vs. losers in Tanner Stage 5, it was indicated that fat mass levels strongly correlated with changes in BMI z score and weight, whereas LBW appeared to have less effect in terms of contributing to MREE and changes in weight. One explanation may be that in Tanner Stage 5, these youth are most metabolically closest to adult-hood, such that 'growth' as defined by metabolically active tissue with increased organ mass, and skeletal muscle (i.e. LBW) has slowed. Thus, as far as growth and development, there is more potential contribution from fat mass at this pubertal stage. In terms of adipose tissue metabolism, Muller et al (34) bring up the point that it is important to consider both adipocyte size as well as fat cell number. Adipocyte number is a major determinant of fat mass, which is then deposited differently (i.e. visceral vs. subcutaneous; abdominal vs. gluteal). Fat cell size and structure have implications with regard to mitochondrial oxidative metabolism which affects rates of lipolysis and energy expenditure (73). As one can ascertain, the metabolic consequences of excess fat, and in this case morbid obesity have yet to be truly be explained, and further exploration with studies that utilize detailed body composition analyses and cellular morphology are indicated for future research.

The mechanisms involved in fat mass and its effect on energy metabolism are still largely unknown; however leptin has been considered a prime 'candidate' (34). As discussed in Chapter 3, leptin's main function is to decrease appetite and increase REE via centrally mediated

mechanisms (48). The concept of leptin resistance such that high levels of leptin do not have the expected appetite suppressant and thermic effect is noted in obesity (48). The investigation as described in Chapter 3 demonstrates that, in older children (Tanner Stage 5), higher levels of leptin are negatively associated with BMI z-score and weight change. This is similar to what Reinehr and associates (46) found in their study with lifestyle intervention of obese adolescents. There are a few proposed mechanisms by which leptin may act affect weight change in the obese. In recent years it has been found that leptin also acts peripherally on tissues such as adipose and skeletal muscle (34,65). In line with the concept of leptin resistance, perhaps on the central level, leptin may not perform normally on the hypothalamic-pituitary-adrenal axis in decreasing appetite. Wang et al (52) reported that leptin may also act on adipose tissue. These researchers found that in healthy rat tissue, leptin stimulated lipolysis within adipose tissue, and increased mRNA of enzymes such as CPT-1 and acylcoA oxidase, while down regulating fatty acid synthase. When compared to the ob/ob zucker rat, known as a model for 'leptin resistance' where the leptin receptor is defective, this lipolysis in adipose did not occur. Thus, perhaps in human adipose, when leptin resistance is present, lipolysis within adipose tissue is blunted which in turn, may contribute to overall outcomes of less weight loss. Finally, Solinas et al (65) demonstrate that leptin has an effect on skeletal muscle thermogenesis. This interesting study proposes that leptin mediates a cycle between de novo lipogenesis and lipid oxidation in skeletal muscle which requires PI3 kinase and AMP kinase. The authors show a link between glucose and lipid metabolism whereby leptin may in a sense 'protect' skeletal muscle from excessive fat storage. In the case of obesity, where leptin resistance may occur, perhaps there is dysfunction in this mechanism such that via defective leptin receptors, or PI3/AMPK signaling, skeletal

muscle thermogenesis is geared more towards lipid storage; hence decreasing the thermogenic affect of muscle on REE.

No studies to date have investigated MREE in a morbidly obese pediatric population with the use of indirect calorimetry in the clinical or 'real world' setting, or in these terms as related to outcome measures such as change in BMI z-score or weight. This investigation is predicated on the hypothesis that MREE would be a predictor of success with weight loss in this population upon participation in a healthy weight clinic where lifestyle changes were taught.

Even while rates of childhood obesity are leveling out, the long term metabolic, social, and health consequences continue to be of great concern in the medical and scientific communities. The current project set out to investigate outcomes in the clinical and 'real world' setting with regard to MREE in an obese pediatric population participating in a healthy weight program in a rural southern community. The overarching hypothesis stated that those youth who had a higher baseline MREE would have more success with weight loss after follow-up with the postulation that MREE would be an independent predictor of outcomes such as change in BMI z score and weight. Overall, there was a negative finding such that in the combined group of all 80 subjects, MREE was not an independent predictor of weight change.

There were some important findings; however. It was determined that Tanner Stage and pubertal maturity was an independent predictor of weight change, and in particular there were different metabolic characteristics in the Tanner Stage 5 group. If one factors the relationship between fat mass and leptin, perhaps the most concise statement to describe the unique finding is baseline leptin concentration is negatively associated with change in BMI z score and weight change in older obese youth. A limitation to this study is that the sample size of Tanner Stage 5

youth was small (n=17). Further study is needed to expand upon the results indicated in this investigation.

In terms of MREE and the exploration of the usefulness in adjusting to body weight , fat mass and percent body fat, it appears that perhaps fat mass, not LBW may help predict success with weight change in older youth; although the metabolic factor may be more related to the higher leptin levels that tend to go along with higher ratios of fat mass. Important conclusions in Chapter 3 are that in older youth, those with a lower baseline leptin concentration and lower baseline fat mass were more likely to decline in BMI z score. Also MREE normalized to FM in this group indicated that those with a higher kcal/kg FM tended were more likely to decrease in BMI z score. This indicates that perhaps in older youth it may be beneficial to adjust MREE to fat mass to help predict success with weight change status. This also demonstrates that it may be important to take into account fat mass in older obese youth when determining energy needs. These data also suggest that baseline leptin levels may be a predictor of weight change status in Tanner Stage 5 youth. Further investigation is needed, and , as stated earlier, a limitation to this study is that the sample size in this group was small, and further investigation is required to expand upon these results.

