

RELATIONSHIP OF DIETARY FACTORS AND PHYSICAL ACTIVITY TO BODY
FAT IN 3-5 YEAR-OLD CHILDREN

A DISSERTATION

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BY

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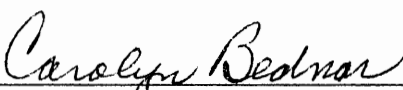
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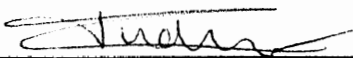
I am submitting herewith a dissertation written by Tina A. Crook entitled "Relationship of Dietary Factors and Physical Activity to Body Fat in 3-5 Year-Old Children." I have examined this dissertation for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Doctor of Philosophy with a major in Nutrition.

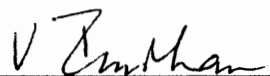


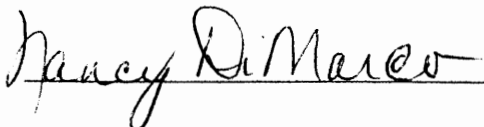
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We have read this dissertation and recommend its acceptance:











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DEDICATION

This project is dedicated to Norm, Mom, Dad, and Tessa. You have shown me what unconditional love, patience, and support are. These lessons are far more valuable than anything I will ever learn from a textbook. Thanks, guys. I love you all!

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ABSTRACT

TINA A. CROOK

RELATIONSHIP OF DIETARY FACTORS AND PHYSICAL ACTIVITY TO BODY FAT IN 3-5 YEAR-OLD CHILDREN

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The aims of the study were to 1) examine associations between environmental factors and percent body fat (PBF), and 2) determine if air-displacement plethysmography (ADP) and dual-energy x-ray absorptiometry (DXA) were accurate methods for assessing body fat in children 3 to 5 years old as compared to deuterium dilution (^2H). Data including PBF, diet during infancy, 3-day food records, and the Netherlands Physical Activity Questionnaire (NPAQ) were collected from 78 children. Pearson's correlation analyses showed a significant, positive relationship between PBF and Healthy Eating Index (HEI) scores, $r(36) = .332, p < .05$, and a significant, negative relationship between PBF and NPAQ scores, $r(36) = -.441, p < .01$, in male participants. Repeated measures multi-variate analysis of variance showed main effects for gender and body fat methods were significant, $F(1, 74) = 17.432, p < .0001$, partial eta squared = .191 and $F(2, 148) = 42.892, p < .0001$, partial eta squared = .367, respectively. Main effect for feeding group was not significant, $F(1, 74) = .553, p = .460$, partial eta squared = .007. Mean PBF measured by DXA ($M = 27.531, SD = 5.674$) was significantly different from mean PBF measured by ^2H ($M = 22.712, SD = 4.539$) and ADP ($M = 21.419, SD = 7.829$). Correlation between ^2H and ADP was moderate ($r = .461$,

$p < .01$). The results show obesity prevention should emphasize increased PA. The data failed to show breastfeeding influenced adiposity in young children. ADP may be an acceptable method when determining average PBF values in groups of children. However, the moderate correlation between ADP and ^2H shows that ADP needs additional adjustments before it can be used effectively in individual assessments. Evidence was provided that DXA overestimates PBF in children by approximately 5-6%. This overestimation was fairly consistent. Therefore, DXA may be useful in a clinical setting to monitor PBF of children as long as the absolute value obtained is not crucial.

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CHAPTER I

INTRODUCTION

Purpose

The purpose of this study was two-fold. The first purpose was to examine associations between environmental factors related to diet and physical activity and percent body fat in children 3 to 5 years old. The second purpose was to determine if air-displacement plethysmography (ADP) and dual-energy x-ray absorptiometry (DXA) were accurate methods for assessing body fat in children 3 to 5 years old as compared to a criterion method, deuterium dilution.

Hypotheses

The null hypotheses were as follows:

Ho₁: There will be no significant relationship between the percentage of body fat in 3 to 5 year old children and the following variables: mean daily intake of total fat in grams, polyunsaturated fatty acid in grams, fruit juice in fluid ounces, and calcium in milligrams; total daily energy in Kilocalories; Healthy Eating Index (HEI) scores; and Netherlands Physical Activity Questionnaire (NPAQ) scores.

Ho₂: There will be no significant difference in the percentage of body fat of 3 to 5 year old children fed breast milk vs. those fed infant formula during the first 6 months of life.

Ho₃: There will be no significant difference in the percentage of body fat of 3 to 5 year old children as measured using DXA vs. deuterium dilution, the criterion method.

Ho₄: There will be no significant difference in the percentage of body fat of 3 to 5 year old children as measured using ADP vs. deuterium dilution, the criterion method.

Ho₅: There will be no significant difference in the percentage of body fat of children 3 to 5 years old as measured using DXA, ADP or deuterium dilution.

Rationale

The most recent 2003-2004 data from the National Health and Nutrition Examination Survey (NHANES) shows that 17.1% of U.S. children ages 2 to 19 years are overweight, and that number increases to 33.6% when those children at risk for becoming overweight are included (Ogden et al., 2006). Numerous chronic health problems are associated with being overweight in childhood. For example, an increase in the rate of type 2 diabetes mellitus in adolescents has been observed over the past two decades as the prevalence of overweight has risen (Boney, Verma, Tucker, & Vohr, 2005). In addition, overweight children are at increased risk for developing other life-long health problems such as cardiovascular disease, hypertension, pulmonary disease, liver disease and orthopedic problems (Angulo, 2007; Boney et al., 2005; Patton et al., 2006; Zametkin, Zoon, Klein, & Munson, 2004).

Environmental factors, including excess dietary intake and physical inactivity are the most important determinants of obesity development (Parikh & Yanovski, 2003). Links between several dietary factors and percent body fat have been reported . Skinner, Bounds, Carruth, and Ziegler (2003) observed intake of total dietary fat and saturated fat

are positively associated with percent body fat while calcium intake and polyunsaturated fat intake were found to be negatively related to percent body fat in a group of 8-year-old children ($N = 52$). Manios et al. (2008) reported a positive association between total daily energy intake and weight in a group of 1 to 5 year-old children ($N = 2374$). Several researchers have linked poor diet quality to weight status in children (Hassapidou, Fotiadou, Maglara, & Papadopoulou, 2006; Langevin et al., 2007). Excessive fruit juice consumption is another factor suggested to increase a child's risk for weight gain (AAP, 2001). Additionally, the effects of early feeding on later obesity have begun to emerge, and there is some evidence to suggest that breastfeeding may be protective (Martorell, Stein, & Schroeder, 2001; Owen et al., 2005).

The relationship between physical activity and body fat has also been explored. Skinner et al. (2003) reported a positive association between sedentary activity and percent body fat in 8-year-old children. Some of the sedentary activities studied include watching TV, playing video games, and using the computer. Lumeng et al. (2006) concluded that TV exposure equal to or greater than two hours per day was an independent risk factor for a child being overweight in a cross-sectional study of 1,016 three-year-old children. Others suggest that time spent playing video games is more detrimental than watching television (Vandewater, Shim, & Caplovitz, 2003). Thus, it is important to assess all forms of "screen time" including television, video games, and computer usage since all are sedentary behaviors and have been associated with weight status in children.

Based on these data, it was expected that a positive relationship would be found between percent body fat and the following variables: total fat grams, fruit juice consumption, and total daily energy intake. A negative relationship was predicted between percent body fat and the following variables: polyunsaturated fatty acids grams, calcium intake, HEI scores, and NPAQ scores. It was also anticipated that children who were breastfed during infancy would have a lower percent body fat.

Adult obesity and associated chronic diseases originate early in life and fat deposition in infancy and childhood may be a factor (Barker, 2004). Consequently, there is a need for accurate, practical methods for monitoring body composition in the pediatric population. Several techniques exist for measuring body fat but they are not all appropriate for use with infants and children. The safety, invasiveness, and time required to acquire results are all factors to consider when selecting a body composition assessment method for children. Young children also have difficulty remaining still throughout a long test procedure, so an ideal method would allow for some movement during testing.