Another important finding resulting from this project is the ability to report the use and usefulness of indirect calorimetry in the clinical setting using an obese pediatric population, particularly in morbidly obese youth. As stated previously, the ADA has encouraged the use of indirect calorimetry to determine caloric targets for weight loss in the obese. To date, there is no literature that describes the use of this clinical tool in obese children and adolescent and as compared to commonly used predictive equations. Not only does this research contribute a unique finding in terms of MREE measured in the clinical setting using an obese pediatric

population, it adds to a growing and needed body of literature that reports the usefulness of indirect calorimetry used in the clinical setting.

. This investigation prompts further research in obese youth in a variety of ways. It would be beneficial to add to the existing literature in terms of validating indirect calorimetry in this population as compared to the research setting using a metabolic cart, and perhaps with a control group of lean subjects. Also further research is needed to determine within subject variability in this population, as recommended by Cooper and associates (4). The current study also prompts further research particularly in an older obese population (Tanner Stage 5). It would be beneficial to investigate this population in terms of body composition using DEXA ,for example, and further exploring relationships between leptin concentration, fat mass and MREE.

As suggested by Muller M and associates (34) further exploration into fat metabolism as related to REE utilizing detailed body composition and cell morphology in this population would also help elucidate mechanisms involved in metabolic dysregulation that not only leads to the obese state in a pediatric population, but that which may have implications for success with weight loss. Also as previously stated the intent of this project was to report findings as developed in the clinical setting and in the medical home of these pediatric patients. Further research is needed in this realm as a randomized clinical trial, and with a control group such as healthy weight youth for comparison. This investigation may be utilized as a springboard of sorts to propel further research with obese youth in the clinical setting and who are participating in a healthy weight program.

In conclusion the important findings of this project are as follows: First, measuring resting energy expenditure (MREE) in the clinical setting via indirect calorimetry is particularly important in older youth- Tanner Stage 5. In this age group, it appears that adjusting MREE to fat mass may help predict weight change status in this population- such that those with a lower baseline fat mass may have more success with decline in BMI z score. A second finding is that when indirect calorimetry is not available in the clinical setting, the best predictive equation to use in obese youth > 99^{th} percentile BMI for age and gender is the Harris Benedict equation. Finally, based on the results in this project, both leptin and insulin appear to be involved in predicting success with weight change status in obese youth. The mechanisms related to leptin and insulin in influencing MREE and metabolic dysregulation need to be further explored.

REFERENCES

1. **ADA Evidence Library**. ADA Pediatric Weight Management Evidence Analysis Project. 2006, http://www.adaevidencelibrary.com/topic.cfm?cat=3010 Accessed: June 12, 2010

2. **Ogden, CL, Carroll, MD, Flegal, KM**. High body mass index for age among US children and adolescent, 2003-2006. *JAMA* 299 (20): 2401-2405, 2006.

3. **Orr.J** Evaluation of a novel resting energy metabolic rate measurement system. http://www.korr.com/products/reevue_clinical.pdf downloaded June 9, 2010

4. **Cooper JA, Watras AC, O'Brian MJ, Luke, A, Dobratz JR, Earthman CP, Shoeller DA.** Assessing validity and reliability of resting metabolic rate in six gas analysis systems. *J. Am. Dietetic Assoc.* 109 (1): 128-133, 2009

5. **Bosy-Westphal A, Reinecke U, Schlorke T, Illner K, Kutzner D, Heller M, Muller MJ.** Effect of organ and tissue masses on resting energy expenditure in underweight, normal weight and obese adults. *Int. J of Obesity*. 28:72-79, 2004

6. **Adair LS**. Child and adolescent obesity: epidemiology and developmental perspectives. *Physiology and Behavior*.94: 8-16, 2008

7. **Speiser PW, Rudolf MC, Anhalt H, Camacho-Hubner C, Chiarelli F, Eliakim A, Freemark M, Gruters A, Hershkovitz E, Iughetti L, Krude H, Latzer Y, Lustig RH, Pescovitz OH, Pinhas-Hamiel O, Rogol AD, Shalitin S, Sultan C, Stein D, Vardi P, Werther GA, Zadik Z, Zuckerman-Levin N, Hochberg Z; Obesity Concensus Working Group**. Childhood obesity. *J Clin Endocrinol Metab*. 90(3): 1871-1887,2005

8. **Libman I, Arslanian S**. Type 2 diabetes in childhood: the American perspective. *Horm. Res.* 59 (supplement1): 69-76, 2003

9. **Whitaker RC, Wright JA, Pepe MS, Seidel KD, Dietz WH**. Predicting obesity in young adulthood from childhood and parental obesity. *N Engl J Med.* 337(13): 869-873, 1997

10. **Wang S, Soni KG, Semache M, Casavant S, Fortier M, Pan L, Mitchell GA**. Lipolysis and the integrated physiology of lipid energy metabolism. *Molecular Genetics and Metabolism*. 95: 117-126, 2008