Some of the most popular methods for assessing body composition include skinfold (SF) measures, bioelectrical impedance analysis (BIA), underwater-weighing (UWW), DXA and ADP (Ellis, 2000). Studies indicate that SF and BIA measures are less accurate than UWW, DXA and ADP in measuring body fat, although these techniques may still be useful in field studies because of their portability (Bray et al., 2002; Roemmich, Clark, Weltman, & Rogol, 1997). Underwater-weighing has limited use in young children because of its requirement for submersion in water (Ellis, 2000;

Ellis 2001). DXA delivers a small dose of radiation and requires that children remain still throughout the test. Additionally, there are some reports that claim it is less accurate at assessing body composition in children than adults (Ellis, 2000; Fields & Goran, 2000; Roemmich et al., 1997; Schoeller et al., 2005; Sopher et al., 2004; Testolin, et al., 2000). Air displacement plethysmography has only been validated for children 6 years of age and older (Higgins et al., 2006). Air displacement plethysmography may be useful in younger children if the calculations for body volume are modified based on age and gender specific equations (Bosey-Westphal et al., 2005). A criterion or reference method must be selected as the standard for comparison to assess the accuracy of a body composition technique. Multi-compartment models are used as reference methods because they measure two or more of the components of the fat free mass (FFM) including bone mineral, protein and water (Reilly, 1998). It is important to measure the various components of FFM because overall composition of FFM varies based on age and gender (Baumgartner, Heymsfield, Lichtman, Wang, & Pierson, 1991; Ellis, 2000; Hewitt, Going, Williams, & Lohman, 1993; Reilly; Wang et al., 1999). Most authorities agree that a four-compartment (4-C) model that takes into account body water, protein, bone mineral and fat mass is ideal (Ellis, 2000; Fields & Goran, 2000; Reilly; Roemmich et al., 1997; Sopher et al., 2004). However, a 4-C model requires equipment that is not available in most facilities to measure the protein component of FFM and it requires moderate radiation exposure (Fields & Goran; Ellis, 2000; Wang et al., 2003). This makes it an impractical choice in the majority of cases, particularly for children. An acceptable alternative recognized as a criterion method by several researchers is total

body water (TBW) analysis (Reilly, 1998; Schoeller et al., 2005; Yao, Roberts, Ma, Pan, & McCrory, 2002). TBW applies dilution principles to calculate lean and fat mass (Sheng & Huggins, 1979). The dilution principles state that if a known amount of a tracer is given and time is allowed for it to become evenly distributed throughout the body's fluids, then the concentration of the tracer in a sample of body fluid can be used to determine TBW (Ellis, 2001; Sheng & Huggins). Deuterium oxide, a stable isotope of hydrogen, was discovered in 1932 and has been used extensively as a tracer in determining TBW in both human and animal studies (Sheng & Huggins; Yoa et al., 2002). Recent reports suggest that dilution techniques are accurate in predicting percent body fat within 1-2% (Bray et al., 2002; Ellis, 2000; Schoeller et al.; Wang et al., 1999).

It was expected that DXA would produce significantly different results than the criterion method in the present study based on reports that it is not as accurate in children compared to adults. It was believed that ADP would produce results similar to the criterion method since age-appropriate adjustments were made to the equations that had been shown to be accurate in older children.

Abbreviations

AAP: American Academy of Pediatrics

ACNC: Arkansas Children's Nutrition Center

ADP: Air displacement plethysmography

BIA: Bioelectrical impedance analysis

BMD: Bone mineral density

BMI: Body mass index

BSA: Body surface area

C1: Constant value 1

C2: Constant value 2

CDC: Center for Disease Control

Db: Body density

Dffm: Density of fat-free mass

DRI: Dietary reference intake

DXA: Dual energy x-ray absorptiometry

FFM: Fat free mass

FRC: Functional residual capacity

H: Height

HDL-C: High density lipoprotein cholesterol

HEI: Healthy eating index

HIPAA: Health Insurance Portability and Accountability Act

k: Constant value

LDL-C: Low density lipoprotein cholesterol

LMi: Life Measurements, Incorporated

MS: Metabolic syndrome

NAFLD: Non-alcoholic fatty liver disease

NASH: Non-alcoholic steatohepatitis

NCRP: National Council of Radiation and Protection

NDSR: Nutrition Data System for Research

NHANES: National Health and Nutrition Examination Survey

NPAQ: Netherland's Physical Activity Questionnaire

PUFA: Polyunsaturated fatty acid

QC: Quality control

SAA: Surface area artifact

SF: skin folds

SoFAAS: Solid fats, alcohol and added sugars

SSF: Subscapular skinfold

TBW: Total body water

TGV: Thoracic gas volume

TSF: Triceps skinfold

TV: Television or tidal volume

T2DM: Type 2 diabetes mellitus

UAMS: University of Arkansas for Medical Sciences

UWW: Under-water weighing

2H : Deuterium oxide

2-C model: two compartment model

3-C model: three compartment model

4-C model: four compartment model

Definition of Terms

Adiabatic conditions: refers to air that freely gains and loses heat during compression and expansion (Fields, Goran, & McCrory, 2002)

Adiposity: a general term that refers to the amount of adipose tissue in the body; Adipose tissue includes both essential and non-essential fat. Non-essential fat is associated with bone marrow, the central nervous system, internal organs, cell membranes, and in females, mammary glands and the pelvic region. Non-essential fat is additional fat stored in the body. It is increased amounts of the non-essential fat that is associated with an increased risk for chronic diseases such as cardiovascular disease and T2DM (Gropper, Smith, & Groff, 2005).

Adiposity rebound: the period of time in childhood when body fat accumulation increases at a rapid rate (Barker, 2004)

At risk for overweight in childhood: a BMI for age percentile between 85 and 95 (CDC, 2006)

Body impedance: a term used to describe the body's resistance to an electric current that is measured in the bioelectrical impedance analysis technique for measuring body fat (Ellis, 2000; Ellis, 2001)

Body mass index (BMI): describes one's weight in relationship to height taking into account body surface area and is calculated by dividing weight in kilograms by height in meters squared (Ellis, 2001)

Bone mineral density (BMD): "the ratio of bone mineral content to bone area" (Ellis, 2000, p. 662)

Closed-circuit spirometer: device used to measure lung function (Beydon et al., 2007)

Densitometry techniques: methods used to calculate body density based on the principle that body density is equal to body mass divided by body volume (Fields et al., 2002)

Developmental origins theory of disease: theory that factors early in life impact disease development later in life (Barker, 2004)

Diet quality: a quantitative way to describe the nutritional merit of one's diet as a whole instead of focusing on individual nutrients by looking at groups of key nutrients or using a standardized tool

Effective dose of radiation: measurement of the biological risk associated with radiation exposure (Thomas, Kalkwarf, Buckley, & Heubi, 2005)

Fat-free mass (FFM): body component made up of body water, protein and bone mineral (Ellis, 2000)

Fat mass: body component comprised of fat tissue and is typically determined by subtracting FFM from total mass (Ellis, 2000)

Frankfort plane: anatomical plane that extends from the outer corner of the eye to the opening in the ear

Fruit juice: a beverage containing 100% fruit juice (AAP, 2001)

Isothermal conditions: refers to a constant temperature (Fields et al., 2002)

Juice drink, beverage or cocktail: defined by the Food Drug Administration as a beverage containing less than 100% fruit juice (AAP, 2001)

Media time: time spent watching television, playing video games, using the computer and reading (Vandewater et al., 2004)

Menarche: the time that menstruation begins in girls

Metabolic syndrome (MS): “association of obesity, insulin resistance, glucose intolerance, hypertension and a characteristic hyperlipidemia” (Boney et al., 2005, p. 291)

Non-alcoholic fatty liver disease (NAFLD): “the accumulation of fat, primarily triglycerides, in hepatocytes that exceeds 5% of the liver weight” (Angulo, 2007, p. S57)

Non-alcoholic steatohepatitis (NASH): a more severe stage of NAFLD that results in inflammation and ballooning of the liver cells (Angulo, 2007)

Overweight in childhood: a BMI for age percentile at or above 95 (CDC, 2006)

Plethysmograph: the enclosed chamber that allows for measuring body volume in the air-displacement plethysmography technique for measuring body fat (Fields et al., 2002)

Plethysmography: “the measurement of size, usually volume” (Fields et al., 2002, p. 454)

Reactance: “measure of the opposition that a circuit or part of a circuit presents to electric current” and present with resistance when currents alternate (Encyclopedia Britannica, 2009)

Residual lung volume: amount of air that remains in the lungs after maximum exhalation (Ellis, 2000)

Resistance: a term used to describe the impedance or opposition of an electric current through the body in the bioelectrical impedance analysis technique for measuring body composition (Ellis, 2000; Ellis, 2001)

Surface area artifact: the effect that the isothermal air near the surface of the skin has on the measurement of body volume using plethysmography (Fields et al., 2002)

Thoracic gas volume (TGV): “the average amount of air in the lungs during normal tidal breathing” (Fields et al., 2002, p. 454)

Tidal volume (TV): “the volume of air inhaled or exhaled with each breath during breathing” (Cherniack, 1977, p. 10)

Weighed food records: a recording of all foods and beverages consumed that uses scales to quantify portions (Hassapidou et al., 2006)

CHAPTER II

LITERATURE REVIEW

The rate of obesity has steadily risen in the United States (U.S.) since the 1960's. Data from the 2003-2004 National Health and Examination Survey (NHANES) showed that 66.3% of American adults were overweight or obese, and 17.1% of U.S. children ages 2 to 19 years were overweight (CDC, 2009; Ogden et al., 2006). That number increases to 33.6% for children when those at risk for becoming overweight are included (Ogden et al.). There has been a significant increase in the rate of overweight children since the 1999-2000 NHANES survey that showed 13.9% of children 2 to 19 years were overweight with 28.2% in the at risk and overweight categories combined (Ogden et al.). This emphasizes the alarming rate at which this epidemic continues to grow in the pediatric population.

The term obesity has not been used in the U.S. to describe children and adolescents because obesity is a measure of body fatness. Body mass index, the tool most commonly used to define weight categories in children, is not a measure of body fatness. Therefore, overweight was determined to be a more appropriate term than obesity for use in children (Himes & Dietz, 1994). Overweight in childhood is defined as a body mass index (BMI) for age percentile at or above 95 and at risk for becoming overweight in childhood as a BMI at the 85 to 95 percentile (CDC, 2006).

Physical Problems Associated with Overweight Children

Numerous chronic health problems are associated with being overweight in childhood. These include type 2 diabetes mellitus (T2DM), cardiovascular disease, hypertension, pulmonary disease, orthopedic problems, and neurological impairments (American Academy of Pediatrics, 2003; Boney, Verma, Tucker, & Vohr, 2005; Zametkin, Zoon, Klein, & Munson, 2004). Zametkin et al. attributed neurological impairments to obstructive sleep apnea, a common condition in many obese children, that can lead to learning and memory deficits. Overweight children are also at increased risk for gallstone formation (Zametkin et al.).

Another chronic condition in children that has become more prevalent as childhood obesity rates have risen is nonalcoholic fatty liver disease (NAFLD). NAFLD is defined by Angulo (2007, p. S57) as “the accumulation of fat, primarily triglycerides, in hepatocytes that exceeds 5% of the liver weight”. Nonalcoholic fatty liver disease, as the name implies, is not caused by alcohol ingestion, and the diagnosis should only be made after ruling out other causes of liver damage such as excessive alcohol intake or use of hepatotoxic drugs (Angulo). A more severe stage of NAFLD that results in inflammation and ballooning of the liver cells is referred to as nonalcoholic steatohepatitis or NASH (Angulo). The first observations of fibroses in children were made in the early 1980’s (Patton et al., 2006).

NHANES III data suggests that 6-10% of overweight adolescents may have NAFLD, while the Child and Adolescent Trial for Cardiovascular Health study reported that as many as 23% of overweight adolescents may have the condition (Patton et al.). Central or

visceral obesity along with insulin resistance, T2DM, high blood pressure, and abnormal lipid levels are risk factors for NAFLD, but it may occur even in the absence of those conditions (Angulo).

Several of the aforementioned health problems are characteristic of the metabolic syndrome (MS), defined as the “association of obesity, insulin resistance, glucose intolerance, hypertension and a characteristic hyperlipidemia.” (Boney et al., 2005, p. 291). Pan and Pratt (2008) analyzed data from 4,450 children ages 12 to 19 who participated in the 1999-2002 NHANES Survey and found that 3.5% of the sample had MS. Further, they reported it was more common in males at a rate of 5.1% than in females at a rate of 1.7%. It was also dramatically higher in the children classified as overweight (BMI \geq 95 percentile) than in those with a BMI less than the 85 percentile (14.5% v. 0.9%, respectively).

Psychosocial Problems Associated with Overweight Children

The American Academy of Pediatrics (AAP, 2003, p. 425) states, “The psychologic stress of social stigmatization imposed on obese children may be just as damaging as the medical morbidities.” Some children become so afraid of becoming obese that they restrict food intake to the point that growth failure and delayed development result (AAP, 2003). Some researchers have even become concerned that intervention programs for obese children could illicit unhealthy eating and physical activity behaviors because the children become so focused on the potential health problems associated with the condition (Huang, Norman, Zabinski, Calfas, & Patrick,

2007). A negative body image and lowered self-esteem may result from heightening an obese child's awareness of his condition (Huang et al.).

Huang et al. (2007) conducted a one-year randomized controlled trial behavior intervention study in a group of 657 adolescents. Data was collected at baseline, six months, and 12 months to assess body image, self-esteem, and weight status. Baseline data showed that obese subjects had significantly lower body image and self-esteem scores than the normal weight control subjects ($p < 0.001$ for both variables). Female subjects who lost or maintained their weight at 6 and/or 12 months had significant improvements in their body image compared to females who gained weight ($p = 0.02$). This effect was not seen in the male subjects. Neither male nor female subjects showed significant decreases in self-esteem or body image in the intervention group regardless of whether they gained, maintained, or lost weight. This is an intriguing finding that may suggest negative body image and lowered self-esteem reported by other researchers was not observed in this study because the intervention concentrated on healthy behaviors and health outcomes instead of weight loss. On the other hand, lack of observed negative body image and lowered self-esteem in the Huang et al. study may be due to differences in study participants or the tools used to assess these factors.

Causes of Overweight in Childhood

Clearly, children and teens are becoming overweight at epidemic proportions. The literature suggests several causes including poor eating habits, physical inactivity, the environment, cultural beliefs, genetics, and some illnesses (AAP, 2003; Ebbeling, 2002; Gillman et al., 2001; Patterson, Satia, Kristal, Neuhouser, & Drewnowski, 2001;

Rolls, Engell, & Birch, 2000; Surgeon General, 2001). Most experts believe it is a combination of several of these factors that causes one to become overweight (Parikh & Yanovski, 2003; Pereira et al., 2002). The Surgeon General's Call to Action to Prevent and Decrease Overweight and Obesity (2001) stated that the greatest areas for prevention and treatment are changes in behavior and environment.

Dietary Factors

Tools for assessing dietary intake. Several tools are available for assessing dietary intake. Food frequency questionnaires serve mainly as a tool for ranking nutrient intakes while food recalls and food records allow for quantification of nutrients (Serdula, Alexander, Scanlon, & Bowman, 2001). Each method has advantages and disadvantages that researchers must consider.

Lanigan et al. (2004) cited over-reporting and under-reporting as common occurrences with food recalls and food records. Individuals who do food recalls rely heavily on memory, and they are less likely to forget items if intake is recorded as it occurs. Lanigan et al. suggested that because the diets of infants and young children are less varied than adults, fewer days of food records are required for accurate dietary assessment. They estimated the number of days of food records required to accurately assess various nutrients and found that for micronutrients, such as calcium, two days were necessary while accurate assessment of fat intake required four days and energy intake five days.

When determining the number of days to collect food records, one should consider the burden placed on the participant or caregiver. The responsibility for data

collection using food records in young children falls to the caregiver. Compliance may be poor if asked to record too many days. These factors must be considered when planning studies that assess nutrient intake.

Total fat intake. Fat intake has been hypothesized to be linked to weight status and body fat in children. Skinner, Bounds, Carruth and Ziegler (2003) created prediction models using multiple regression analyses to study this association. Multiple regression analysis provides an R^2 statistic that indicates the amount of variance in a model that is attributed to the dependent variable (Pallant, 2007). They found total fat intake along with female gender, hours spent in sedentary activities, and father's BMI to be positive predictors of percent body fat, as measured by dual energy x-ray absorptiometry (DXA), in three prediction models ($R^2 = 0.26-0.336$, $p = 0.002-0.006$). However, other researchers' data failed to associate fat intake to weight status (Aeberli, Kaspar, & Zimmermann, 2007; Hassapidou, Fotiadou, Maglara, & Papadopoulou, 2006; Magarey, Daniels, Boulton, & Cockington, 2001). Differences in findings may be due to the different study designs and outcome measures used among researchers. Skinner et al. used body fat measured by DXA, Aeberli et al. and Hassapidou et al. used body weight, and Magarey et al. used BMI and skin fold measures as outcome measures. Body fat is a better indicator of overall health status than body weight and, therefore, more studies using a reliable measure of body fatness should be designed to explore this possible link.