11. **Speakman J**. Obesity: the integrated roles of environment and genetics. *J of Nutr*. 134: 2090S-2105S, 2004

12. **Stiegler P, Cunliffe A**. The role of diet and exercise for the maintenance of fat free mass and resting metabolic rate during weight loss. *Sports Med*. 36 (3): 239-262, 2006

13. **Rodriguez G, Moreno LA, Sarria A, Pineda I, Fleta J,Perez-Gonzalez JM, Bueno M**. Determinants of resting energy expenditure in obese and non-obese children and adolescents. *J Physiol Biochem*. 58 (1): 9-15, 2002

14 **Derumeaux-Burel H, Meyer M, Morin L, Boirie Y.** Prediction of resting energy expenditure in a large population of obese children. *Am J. Clin Nutr*. 80: 1544-1550, 2004

15. **McDuffie JR, Adler-Wailes DC, Elberg J, Steinberg EN, Fallon EM, Tershakovek AM, Arslanian SA, Delaney JP, Bray GA, Yanovski JA.** Prediction equations for estimating energy expenditure in overweight and normal-weight black and white children. *Am J Clin Nutr* 80(2): 365-373, 2004

16. **Lazzer S, Agosti F, De Col A, Mornati D, Sartorio A**. Comparison of predictive equations for resting energy expenditure in severely obese Caucasian children and adolescents. *J Endocrinol Invest* 30 (4): 313-317, 2007

17. **Wong WW, Butte NF, Hergenroeder AC, Hill RB, Stuff JE, Smith EO.** Are basal metabolic rate prediction equations appropriate for female children and adolescents? *J Appl. Physiol*. 81 (6): 2407-2414, 1996

18. **Schmelzle H, Schroder C, Armburst S, Unverzagt S, Fusch C**. Resting energy expenditure in obese children aged 4 to 15 years: measured versus predicted data. *Acta Paediatr*. 93: 739-746, 2004.

19. **Amirkalaii B, Hosseini S, Heshmat R, Larijani B**. Comparison of Harris Benedict and Mifflin St. Jeor equations with indirect calorimetry in evaluation resting energy expenditure. *Indian J Med Sci.* 62 (7): 283-290, 2008

20. **Spears KE, Kim H, Behall KM, Conway JM.** Hand held indirect calorimeter offers advantages compared with prediction equations, in a group of overweight women, to determine resting energy expenditures during research screening. *J Am Diet Assoc.* 109:836-845, 2009

21**. Rodriguez G, Moreno LA, Sarria A, Fleta J, Bueno M.** Resting energy expenditure in chi;dren and adolescents: agreement between calorimetry and prediction equations. *Clinical Nutrition.* 21 (3): 255-260, 2000.

22. **Dietz WH, Bandini LG, Shoeller DA**. Estimates of metabolic rate in obese and nonobese adolescents. *J Pediatr*. 118 (1): 146-149, 1991.

23. **Lazzer S, Agosti F, De Col A, Sartorio A**. Development and cross-validation of prediction equations for estimating resting energy expenditure in severely obese Caucasian children and adolescents. *Br. J Nutr.* 95 (5): 973-979, 2006

24. **Hofsteenge GH, Chinapaw MJM, Delemarre-van de Waal HA, Weijs PJM**. Validation of predictive equations for resting energy expenditure in obese adolescents. *Am J Clin Nutr*. doi: 10.3945/ajcn.2009.28330, 2009.

25. **Schneider P, Meyer F**. Are basal metabolic rate prediction equations appropriate for overweight and obese adolescents? *Rev Bras Med Esporte*. 11 (3): 185e-188e, 2005

26. **Butte NF, Puyau MR, Vohra FA, Adolph AL, Mehta NR, Zakeri I**. Body size, body composition, and metabolic profile explain higher energy expenditure in overweight children. *J Nutr*. 137: 2660-2667, 2007

27. **DeLaney JP, Bray GA, Harsha DW, Volaufova J**. Energy expenditure in preadolescent African American and white boys and girls: the Baton Rouge Children's Study. *Am J Clin Nutr*. 75: 705-713, 2002

28. **Vogels N, Diepvens K, Westerterp-Plantenga MS**. Predictors of long-term weight maintenance. *Obesity Research.* 13 (12): 2162-2168, 2005.

29. **Bosy-Westphal A, Wolf A, Buhrens F, Hitze B, Czech N, Monig H, Sleberg O, Settler U, Pfeuffer M, Schrezenmeir J, KrawczakM, Muller MJ**. Familial influences and obesity-associated metabolic risk factors contribute to the variation in resting energy expenditure: the Kiel Obesity Prevention Study. *Am J Clin Nutr.* 87: 1695-1701, 2008

30. **McArdle W, Katch F, Katch V**. Exercise Physiology, 5th ed. *Energy Nutrition and Human Performance,* 2001

31. **Zurlo F, Larson K, Bogardus C, Ravussin E.** Skeletal muscle metabolism is a major determinant of resting energy expenditure. *J of Clin.Invest.* 86: 1423-1427, 1990.