Polyunsaturated fatty acid intake. Similarly, equivocal findings have been reported regarding the relationship between polyunsaturated fatty acid (PUFA) intake and weight or body fat status in children. Skinner et al. (2003) found PUFA to be negatively

associated with body fat, along with calcium, in four different models ($R^2 = 0.26-0.336$, $p = 0.002-0.006$). On the other hand, Aerberli et al. (2007) were unable to establish a relationship between PUFA and BMI levels. Again, different study designs and outcome measures may explain the disparity in findings.

Fruit juice consumption. Americans consume an average of 2 billion gallons of fruit juice per year with children representing the largest group of juice drinkers (AAP, 2001). Fruit juice is defined by the Food Drug Administration as a beverage containing 100% fruit juice. Any beverage with less than 100% juice must be labeled as a juice drink, beverage, or cocktail (AAP). Juice contains some essential nutrient such as vitamin C and folate, but excessive juice consumption may have negative effects such as gastrointestinal distress, over-nutrition, under-nutrition, short stature, and dental caries (AAP). Thus, the AAP recommends no fruit juice until infants are 4-6 months old and then limiting fruit juice to 4-6 fluid ounces per day for children 1-6 years, and 8-12 fluid ounces per day for children 7-18 years of age (AAP).

O'Connor, Yang, and Nicklas (2006) investigated the beverage intake of preschool children who participated in the 1999-2002 National Health and Nutrition Examination Survey (NHANES). They conducted a secondary analysis of the dietary data collected from 24-hour recalls on 1,160 children ages 2-5 years and compared that to the children's BMI to determine if beverage consumption was linked to weight status. They found an average fruit juice consumption of 4.7 fluid ounces per day, while the average fruit drink and soda consumption combined was 4.98 fluid ounces per day. The children in the sample were within the AAP's recommendation of 4-6 fluid ounces per

day of 100% fruit juice; however, they were consuming other sweetened beverages not recommended by the AAP. No significant associations were found between the beverages consumed and the children's weights. There was, however, a link between total energy intake and consumption of all beverages including milk, fruit juice, juice drinks, and soda. Total energy increased as consumption of all beverages increased, but there was not a statistically significant difference in BMI values based on the amount of beverages consumed.

Calcium intake. In recent years, studies examining the relationship between calcium intake and body fat in children have begun to emerge. Interest in this potential link stemmed from animal studies and epidemiological data that suggested those who consumed more dairy products were less likely to be overweight (Parikh & Yanovski, 2003; Zemel, Shi, Greer, Dirienza, & Zemel, 2000).

A negative relationship between dairy consumption and percent body fat in children was documented by Carruth and Skinner (2001) and Barba, Troiano, Russo, Venezia, and Siani (2005). Carruth and Skinner conducted regression analyses and determined that longitudinal calcium intake as well as the number of servings of dairy products consumed were negatively associated with body fat as measured by DXA ($p = 0.0003$ and $p = 0.001$, respectively) in a group of 53 white children ages 2 months to 8 years. Likewise, Barba et al. studied 884 children ranging in age from 3 to 11 years and showed in multiple regression models that milk consumption was inversely related to BMI z scores ($p = 0.003$) after controlling for several variables including sex, age, physical activity, birth weight, and parental overweight and education.

Other researchers have had contradictory findings. Phillips, Bandini, Cyr, Colclough-Douglas, and Must (2003) conducted a longitudinal study of 178 children similar to the study by Carruth and Skinner (2001) but studied older children ages 8 to 12 years and used bioelectrical impedance analysis (BIA) instead of DXA to measure body fat. They did not find an association between daily servings of dairy foods or percentage of daily calories from dairy foods and body fat. Similar to Carruth and Skinner, they controlled for several variables including physical activity levels, parental overweight, sex, and ethnicity. Perhaps the different outcome measures used and ages of the children in the two studies explain the discrepancy in findings.

O'Connor et al. (2006) reported no relationship between milk consumption and BMI levels in a sample of 1,160 children ages 2 to 5 years. Like Barba et al. (2005), BMI was used as an outcome measure, but the findings from the two studies are conflicting. O'Connor et al. grouped milk consumption into categories of none, >0 to 8 fluid ounces per day, >8 to 16 fluid ounces per day, >16 to 24 fluid ounces per day, and >24 fluid ounces per day. These groupings differed from the groupings used in the study conducted by Barba et al. and may partially explain the inconsistency in findings between studies. Additionally, Barba et al. studied older children which may have impacted the results.

Few randomized experimental studies investigating the link between calcium intake and weight or body fat status in children exist. One experimental study conducted by Lappe, Rafferty, Davies, and Lypaczewski (2004) was conducted in a group of 9-year-old females ($N = 59$) to determine if increased calcium intake affected weight and body

fat. Participants were randomly assigned to either a high calcium group receiving ≥ 1500 mg of calcium per day or a control group instructed to consume a regular diet. Subjects were followed quarterly for two years. Both groups provided 3-day food diaries and had weight and height measures taken at each visit. DXA was used to assess lean and fat mass of the participants at baseline and the end of the study. No differences in body weight, BMI, fat mass, or lean mass was found between the two groups at the end of the study. It is important to consider that both groups had fairly high calcium intakes (control = 961 ± 268 mg/day, high calcium = 1656 ± 191 mg/day). In addition, unlike the other studies that assessed dairy or milk consumption, Lappe et al. focused on calcium intake. Perhaps other components in dairy products are responsible for the link to percent body fat identified by Carruth and Skinner (2001) and Barba et al.

These inconsistent findings regarding dairy consumption and weight or fat status in children warrant further investigation. Future studies must focus on specific components in dairy foods to more clearly identify the potential protective factors involved. Additionally, narrower age ranges of children should be used in analyses so that it can be determined if age impacts results.

Total daily energy intake. Total daily energy intake is another factor that must be considered when investigating the causes of overweight in childhood. Aeberli et al. (2007) and Manios et al. (2008) both showed that increased energy intake related to increased BMI levels in children. Aeberli et al. found a correlation between energy intake and BMI in female subjects ($p = 0.002$) but not in the male subjects. Manios et al. found that both male and female children at risk for becoming overweight and those who

were overweight consumed significantly more kilocalories per day than normal weight children (1,434 kcals/day, SD = 242, v. 1,445 kcals/day, SD = 270, v. 1,386 kcals/day, SD = 233, respectively, $p < 0.001$).

Langevin et al. (2007) did not find a relationship between kilocalorie intakes and BMI levels in a cross-sectional study of 193 low income children. Nor, were Hassapidou et al. (2006) able to relate energy intake to weight status. These data must be interpreted cautiously. The study by Langevin et al. had more children who were at risk for being overweight (22%) or overweight (36%) than NHANES data revealed for the rest of the population (Ogden et al., 2006). This, along with the fact that the children were all from low income families, should be considered when interpreting the results. Another consideration is that study participants may have been inclined to underestimate food intake as is the case with all studies using self-reported data.

Diet quality. The quality of the diet is another area of interest when studying potential causes of increased rates of overweight in children. Diet quality is a way of determining the nutritional merit of the diet as a whole instead of focusing on individual nutrients. Some researchers make inferences about diet quality by focusing on groups of key nutrients and others use standardized tools developed for measuring diet quality. Such tools take into account factors like nutrient density of the foods consumed as well as the overall balance and variety of food choices in the diet.

The Healthy Eating Index (HEI) (2005) is an example of a diet quality tool developed by the United States Department of Agriculture's (USDA) Center for Nutrition Policy and Promotion. It was designed for Americans over the age of two to measure

how closely diet matches the 2005 recommendations of the Dietary Guidelines for Americans and the Food Guide Pyramid. A high score indicates good diet quality while a low score indicates poor diet quality. Low scores reflect both overconsumption and under-consumption of nutrients recommended by the USDA. Guenther et al. (2007) measured the content validity, construct validity, and reliability of the HEI.

Content validity of the HEI was assessed in two ways. First, components that make up the HEI were checked against the 2005 Dietary Recommendations for Americans to determine if they were represented in the HEI. All of the recommendations were reflected in the HEI with the exception of those related to body weight, physical activity, and food safety. The authors stated that these were intentionally eliminated when the HEI was developed. The second way that content validity was assessed was by measuring face validity. Using food recalls from NHANES 2001-2002, researchers divided the recalls into 10 groups based on HEI scores and selected four diets from each group. Professional judgment was used to determine if the 40 diets selected reflected the Dietary Guidelines for Americans. Those with the highest HEI scores were identified as most closely matching the Dietary Guidelines, while diets with the lowest scores were identified as having the least in common with the Dietary Guidelines.

Construct validity was measured five different ways. First, researchers compared four different sets of menus that were developed by nutrition experts and believed to have high diet quality to HEI scores. The menus included 1) the seven-day 2000 Kilocalorie menus on the My Pyramid website, 2) the seven-day sample menus developed for the DASH diet, 3) the two one-week menus developed for Harvard Medical School's

Healthy Eating Pyramid, and 4) the two one-day menus by the American Heart Association for the No-Fad Diet. All of the menus received high HEI scores ranging from 90.9 to 100.

Second, construct validity was examined by testing to see if the HEI could differentiate between diets of groups with known differences. NHANES 2001-2002 recalls from diets of smokers and non-smokers were compared to individual HEI component scores between the groups. Researchers found that nine of the 12 component scores were significantly lower for the smokers ($p < 0.01$).

Third, construct validity was measured to determine that diet quality was unrelated to diet quantity. Researchers examined correlations between total HEI and component scores and total energy intake. Correlation analyses provide r statistics that illustrate the strength of the relationship between variables (Pallant, 2007). They found low relationships for all components. The highest correlation was between the solid fats, alcohol, and added sugars category (SoFAAS) and energy intake ($r = -0.22$) while all other components had absolute values of 0.11 or less.

Fourth, construct validity was investigated by reviewing the distribution of HEI scores as it is important that tools provide a range of scores to reflect the population of interest. Most components provided an acceptable range of scores but the whole fruit; dark green vegetables, orange vegetables, and legumes; and whole grain components were not as varied because many individuals reported no intake of these foods.

Fifth, construct validity was evaluated by using principal components analysis to determine how many factors influence the HEI. A screen plot showed that

between five and eight factors appeared to influence the total score. This indicates that the HEI provides a multifaceted approach for measuring diet quality.

Reliability of the HEI was measured for internal consistency using Cronbach's coefficient alpha. The analysis found a moderate correlation of 0.43, and the component scores with the strongest relationships to the overall score were SoFAAS (0.57), total fruit (0.43), and whole fruit (0.45). The other scores ranged from absolute values of 0.01 to 0.26. The researchers expected to find low correlations since many factors impact diet quality. This shows the HEI is not influenced to a large degree by individual components and meets its goal of reflecting various characteristics of diet quality.

Several researchers have explored diet quality as it relates to weight status in children, and most agree that a positive relationship exists between variables (Hassapidou et al., 2006; Langevin et al., 2007; Receveur, Morou, Gray-Donald, & Macaulay, 2008). However, the different approaches used to determine diet quality among studies makes it difficult to draw concise conclusions from the data. Langevin et al. used the Block Food Frequency Questionnaire for Children 2004 while Hassapidou et al. assessed intake of individual food items. Receveur et al. considered several factors including total energy intake, percent kilocalories from fat, energy density and diet diversity. A standardized tool for assessing diet quality, such as the HEI, would be beneficial in future studies so that inferences can be made about children's diets compared to an acceptable standard such as the Dietary Guidelines for Americans.

Infant feeding. The AAP Section on Breastfeeding recommends breast milk as the sole source of nutrition for infants until 6 months of age (AAP, 2005). Many benefits

have been documented for breastfeeding during infancy including decreased rates and severity of infectious diseases, lower rates of infant mortality, improved immune function, and better cognitive development (AAP). Breastfeeding is also more cost-effective than formula feeding and provides a unique opportunity for mother-child bonding (AAP). It is considered the gold standard in terms of nutrition quality. The AAP (p. 496) stated, “Exclusive breastfeeding is the reference or normative model against which all alternative feeding methods must be measured with regard to growth, health, development, and all other short- and long-term outcomes.” Protection against obesity later in life is another potential benefit of breastfeeding during infancy which has emerged in the literature (AAP; Toscheke et al., 2007). The AAP recognized this possible association and stated that more research was needed on the topic.

Owen et al. (2005) conducted a systematic review of literature and performed a meta-analysis on 36 studies to examine the mean differences in BMI later in life between those participants who were breastfed versus formula-fed during infancy. A lower mean BMI was found in the groups who had been breast fed in 23 of the 36 studies reviewed ($p < 0.001$). Eighteen of the studies also showed that a longer duration of breastfeeding offered more protection. The researchers performed a meta-regression and found a decrease of 0.04 in mean BMI for each added month of exclusive breastfeeding. However, the effects diminished when confounding variables were considered. The effect was cut in half when maternal BMI in early life was considered, and it was completely eliminated when maternal BMI, maternal SES, and maternal smoking were

taken into account in 11 of the studies. Thus, the lower mean BMI levels observed in the breastfeeding group cannot be attributed to infant feeding.

Individual studies reviewed reported similar findings. Burdette, Whitaker, Hall, and Daniels (2006) conducted regression analyses on a group of 5-year-old children ($N = 313$) and found no link between breastfeeding or duration of breastfeeding and the child's body fat level as measured by DXA at age 5. Likewise, a birth cohort study conducted in the United Kingdom in a group of 881 children failed to show a relationship between breastfeeding during infancy and lower BMI later in life (Ong et al., 2006). Michels et al. (2007) also concluded that breastfeeding did not appear to be a factor in obesity prevention in a group of 35,830 participants in the Nurses' Health Study II. Some limitations of these studies are that infant feeding history was self-reported and in some cases data were collected several years later. Thus, reliance on memory for the quantity and duration of breastfeeding may have affected results. In addition, weight data were also self-reported in the study by Michels et al.

Studies suggest that duration of breastfeeding should also be considered when examining its potential protective effects against weight and fat gain later in life. Toscheke et al. (2007) concluded that breastfeeding for 6 months or more may offer protection from obesity later in life while Apfelbacher et al. found that breastfeeding for more than 3 months was negatively related to being overweight or obese. Therefore, breastfeeding duration is an important variable to assess in future studies.

No definitive conclusions have been drawn about the relationship between breastfeeding and weight later in life although several researchers have explored the

topic. Many of these studies have been retrospective and have used different outcome measures making it difficult to compare results. In addition, it becomes challenging to design studies that show a direct relationship between breastfeeding and weight or adiposity because of the many environmental factors that can influence weight gain over time.

Other dietary factors. Other dietary factors that are potential contributors to weight imbalances in children include soft drink consumption, portion sizes, increased fast food intake, skipping breakfast, and a decrease in the number of meals families consume together. Several health problems that may be linked to excessive soft drink consumption in children include weight gain, bone disease, and dental caries (AAP, 2004). Other researchers have linked soft drink consumption to sedentary behaviors. Coon, Goldberg, Rogers, and Tucker (2001) found that soft drink consumption in a group of fourth to sixth grade children was positively related to watching television at two or more meals.

Rajeshwari, Yang, Nicklas, and Berenson (2005) confirmed an inverse association between milk and the consumption of sweetened beverages including soda, fruit juice and sweetened tea and coffee. They studied the dietary habits of 1,548 children ten years of age using 24-hour diet recalls over a 21 year period in Bogalusa, Louisiana. They found that children consuming the most sweetened beverages consumed significantly less milk ($p < 0.0001$). However, no association between sugary beverage consumption and BMI levels was observed.

Other researchers have looked at the relationship between weight and soda intake. O'Connor et al. (2006) found no association between BMI levels and beverage consumption in a group of 1,160 preschool children ages 2 to 5 years despite the fact that those who consumed more milk, juice, and soda had higher energy intakes. The authors concluded that the effects of increased energy intake on BMI may not be apparent until after children go through adiposity rebound that usually occurs later at 5 to 6 years of age.

The increasing portion sizes at restaurants, fast food chains, and convenience stores may play a part in the observed increased rates of obesity as well. Rolls et al. (2000) examined the effects of portion size on children at 3 and 5 years of age. They found that 5 year olds ate greater amounts when presented with larger portions, but 3 year olds were not affected by portion size. These results are consistent with other reports that suggest as children develop, their food intake is influenced by a variety of social, cultural and environmental factors (Ebbeling, 2002).

A similar study by Fisher, Rolls, and Birch (2003) explored the influence of large portions in a group of 30 preschool children. They presented the children with age-appropriate portions and larger than appropriate portions on two separate occasions at the lunch meal. They assessed intake by weighing the food before and after the children ate. Bite size was also assessed and children's comments regarding portions were recorded. The subjective data indicated that the children did not recognize a difference in the amount of food presented at the two meals. However, the children consumed 25% more of the entrée, and 15% more total Kilocalories of the large portion lunches. The

researchers observed that the children took larger bites and did not cut back total intake to compensate for this at the large portion meal. In addition, the researchers allowed the children to serve themselves at another lunch meal, and they observed a 25% decrease in the consumption of the main dish compared to when children were served the larger portion.