32 **Tershakovec AM, Kuppler KM, Zemel B, Stallings VA**. Age, sex, ethnicity, body composition, and resting energy expenditure of obese African American and white children and adolescents. *Am J Clin Nutr*. 75: 867-871, 2002

33. **Sun M, Gower BA, Bartolucci AA, Hunter GR, Figueroa-Colon R, Goran MI**. A longitudinal study of resting energy expenditure relative to body composition during puberty in African American and white children. *Am J Clin Nutr.* 73: 308-315, 2001.

34. **Muller MJ, Bosy-Westphal A, Later W, Haas V, Heller M**. Functional body composition: insights into the regulation of energy metabolism and some clinical applications. *European Journal of Clinical Nutrition.* 63: 1045-1056, 2009.

35. **Sweeting HN.** Review: Gendered dimensions of obesity in childhood and adolescence. *Nutrition Journal.* 7:1 doi:10.1 186/1475-2891-7-1, 2008.

36. **Lazzer S, Boirie Y, Montaurier C, Vernet J, Meyer M, Vermorel M**. A weight reduction program preserves fat free mass but not metabolic rate in obese adolescents. *Obesity Research.* 12(2): 233-240, 2004.

37. **Vogels N, Westerterp-Plantenga MS**. Successful long-term weight maintenance: a 2-year follow-up. *Obesity*. 15 (5): 1258-1266, 2007.

38. **Luke A, Dugas L, Kramer H**. Ethnicity, energy expenditure and obesity: are the observed black/white differences meaningful? *Curr Opin Endocrinol Diabetes Obes* 14: 370-373, 2007

39. **Morrison JA, Alfaro MP, Khoury P, Thornton BB, Daniels S.** Determinants of resting energy expenditure in young black girls and young white girls. *J of Pediatr*. 129 (5): 637-642, 1996.

40. **Kaplan AS, Zemel BS, Stallings VA**. Differences in resting energy expenditure in prepubertal black children and white children. *J Pediatr*. 129: 643-647, 1996

41. **Lee S, Arslanian SA.** Fat oxidation in black and white youth: a metabolic phenotype potentially predisposing black girls to obesity. *J Clin Endocrinol Metab*. 93 (11): 4547-4547, 2008

42. **Enriori PJ, Evans AE, Sinnayah P, Cowley MA**. Leptin resistance and obesity. *Obesity.* 14(S): 254S-258S, 2006.

43. **Liuzzi A, Savia G, Tagliaferri M, Lucatoni R, Berselli ME, Petroni ML, Medici CD, Viberti GC**. Serum leptin concentration in moderate and severe obesity: relationship with clinical, anthropometric and metabolic factors. *Int. J Obes*. 23: 1066-1073, 1999

44. **Niskanen L, Haffner S, Karhunen LJ, Turpeien AK, Miettinen H, Uusitupa MI.** Serum leptin in relation to resting energy expenditure and fuel metabolism in obese subjects. *Int J Obes Relat Metab Disord*. 14: 309-313, 1997

45. **Savoye M, Dziura J, Castle J, DiPietro I, Tamborlane WV, Caprio S**. Importance of plasma leptin in predicting future weight gain in obese children: a two-and-a-half-year longitudinal study. *Int J of Obes.* 26: 942-946, 2002

46. **Reinehr T, Kleber M, DeSousa G, Andler W**. Leptin concentrations are a predictor of overweight reduction in a lifestyle intervention. *Int J Pediatr Obes*. 4: 215-223, 2009

47. **Muoio D, Dohm GL**. Peripheral metabolic actions of leptin. *Best Practice and Research Clin. Endocr Metab*. 16 (4): 653-666, 2002

48. **Margetic S, Gazzola C, Pegg GC, Hill RA**. Leptin: a review of its peripheral actions and interactions. *Int. J Obes*. 26: 1407-1433, 2002

49. **Muoio DM, Dohm GL, Tapscott EB, Coleman RA.** Leptin opposes insulin's effects on fatty acid partitioning in muscles isolated from obese ob/ob mice. *Am J Physiol.* 276 (5 pt 1): E913-921, 1999

50. **Muoio DM, Dohm DL, Fiedorek FT Jr, Tapscott EB**, Coleman RA. Leptin directly alters lipid partitioning in skeletal muscle. *Diabetes* 46(8): 1360-1363, 1997

51. **Minokoshi Y, Kim YB, Peroni OD, Fryer LG, Muller C, Carling D, Kahn BB**. Leptin stimulates fatty acid oxidation by activation AMP-activated protein kinase. *Nature* 415 (6869): 338-343, 2002.

52**. Wang M, Lee Y, Unger RH.** Novel form of lipolysis induced by leptin. *J Biological Chemisty*. 274 (25): 17541-17544, 1999

53. **Nieman DC, Austin MD, Chilcote SM, Benezra L**. Validation of a new handheld device for measuring resting metabolic rate and oxygen consumption in children. *Int J Sport Nutr Exerc Metab.* 15 (2): 186-194, 2005

54. **Mosteller RD**. Simplified calculation of body-surface area. *N Eng J Med*. 317(17): 1098, 1987

55. **Thanh V**. Standardization of body surface area calculations. 2008 http://www.halls.md/bsa/bsaVuReport.htm, Accessed June 12, 2010

56. Halls SB. Body surface area calculator for medication doses. http://www.halls.md/body-surface-area/bsa.htm Accessed June 17, 2010.