Fast food consumption is another factor to consider as a possible contributor to the obesity epidemic. A study by Utter, Scragg, Schaaf, Fitzgerald, and Wilson (2007) documented a relationship between takeout food and BMI levels in a group of 3,250 children ages 5 to 14 years living in New Zealand ($p = 0.04$). The researchers collected information specifically on takeout food consumption at lunch during school which points to the effect the school environment can have on nutrient intake and weight status.

Data from the Bogalusa Heart Study indicates that meal patterns have changed substantially since the 1970's (Nicklas et al., 2004). Breakfast consumption patterns have been of particular interest to several researchers because of the theorized link between skipping breakfast and increased body weight. O'Dea and Wilson (2006) reported on a national study conducted with 4,441 children in Australia. The nutritional quality of the participants' breakfast was measured using a questionnaire and weight and height measures were obtained by the researchers. They found a negative association between nutritional quality of breakfast for participants with low socioeconomic status and BMI levels ($p < 0.01$). Similarly, Utter et al. (2007) established a significant relationship between skipping breakfast and increased BMI levels in a group of 3,250 children in New Zealand ($p = 0.007$).

The number of meals eaten together as a family is another factor which may relate to a child's weight status. Epstein, Valoski, Wing, and McCurley (1994) reported that the number of meals children ate at home was one of several variables that predicted success in weight loss programs in children. More data on the importance of family meals is presented in a later section pertaining to environmental influences on obesity.

Physical Inactivity

Positive energy balance results not only from excess energy intake, but also from decreased energy output. Thus, one cannot solely focus on dietary factors as the cause of the overweight epidemic. The increased sedentary lifestyle of Americans in the past decades must also be considered. Thus, it is important for researchers to measure physical activity patterns when exploring factors related to weight and adiposity. This can be done through a variety of methods in children including observation by caregivers, use of activity monitors, and standardized questionnaires.

The Netherland's Physical Activity Questionnaire (NPAQ) is a standardized questionnaire that was developed for young children less than 10 years of age and is intended to be completed by parents or teachers (Janz, Broffitt, & Levy, 2005). It assesses children's daily activity preferences and typical behaviors that are thought to be associated with physical activity levels in a short questionnaire format. It does not require that the respondents quantify duration, frequency, and intensity of the child's physical activities which is an advantage since these components of activity are hard for untrained individuals to accurately estimate.

Janz et al. (2005) tested the NPAQ for reliability and validity in a group of 204 children ages four to seven years. Spearman's correlation coefficient (ρ) was used because data did not meet the assumptions to use a parametric test. Moderate to good test-retest reliability was observed for all items (overall NPAQ $\rho = 0.61, p < 0.01$; TV viewing $\rho = 0.68, p < 0.01$). The NPAQ results were compared to accelerometer readings to determine validity and they were found to be significantly related, although the relationship was low to moderate (overall NPAQ $\rho = 0.33, p < 0.01$; TV viewing $\rho = -0.14; p < 0.05$). The researchers also divided the data into groupings to measure the tool's ability to sort children into activity categories of low, moderate, and high. They calculated an odds ratio of 2.7 for children with a high NPAQ score to be in the high category for activity and found that children who watched the most TV were 10% less likely to be in the high category. The authors concluded that the NPAQ was a good tool for classifying children into activity categories and that the tool was an easy and practical method for measuring physical activity in young children.

The AAP has established physical activity recommendations at various stages throughout a child's development (Council on Sports Medicine, 2006). They suggest that parents of infants and toddlers provide environments for play that are safe and foster physical activity and introduce them to unstructured outdoor play under parental supervision. Pre-school children, ages 4 to 6, should be prompted to engage in free play that is fun and encourages them to explore their environment. Children this age should also be able to walk with family members without the aid of a stroller for reasonable distances. Children ages 6 to 9 should be encouraged to continue free play and can begin

participating in organized sports that they enjoy. By 10 to 12 years of age, children are more agile and have developed better motor skills allowing them to participate in more advanced sports. They may also start to participate in supervised weight training using lighter weights and more repetitions. It is important that children identify and participate in physical activities they enjoy by the time they reach adolescence. Children of all ages should be encouraged to participate in activities they enjoy with family members and friends and to limit sedentary behaviors.

Screen time. Television (TV) is a sedentary activity in which many children engage. The AAP (2001) recommends that children two years and older restrict media time to one to two hours per day for several reasons, including its link to weight gain. However, many American children exceed this limit. Lumeng, Rahnam, Appugliese, Kaciroti, and Bradley (2006) reported that approximately two-thirds of a group of 3-year old children watched TV more than two hours per day ($N = 1,016$). Lumeng et al. (2006) concluded that TV exposure equal to or greater than two hours per day was an independent risk factor for a child being overweight (odds ratio = 2.92, confidence interval = 95%, $p = 0.006$). TV exposure was defined as being in a room where the TV was on which the authors stated was a more appropriate definition for children this age who seldom sit still for long periods without other activities. Therefore, their findings suggest that TV, even as background noise, may have detrimental effects on weight status by deterring children from engaging in more physical activities. They also explored several confounding variables including hours per day in non-parental care, maternal depressive symptoms, income-needs ratio, child behavior problems, quality of the home

environment, and proportion of TV exposure that was educational. None of these significantly changed the relationship observed between TV exposure and overweight status at age three. The authors pointed out that a cause-effect relationship could not be established due to the cross-sectional nature of the study.

The positive correlation between hours spent watching TV and body weight may result from TV taking the place of physical activities, thus decreasing energy output (Vandewater, Shim, & Captovitz, 2004). Other potential contributors may be advertisements for unhealthy foods and the tendency to snack while watching TV (Lumeng et al., 2006). Not only do advertisements encourage unhealthy snacks, they may also convey inaccurate nutrition messages to children. Vandewater et al. pointed out that the actors in the commercials are typically of a normal body weight which may prevent children from recognizing the relationship between high fat, high sugar foods, and becoming overweight.

Other researchers have focused on the impact of watching TV during mealtimes. Coon et al. (2001) described how food consumption patterns of fourth through sixth grade children (N = 91) are influenced by watching TV during meals. Data collected included three 24-hour diet recalls from the children. Parents were queried about frequency of TV use during meals and the number of quick and easy to fix suppers prepared per week. Additionally, the nutrition knowledge, attitudes, and norms of the parents were measured. The chief findings were that children who watched TV during two or more meals per day (n = 41) consumed more meat, pizza, salty snacks, soda, and caffeine while consuming fewer fruits, vegetables, and juices than children who never

watched TV or only watched TV during one meal per day (n = 50). Other variables positively related to TV use during meals included lower household income, lower education of mother, single parent household, and number of nights quick meals were prepared. On the other hand, knowledge regarding nutrition and diet were negatively associated with TV use during meals. These data suggest that TV not only influences a child's desire for unhealthy snack foods, but it may also impact the overall diet of children by under-emphasizing foods such as fruits and vegetables which are rarely advertised.

Other researchers report opposing findings. Vandewater et al. (2004) failed to find a link between TV exposure and body weight in a sample of 2,831 children ranging in age from one to twelve years. Other forms of media use were measured in addition to television use including electronic game use, computer use, and print use which captured time spent reading and being read to. Twenty-four hour time-use diaries were used to collect the data, and parents recorded all the child's activities for two random days that included one weekday and one weekend day. A significant correlation was noted between video game use and weight, although there was no evidence of an association between TV use and body weight. Other relevant findings were that more time spent in sedentary activities, such as talking on the phone, puzzles, board games, and relaxing, were positively associated with weight while non-game computer use and use of print media were negatively associated with weight. This study illustrates the importance of obtaining information on all forms of media use when collecting data in both research and clinical settings.

Physical activity levels. The Council on Sports Medicine and Fitness and the Council on School Health (2006) summarized several issues that may play a part in the physical inactivity observed among children. They included inactive role models, competing demands/time pressures, unsafe environments, lack of recreational facilities or insufficient funds to begin recreation programs, and inadequate access to daily physical education (PE). Several of these factors were identified in the literature and the issue of children's perceptions about exercise surfaced, as well.

Snethen and Broome (2007) conducted semi-structured interviews with 17 children ages 8 to 12 years who were classified as overweight (BMI for age $\geq 95\%$ tile) to determine their perceptions of weight, exercise, and health status. The children's responses were studied by the investigators to see if there were similarities. One of the questions asked pertained to exercise. Several of the children reported exercising more in the summer than in the winter. The only consistent physical activity noted in the children's routine was that provided at school. It appeared they were not getting the recommended duration and frequency of exercise, although the researchers did not quantify the children's current physical activity levels, and some children seemed unaware of what the recommendations were. One respondent stated she thought exercise should be done once a week while another said twice a week. Although from a small sample, these findings suggest that additional education on exercise duration and frequency may be needed and that school-based physical activity programs are crucial since this may be the primary site for exercise activities.