57. **LazzerS, Bedogni G, Agosti F, De Col A, Mornati D, Sartorio A**. Comparison of dual-energy x-ray absorptiometry, air displacement plethysmography and bioelectrical impedence analysis for the assessment of body composition in severely obese Caucasian children and adolescents. *British J of Nutr*. 100: 918-924,2008

58. **Barlow SE, Expert Committee**. Expert committee recommendations regarding the prevention, assessment, and treatment of child and adolescent overweight and obesity: summary report. *Pediatrics* 120 Suppl 4: S164-192, 2007.

59. **KORR- ReeVue Indirect Calorimeter**. http://www.korr.com/products/reevue.htm Accessed June 12, 2010

60. Theberge C. The Nutrition and Food Web Archive, 2009. http://www.nafwa.org/clinical_calculators.php. Accessed June 17, 2010

61. **SPSS,** Chicago Illinois, *Version 17*, 2009

62. **Ravussin E, Gautier JF**. Metabolic predictors of weight gain. *Int J Obes*. 23 Suppl: 37-41, 1999

63. **Zakeri I, Puyau M, Adolph AL, Vohra FA, Butte N.** Normalization of energy expenditure data for differences in body mass or composition in children and adolescents. *J Nur*. 136: 1371-1376, 2006

64. **Porter RK, Andrews JF**. Effects of leptin on mitochondrial 'proton leak' and uncoupling proteins: implications for mammalian energy metabolism. *Proc Nutr Soc*. 57 (3): 455-460, 1998

65. **Solinas G, Summermatter S, Mainieri D, Gubler M, Pirola L, Wymann MP, Rusconi S, Montani JP, Seydoux J, Dulloo AG.** The direct effect of leptin on skeletal muscle thermogenesis is mediated by substrate cycling between de novo lipogenesis and lipid oxidation. *FEBS Letters* 577: 539-544, 2004

66. **Summermatter S, Mainieri D, Russell AP, Seydoux J, Montani JP, Buchala A, Solinas G, Dulloo AG.** Thrifty metabolism that favors fat storage after caloric restriction: a role for skeletal muscle phosphatidylinositol-3-kinase activity and AMP-activated protein kinase. *FASEB J.* 22(3): 744-785, 2008

67. **Lazzer S, Bedogni G, Lafortuna C, Marazzi N, Busti C, Galli R, DeCol A, Agosti F, Sartorio A**. Relationship between basal metabolic rate, gender, age, and body composition in 8,780 white obese subjects. *Obesity*. 18: 71-78, 2010

68. **Ziegler J, Rothpletz-Puglia P, Touger-Decker, R, Byham-Gray L, Maillet J, Denmark, R.** Resting energy expenditure in overweight and obese adults. Agreement between indirect calorimetry and predictive formulas. *Top Clin Nutr*. 25 (2): 180-187, 2010

69. **Cummings DM, Henes S, Kolasa K, Olsson J, Collier D**. Insulin resistance status. Predicting weight response in overweight children. *Arch Pediatr Adolesc Med*. 162 (8): 764-768, 2008

70. **Cornier MA, Donahoo WT, Periera R, Gurevich I, Westergren R, Enerback S, Eckel PJ, Goalstone ML, Hill JO, Eckel RH, Drazin B.** Insulin sensitivity determines the effectiveness of dietary macronutrient composition on weight loss in obese women. *Obes Res*. 13 (4): 703-709, 2005

71. **Meier U, Gressner AM.** Endocrine regulation of energy metabolism: review of pathobiochemical and clinical chemical aspects of leptin, ghrelin, adiponectin, and resistin. *Clinical Chemistry* 50(9): 1511-1525, 2004.

72. **Garcia-Mayor RV, Andrade MA, Rios M, Lage M, Dieguez C, Casanueva FF**. Serum leptin levels in normal children: relationship to age, gender, body mass index, pituitary-gonadal hormones, and pubertal stage. *J Clin Endocrinol Metab*. 82 (9): 2849-2855, 1997

73. **Puri V, Czech MP**. Lipid droplets: FSP27 knockout enhances their sizzle. *J Clin.Invest*. 118: 2693-2696, 2008

74. **Scholz GH, Englaro P, Thiele I, Scholz M.** Dissociation of serum leptin concentration and body fat content during long term dietary intervention in obese individuals. *J Hormone and Metabolic Research.* 28(12): 718-723, 1996

75. Frankenfield D, Roth-Yousey L, Compher C. Comparison of predictive equations for resting metabolic rate in healthy non-obese and obese adults: a systematic review. *JADA* 105 (5): 775-789, 2005

APPENDIX 1

UNIVERSITY AND MEDICAL CENTER INSTITUTIONAL REVIEW BOARD
HUMAN BIOMEDICAL CONTINUING REVIEW OR STUDY CLOSURE

*****Note: Each section should be completed regardless of whether this form is being submitted for continuing review or closure of a research study.*****

DEMOGRAPHIC INFORMATION

UMCIRB Number: 07-0618 **Date this form was completed:** 9.10.09

Title of research (this title must match protocol, consent form and funding application, if applicable): Medical Nutrition Therapy for Overweight Youth: An Outcomes Study

Principal Investigator, credentials, department, section and school: Kathryn Kolasa PhD, RD, LDN Professor and Associate Director for Dietary Interventions, Pediatric Healthy Weight Research and Treatment Center, Brody School of Medicine; David Collier, MD, PhD, Department of Pediatrics; Adjunct Faculty to Department of Family Medicine, Brody School of Medicine; Director Pediatric Healthy Weight Research
and Treatment Center (PHWRTC).