Lazaar et al. (2007) conducted a six-month intervention study in 425 school children ages 6 to 10 years to see if physical activity would improve body composition. Nineteen schools participated in the study with 14 schools receiving a physical activity program and five schools serving as the control. The physical activity program consisted of two weekly 1-hour exercise sessions that included a variety of exercises. Anthropometric measurements taken on the subjects included weight, height, waist circumference, and skinfold measures of the biceps, triceps, subscapular, and suprailiac. There were no differences in the anthropometric parameters of the control and intervention groups at baseline. However, the girls in the intervention group were found to have significantly lower BMI z-scores, waist circumference measures, and sum of skinfolds and higher levels of fat free mass after the six-month intervention ($p < 0.05-0.001$). The boys in the intervention group showed significant improvements over the control group for BMI z-scores and fat free mass ($p < 0.001$). This adds to the evidence that school-based physical activity interventions are effective in addressing the overweight crisis in children.

A study conducted in Australia investigated the relationship between the home environment, physical activity, and dietary behaviors of 280 children ages 3 to 4 years (Spurrier, Magarey, Golley, Curnow, & Sawyer, 2008). Home interviews were conducted by the researchers to determine characteristics of the home environment and standardized questionnaires were given to the parents to assess the children's sedentary behaviors and physical activity levels. Children were also weighed and height measures taken in order to calculate BMI. Children were more likely to play outside when their

mothers walked >30 minutes per day ($p = 0.008$), when their mothers were more involved in organized sports ($p = 0.04$), when they had larger backyards ($p = 0.001$), and when they had more outdoor play equipment ($p = 0.003$). On the other hand, children were likely to engage in more indoor sedentary behaviors when there were fewer rules in the household about TV viewing ($p < 0.001$) and when there was a playstation in the household ($p = 0.02$). These findings indicate that the home environment and parental behaviors appear to influence physical activity patterns among children.

Environmental Influences

A child's environment influences both food intake and activity patterns. There are various layers of a child's environment including the home, school, and community but perhaps the most influential of these is the home environment because children tend to mimic the actions of their parents. The AAP (2003) cited parental obesity as a more powerful predictor of adult obesity than current weight in children less than 3 years old. In addition, the Surgeon General (2001) reported that overweight adolescents had a 70% chance of becoming overweight adults, and that number increased to 80% if one or more parents were overweight or obese. One may conclude from these statements that genes are to blame, but a number of studies point to environmental influences in the home instead of genetics as the culprit (Gable, Chang, & Krull, 2007; Gibson et al., 2007; Spurrier, et al., 2008).

Several aspects of the home environment have been reported to influence a child's food intake and weight status. Gable, Chang, and Krull (2007) studied a group of 8,000 U.S. children at four time points from kindergarten to third grade. They calculated odds

ratios (OR) for the children becoming overweight using several variables. They found the odds of the child being overweight by third grade increased by an OR of 1.08 for each breakfast or lunch meal not eaten with the family during the first two years. They also reported that fewer family meals per week was a risk factor for becoming persistently overweight.

Gibson et al. (2007) identified single-parent households as another predictor of a child's BMI. They studied 329 Australian children and 265 of their mothers in a prospective study from January 2004 to December 2005. Children in households with a single-parent mother had significantly higher BMI z scores than those in two-parent households ($P < 0.05$). They also reported a link between maternal BMI and the children's BMI z scores ($P < 0.01$). Other family factors such as maternal depression and parenting style were assessed, but no other associations to the children's BMI z scores were found.

The study presented previously by Spurrier et al. (2008) also illustrates the impact of the home environment on eating behaviors. This study showed that children who had limited availability to fruit juice and high fat, high sugar snacks consumed more fruits and vegetables ($p = 0.02$ and 0.0009 , respectively). On the other hand, those whose parents prompted them to eat everything on their plates, offered food rewards for finishing a meal and allowed children to eat while watching TV consumed fewer fruits and vegetables ($p = 0.007, 0.04, 0.01$, respectively). There were also associations found between fewer meals eaten together as a family and increased sugary beverage

consumption ($p = 0.05$), dinner eaten while watching TV ($p = 0.02$) and use of food rewards for desired behaviors ($p = 0.02$).

Cultural Beliefs

Cultural beliefs may also play a role in how one perceives weight and its associated health risks. There has been a reported disconnect between how individuals perceive their weight or the weight of their children and established weight standards. A study by Hackie and Bowles (2007) measured mothers' perceptions of their children. The children in the study were participants in the WIC program in Nevada and were between 2 and 5 years of age. Sixty-two percent of the mothers did not consider their children to be overweight even though all 38 children in the study were classified as overweight with a BMI for age ≥ 95 percentile. The researchers did not find age or educational level of the mother to be a factor in the results. A larger study consisting of 99 overweight children examined this same issue and found that 79% of the mothers with overweight children did not consider their child to be overweight (Baughcum, Chamberlin, Deeks, Powers, & Whitaker, 2000). The researchers found an association between low maternal education and failure to perceive the child as overweight.

Mothers appear to define overweight for their children differently than health experts. Eighteen low-income mothers with overweight children participated in focus groups to determine perceptions about their children's weights (Jain et al., 2001). The mothers reported being teased about weight and developing limitations in physical activities as indications that their child was overweight as opposed to where they plotted on a growth chart. The mothers agreed with health experts that children should be active

and eat healthy foods indicating that weight management interventions may be more effective if healthy lifestyle factors are the focus instead of weight.

Jain et al. (2001) also examined views of mothers on why children become overweight and obstacles in preventing and treating childhood obesity. Several themes emerged from the focus groups including: 1) mothers believed that nature, genetics, or heredity determined weight; 2) parents' behaviors and family environment also influenced a child's diet and activity patterns; 3) mothers had trouble controlling their children's eating habits; 4) mothers used food to shape children's behavior; 5) mother's control over her child's diet was challenged by other family members; and 6) mother's own obesity affected her outlook on children's weight management.

Genetics

Understanding the role that genes play in obesity development is an important component of prevention and treatment, but distinguishing the influences of genetics from environment is difficult. Nelson, Gordon-Larsen, North and Adair (2006) pointed out that the environment in which a child is raised can affect his health behaviors even into adulthood. Thus, attempts to measure genetic and environmental influences separately become complicated.

Nelson et al. (2006) designed a study to measure the cumulative effects of genetics and influences of the home environment on BMI and related health behaviors by studying siblings who lived together for at least ten years during childhood and following them into adulthood (N=4,368). They assessed the participants at baseline when they were in grades seven to twelve, one to two years later when they were a mean age of 16.5

years, and a third time when they were young adults with a mean age of 22.4 years. Twin pairs, including 580 monozygotic (MZ) and 726 same-sex dizygotic (DZ) twins were included in the analysis. The researchers analyzed correlations for several variables including weight, physical activity, and various dietary factors between the twins living together and those living apart in young adulthood to see if they were related. They found significant differences for two variables among the MZ twins including BMI and sedentary behaviors. Those who were living apart had greater differences in BMI levels ($p < .05$) but more similar sedentary behavior scores ($p < .05$) than those living together at both follow-up assessment time points. The DZ twins who were living apart in adulthood had greater differences in mean vigorous physical activity bouts per week ($p < .05$) than the DZ twins living together at both follow-up assessments. The authors stated that the data highlights the strong influence of the household and physical environment on health behaviors.

Research indicates that one's genetic makeup can result in varying hormone and neurotransmitter profiles causing some to be more susceptible to obesity development than others (AAP, 2003). Scientists have explored many hormones including insulin, growth hormone, leptin, and ghrelin to determine how they may affect appetite control and energy metabolism (Grottoli et al., 2003; Mark, Correia, Rahmouni, & Haynes, 2004; Soderberg, Ahren, Eliasson, Dinesen, & Olsson, 2002; Yildiz, Suchard, Wong, McCann, & Licinio, 2004).

Leptin is a hormone derived from adipose tissue and is an indicator of energy stores and energy balance (Grottoli et al., 2003). Animal studies suggest it may be

responsible for appetite suppression and weight loss in obese mice (Gaucher, Miyamoto, & Benner, 2003). However, many obese adults have been found to have excess leptin levels (Gaucher et al.). Several researchers have reported that individuals with hyperinsulinemia tend to have increased leptin levels (Grottoli et al; Mark et al., 2004; Soderberg et al., 2002).