Subinvestigators, credential, department, section and schools: John Olsson, MD, Department of Pediatrics, Brody School of Medicine; Suzanne Lazorick, MD. Department of Pediatrics, Brody School of Medicine. Sarah Henes, MA, RD, LDN. Department of Pediatrics, Brody School of Medicine. Cara Jenkins, MPH, RD, LDN KIDPOWER Dietitian, Department of Pediatrics, Brody School of Medicine ; Doyle Cummings, Pharm D, Family Medicine, Research Division, Brody School of Medicine ; Susan Morrissey, MA Family Medicine, Research Division, Brody School of Medicine; Allison Spain, BS Exercise and Sport Science Wellness Program Specialist, Viquest Exercise Programming; Keeley Pratt, MS, Family Therapy Associate, Department of Child Development and Family Studies, East Carolina University; Kay Craven, RDLDN, Family Medicine, Brody School of Medicine

ITEMS FOR APPROVAL

☒ **Research study being submitted for renewal.**

Version of the most currently approved protocol: October, 2007
Version of most currently approved consent document: September, 2007
List all other items that are currently approved (i.e. advertisements, questionnaires, study measures, etc.) and need to be re-approved for new approval period. Listing these items enhances the renewal process to make sure all research items required to conduct the research study will be re-approved: Cara Jenkins is KIDPOWER dietitian.

☐ **No items need to be approved since study is being closed.**

INVESTIGATOR QUALIFICATIONS

☒ **Research study being submitted for renewal.**

Provide the date of completion for the UMCIRB Human Subjects Protections training modules for the principal investigator, any subinvestigators and coordinators if this study is being renewed (must be current within 3 years):

Kolasa 8/2008
Olsson: 7/09/2007 Jenkins: 9/12/2008
Cummings: 6/2008 Collier: 9/2008
Lazorick: 4/2008 Henes: 9/10/2009
Spain: 9/10/09 Pratt: 01/2008
Morrissey: 7/28/08 Craven: 12/01/08

☐ **No UMCIRB Human Subjects Protection training modules information is necessary since study is being closed.**

Have there been any changes in your credentialing, licensure, certifications or privileges since the last continuing review? ☐ **yes** ☒ **no If yes, describe.**

SOURCE OF FUNDING:

☐ **No source of funding exists for this research**
☒ **Institution or Department Sponsor, Name:** Pitt Memorial Hospital Foundation
☐ **Government Agency, Name**:
☐ **Private Agency, Name**:

Fund number for IRB fee collection (applies to continuing review of all for-profit, private industry or pharmaceutical company sponsored projects):

Fund	Organization	Account	Program	Activity (optional)
		73059		

CHECK ALL INSTITUTIONS OR SITES WHERE THIS RESEARCH STUDY WILL BE CONDUCTED:

☒ **East Carolina University**
☒ **Pitt County Memorial Hospital, Inc**
☐ **Heritage Hospital**
☐ **Beaufort County Hospital**
☐ **Carteret General Hospital**
☐ **Boice-Willis Clinic**
☒ **Other** Pediatric and Family Practice Clinics in Pitt County.

AMENDMENTS / REVISIONS / MODIFICATIONS

☒ **There have been no amendments, revisions or modifications to the research protocol since the last review.**
☐ **Yes, there have been amendments, revisions or modifications since the last continuing review. Attach the UMCIRB revision form for any revision that is being considered for approval along with this continuing review. List the title or reference each item including version and UMCIRB approval date.**

☒ **There have been no amendments, revisions or modifications to the consent document since the last review.**

☐ **Yes, there have been amendments, revisions or modifications to the consent document since the last continuing review. Attach the UMCIRB revision form for any revision that is being considered for approval along with this continuing review. List the title or reference for each the item including version date and UMCIRB approval date.**

☐ **This is not a grant funded study.**
☒ **There have been no amendments, revisions or modifications to the grant since the last review.**
☐ **Yes, there have been amendments, revisions or modifications to the grant since the last continuing review.**
Attach a copy of the updated grant application with changes outlined or highlighted.

PARTICIPANT ACTIVITY

Sample size proposed in the research at all sites Open Enrollment- for all new obese patients seen for Medical Nutrition Therapy in the pediatric and family practices in Pitt County- approx n~ 300.

Total number of participants enrolled at all research sites to date
0

Total number of participants enrolled at this site since the research was initially approved
0

Total number of participants enrolled at this site since the last continuing review
0

Total number of participants completing all aspects of research at this site since the last review
0

Total number of participants involved in the follow-up portion of the research at this site
0

Total number of participants remaining in the active portion of the research
0

Total number of deaths at this site during the active or follow-up portion of this research to date
0

Is this research study followed by a Data Monitoring committee
☐ **yes** ☒ **no**

Total number of participants locally withdrawn prior to research completion
0

Provide specific details regarding all participant withdrawals from the research study, whether voluntary or initiated by the investigator.

Describe any difficulties in participant enrollment, specifically if the enrollment goals have not been reached as originally outlined. Describe the impact this will have on the study. Availability of Follow-up with the Community Dietitian (KIDPOWER), and patients lost to follow-up, may limit utilization of the 7-visit Medical Nutrition Therapy Protocol

If you have exceeded the sample size initially proposed for this research study, provide a rationale. Not Applicable

MONITORING AND ONGOING ACTIVITIES

☒ **There have been no locally occurring serious adverse events or events resulting in unanticipated risks to participants or others since the last review.**
☐ **Yes, there have been locally occurring serious adverse events or events resulting in unanticipated risks to participants or others since the last review. Attach an Adverse Event Reporting Form for any previously unreported serious adverse events. Applicable serious adverse**

events that have previously been reported should be listed by referring to the study participant code/number, date of event, type of event, and date submitted to the IRB office.

☒ There have been no protocol deviations/violations for this research study since the last review.
☐ Yes, there have been protocol deviations/violations for this research study since the last review. Attach a Protocol Deviation Form for any previously unreported protocol deviations/violations. Any protocol deviations/ violations that have previously been reported should be listed by referring to the study participant code/number, date of event, type of event, and date submitted to the IRB office.

☒ There have been no regulatory auditing activities or monitoring visits by a sponsor, institutional officials or outside agency since the last review.
☐ Yes, there have been regulatory auditing activities or monitoring visits by a sponsor, institutional officials or outside agency since the last review. Attach a report of these activities if the outcome was unfavorable or unacceptable. List the auditor/monitor (sponsor, institution, federal agency) and date of the activity.

☒ There has been no analysis or reports by a data monitoring committee since the last review.
☐ There has been an analysis or report by a data monitoring committee since the last review. Attach the report to the continuing review form if not previously submitted. If this report has been previously submitted to the UMCIRB, list that date.

☐ There have been no publications or presentations generated from the local investigator involved in this research since the last review.
☒ There have been publications or presentations generated from the local investigator involved in this research since the last review. List all publications or presentation resulting from information generated by this research, generated by local investigators or sponsors. Attach the published materials to the continuing review form.

☒ There have been no new developments generated by this research that have an impact on the assessment of potential risks or benefits for participation in this research study since the last review.
☐ There have been new developments generated by this research that have an impact on the assessment of potential risks or benefits for participation in this research study since the last review. Describe these new developments.

☒ There are no additional comments or information that may be pertinent to the review of this research.
☐ There are additional comments or information that may be pertinent to the review of this research.

CONFLICT OF INTEREST

☒ There are no potential conflicts of interest involving any member of the research team since the last review.
☐ There is now a potential or actual conflict of interest involving a member of the research team since the last review. Complete and attach an updated UMCIRB Conflict of Interest disclosure form.

REQUIRED ATTACHMENTS FOR CONTINUING REVIEW

*****Note: To determine whether a research study should go to the full committee for review and approval or if the study can be approved by expedited review, see Instructions.**

Full Committee Review:

- **2 copies of protocol**
- **2 copies of publications/presentations**
- **20 copies of continuing review form**
- **20 copies of ALL consents/assents**
- **20 copies of protocol summary**

****These should be collated into individual packets with 2 of the packets containing the protocol and any publications/presentation information.**

Expedited Review:

- **1 copy of protocol**
- **1 copy of continuing review form**
- **1 copy of ALL consents/assents**
- **1 copy of protocol summary**
- **1 copy of publications/presentations**

*****Consent Documents**

1) **Continuing participant enrollment: Attach one clean copy (no notes, no highlighting, no stamps or no signatures) of the current consent document. This clean copy of the consent document will be stamped and returned to the investigator with the current approval period. *This stamped consent document should be the only form used to consent participants.* All previous versions of this consent document are considered invalid and may not be used to consent participants.**
2) **Closed to participant enrollment: Attach one copy of the current consent document. Note: A stamped consent document with the new approval period will not be sent the investigator.**

*****HIPAA Authorizations and Waivers of Authorization do not expire and, therefore, do not need to be resubmitted to the UMCIRB office.**

CLOSURE OF A RESEARCH STUDY

- **Each section should be completed regardless of whether this form is being submitted for continuing review or closure of a research study**
- **No consent documents are necessary.**
- **A copy of the protocol or protocol summary is not required.**

ACTION REQUESTED

☒ **Renew—continued participant enrollment**
☐ **Renew—no additional participant enrollment with follow-up for enrolled participants only, utilizing research related interventions conducted solely for gathering protocol related information**
☐ **Renew—no additional participant enrollment with long-term follow-up for enrolled participants only, utilizing follow-up interventions considered standard of practice that creates no research related burden for participants**
☐ **Renew—no additional participant enrollment; data analysis and interpretation only**
☐ **Terminate—research completed with no additional participant enrollment or collection of follow-up information. Provide rationale for study termination:**

Principal Investigator Signature **Print** **Date**

APPENDIX 2

University and Medical Center Institutional Review Board
East Carolina University • Brody School of Medicine
600 Moye Boulevard • Old Health Sciences Library, Room 1L-09 • Greenville, NC 27834
Office **252-744-2914** • Fax **252-744-2284** • **www.ecu.edu/irb**
Chair and Director of Biomedical IRB: L. Wiley Nifong, MD
Chair and Director of Behavioral and Social Science IRB: Susan L. McCammon, PhD

FILE COPY

TO: Kathryn Kolasa, PhD, RD, LDN, Pediatric Healthy Weight Research & Treatment Center, ECU e-mailed FAXED 10-7-09

FROM: UMCIRB

DATE: October 6, 2009

RE: Expedited Continuing Review of a Research Study MAILED 10-7-09

TITLE: "Medical Nutrition Therapy for Overweight Youth: An Outcomes Study"

UMCIRB #07-0618

The above referenced research study was initially reviewed and approved by expedited review on 10/5/07. This research study has undergone a subsequent continuing review using expedited review on 10/6/09. This research study is eligible for expedited review because it is a collection of data through noninvasive procedures (not involving general anesthesia or sedation) routinely employed in clinical practice, excluding procedures involving x-rays or microwaves. Where medical devices are employed, they must be cleared/approved for marketing. (Studies intended to evaluate the safety and effectiveness of the medical device are not generally eligible for expedited review, including studies of cleared medical devices for new indications.) Examples: (a) physical sensors that are applied either to the surface of the body or at a distance and do not involve input of significant amounts of energy into the subject or an invasion of the subject's privacy; (b) weighing or testing sensory acuity; (c) magnetic resonance imaging; (d) electrocardiography, electroencephalography, thermography, detection of naturally occurring radioactivity, electroretinography, ultrasound, diagnostic infrared imaging, doppler blood flow, and echocardiography; (e) moderate exercise, muscular strength testing, body composition assessment, and flexibility testing where appropriate given the age, weight, and health of the individual. It is also eligible for continuing review suing expedited review because it is research involving materials (data, documents, records, or specimens) that have been collected, or will be collected solely for nonresearch purposes (such as medical treatment or diagnosis). (NOTE: Some research in this category may be exempt from the HHS regulations for the protection of human subjects. 45 CFR 46.101(b)(4). This listing refers only to research that is not exempt.). The Chairperson (or designee) deemed this **Pitt Memorial Hospital Foundation** sponsored study **no more than minimal risk** requiring a continuing review in **12 months**. Changes to this approved research may not be initiated without UMCIRB review except when necessary to eliminate an apparent immediate hazard to the participant. All unanticipated problems involving risks to participants and others must be promptly reported to the UMCIRB. The investigator must submit a continuing review/closure application to the UMCIRB prior to the date of study expiration. The investigator must adhere to all reporting requirements for this study.

The above referenced research study has been given approval for the period of **10/6/09** to **10/5/10**. The approval includes the following items:

- Continuing Review Form (dated 9/10/09)
- Informed consent: parent (version 9/17/07)
- Minor Assent

The Chairperson (or designee) does not have a conflict of interest on this study.

The UMCIRB applies 45 CFR 46, Subparts A-D, to all research reviewed by the UMCIRB regardless of the funding source. 21 CFR 50 and 21 CFR 56 are applied to all research studies under the Food and Drug Administration regulation. The UMCIRB follows applicable International Conference on Harmonisation Good Clinical Practice guidelines.

Chapter 3 Index

A. General:
--Approach to the Adolescent Patient
--Managing Problem Health Behaviors in Adolescents
--Health Screening and Prevention Guidelines for Teens

Tanner Staging

I. Girls

Tanner Stage	Stage of develop	Pubic Hair	Breasts
Stage 1	Early adolescence (10-13 years)	Preadolescent	Preadolescent
Stage 2		Sparse, straight	small mound
Stage 3	Middle adolescence (12-14 years)	Dark, curl	bigger; no contour separation
Stage 4		Coarse, curly, abundant	Secondary mound of areola
Stage 5	Late Adolescence (14-17 years)	Triangle; medial thigh	nipple projects; areola part of breast

I. Boys

Tanner Stage	Stage of develop.	Pubic Hair	Penis	Testes
Stage 1	Early adolescence (10.5-14 years)	None	Preadolescent	preadolescent
Stage 2		Scanty	Slight increase	larger
Stage 3	Middle adolescence (12.5-15 years)	Darker, curls	Longer	larger
Stage 4		adult, coarse, curly	Larger	scrotum dark
Stage 5	Late adolescence (14-16 years)	adult - thighs	Adult	adult

Middle Adolescence (Stages 3 and 4): acceleration of weight and growth as well as above secondary sex characteristics. Pubic hair first, then axillary, then facial hair.

- Female: menarche (average age 12 years) - can occur in Stages 1 and 2; usually 3 and 4 factors affecting: nutrition, genetic - age of

mother's menarche.

- Male: gynecomastia also appears during middle adolescence: up to 70% of normal males.

Should Tanner 1 boys be allowed to play football with Tanner 5s? Controversial. Dr Landry of Madison Wisconsin says that there is no problem. However some literature states that adolescents that have gone through puberty recently are at higher risk of injury. (Clinical Journal of Sport Medicine 1995;5:167-70) Also study of strength, flexibility and maturity correlate better with Tanner staging than with chronological age. (AJDC 1989;143:560-3)

All agree that we worry about these mismatches in sports.

http://www.mcg.edu/pediatrics/CCNotebook/chapter3/tanner.htm

Next Page

APPENDIX 3