Ghrelin is a hormone that plays a role in the regulation of growth hormone secretion and energy balance (Yildiz et al., 2004). Yildiz et al. showed that ghrelin increased pre-prandially, but decreased after meals. They stated that this may be in response to insulin secretions after feeding. They concluded that ghrelin was inversely related to leptin and noted low ghrelin levels in obese individuals.

Adiponectin is another hormone which appears to affect metabolism. Yildiz et al. (2004) reported that adiponectin may work with insulin enhancing its action. Low levels of adiponectin were also found in obese individuals (Yildiz et al.).

The impact these and other hormones and neurotransmitters may have on obesity development is a crucial part in understanding the best approach for prevention and treatment of the problem. Most experts agree that it is the synergistic effect of genes working with environmental, social, and psychological factors that cause an individual to be overweight (AAP, 2003). Thus, a comprehensive approach to obesity management must include a combination of all of these factors.

Illnesses

Certain medical conditions have been linked to weight gain in children including Down's, Prader-Willi, and Bardet-Biedle syndromes (Council on Sports Medicine, 2006).

These conditions are rare, accounting for only 1-2% of overweight children (Council on Sports Medicine). Other single-gene disorders such as congenital leptin deficiency are even more uncommon, but have been observed. Farooqi et al. (2002) treated three morbidly obese children with congenital leptin deficiency with leptin for four years. They found that the leptin therapy decreased appetite, fat mass, hyperinsulinemia, and hyperlipidemia in the children. They also observed an increase in plasma thyroid hormone levels and pubertal development occurred at an appropriate age. Additionally, improvements were noted in immune function of the subjects. It should be noted that while treatment with exogenous leptin was found to be effective in reducing weight in this small group, most obese adults have been found to have high leptin levels instead of deficiencies (Gaucher et al., 2003). Consequently, it is unlikely that leptin therapy would be an effective treatment for most overweight or obese individuals.

Prevention of Overweight in Childhood and Adolescence

It is imperative that modifiable factors be identified so that prevention strategies can be developed and implemented in light of the escalating rates of obesity. The literature suggests it is a combination of factors that causes one to become overweight; therefore, comprehensive programs for prevention and treatment must address multiple issues including dietary factors, physical activity, environmental issues, and cultural beliefs regarding weight (AAP, 2003; Council on Sports Medicine, 2006; Nemet et al., 2005).

Nemet et al. (2005) conducted a randomized prospective study in a group of obese Israeli children ages 6 to 16 years that combined diet, physical activity, and behavior

modification (N = 40). The intervention group and their parents received the following:

- 1) four lectures over a three-month period addressing topics related to childhood obesity,
- 2) six meetings with a registered dietitian that included an initial twenty-four hour recall and a low Kilocalorie diet plan,
- 3) one-hour exercise program twice a week,
- 4) encouragement to do at least one other exercise lasting 30-45 minutes during the week,
- and 5) encouragement to decrease sedentary activities like watching TV and playing video games.

The control group received weekly nutrition counseling and was instructed to do physical activity at least three times per week. Both groups had anthropometric measures taken weekly during the three months and at a 1 year follow-up visit. Two-day food records, a physical activity questionnaire, and a progressive treadmill test were obtained at baseline, three months, and at the one-year follow-up visit. Blood samples were taken at baseline and three months to measure total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and triglycerides. The results at three months showed positive changes for the intervention group including significant decreases in body weight, BMI, percent body fat, energy intake, total cholesterol, and LDL-C. They also showed significant increases in habitual physical activity patterns and endurance time. On the other hand, the control group gained weight and had increases in body fat; they also showed a slight decrease in habitual physical activity patterns, and no changes were noted in their lipid levels. The 1-year follow-up visit showed that 90% of the intervention group had decreased BMI levels while 68% of the control group showed increased BMI levels. Body weight was found to be stable in the intervention group while the control group continued to gain weight.

Habitual physical activity significantly increased in the intervention group compared to the control. This study shows that a multidisciplinary team approach addressing diet, physical activity and behavior change is effective for both short and long-term weight management in children.

The AAP (2003) offered other advice to health professionals emphasizing the importance of early identification of weight gain in children. They discussed the importance of routine weight monitoring using the BMI for age growth charts as a guide. Growth charts are essential tools for health professionals, but it should be reiterated that Jain et al. (2001) found that parents may perceive weight differently than medical professionals. Therefore, it may be more effective to discuss behaviors such as healthy eating and exercise with parents instead of focusing on weight and growth charts.

Treatment of existing weight problems and early identification are necessary. However, prevention is the ideal approach for the childhood obesity epidemic. Several recent studies suggest that prevention measures should begin as early as infancy (Baird et al, 2005; Kinra, Baumer, & Smith, 2005; Martorell, Stein, & Schroeder, 2001; Stettler, Kumanyika, Katz, Zemel, & Stallings, 2003; Stettler et al, 2005). The developmental origins theory of disease is based on the notion that very early in life, even *in utero*, growth patterns may trigger chronic disease development (Barker, 2004).

Nutrition during the prenatal period and the first three years of life was examined in a review by Martorell and associates (2001). They focused on the effects of under-nutrition, over-nutrition, and breastfeeding and the impact these factors had, if any, on obesity later in life. Their primary conclusions were: 1) weak evidence exists for under-

nutrition as a risk factor for future obesity; 2) higher birth weight, especially greater than 10 pounds, is a fairly consistent indicator of fatness in adulthood across studies; and 3) breastfeeding appears to be protective against childhood obesity. They concluded that while prenatal over-nutrition, defined as high birth weight, may increase obesity and breastfeeding may decrease it, they are weaker predictors than other known risk factors such as excessive caloric consumption and physical inactivity.

Stettler et al. (2003) mentioned several possible critical periods for obesity genesis including fetal life, early infancy, the period of adiposity rebound, and adolescence. They stated that critical periods must be considered, along with causes of obesity, when developing prevention strategies. Other researchers had reported an association between rapid weight gain during infancy and childhood obesity. Stettler et al. focused on rapid weight gain during early infancy as a risk factor for obesity development in young adulthood. They conducted a cohort study in 300 African Americans and assessed the relationship of rapid weight gain during early infancy with obesity at age 20. Rapid weight gain was defined as an increase in the weight for age z score between birth and four months of ≥ 1 standard deviation. They found that 86 of the 300 subjects experienced rapid weight gain during the first four months of life, and of those, 14% became obese adults ($BMI \geq 30$). On the other hand, only 6% of the other 214 subjects who did not experience rapid weight gain in early infancy became obese adults. The investigators also categorized subjects as overweight-overfat which they defined as a BMI of ≥ 25 and a sum of the tricep and subscapular skinfold thicknesses of $\geq 85^{\text{th}}$ percentile. Fifteen percent of the subjects with rapid weight gain during early infancy

were found to be overweight-overfat at age 20 while only 6% of those without rapid weight gain were categorized as overweight-overfat using those parameters. These data point to early infancy as one possible critical period to consider in obesity prevention initiatives. However, the authors caution against dismissing other possible critical periods for obesity development. Instead, they suggest early infancy as one of several critical periods to explore.

Stettler et al. (2005) reported on results from another study with similar findings. European American adult subjects who had participated as infants in formula studies were contacted for current weight and height information (N = 653). The outcomes measure used by the researchers was overweight (BMI \geq 25; 32.3% of subjects) since the rate of obesity was low in the sample (5.7%) compared to the general population. Results were reported as medians and percentiles, as in the previous study, because variables were not normally distributed. A life-course plot was created to identify periods of weight gain that might be critical for overweight in adulthood. Two periods were recognized including birth to eight days and the period between birth and 112 days. The chief finding was that weight gain during the first week of life was significantly related to overweight status in adulthood. Furthermore, the risk of becoming overweight in adulthood went up 28% for every 100 gram increase in absolute weight gain during this time period. Although intriguing, these data are limited by several factors including: 1) birth weight was reported by the subjects' mothers when they were 8 days old, 2) adult weight was self-reported by the subjects, 3) few subjects were classified as obese, 4) only formula-fed infants were included in the study, and 5) the study was retrospective.

Consequently, it cannot be concluded that weight gain during the critical time periods identified by the researchers was directly linked to weight status in adulthood. Future studies should look at this question prospectively and explore potential confounding variables that may have influenced adult weight status.

Baird et al. (2005) conducted a systematic review of 24 observational studies that examined the association between infant growth or size during the first two years of life and obesity occurrence at any age after infancy. Twenty-two of the studies reviewed were cohort designs and two were case-controlled studies. The reviewers were interested in both size and growth as independent variables for obesity occurrence because both have been implicated in the literature as potentially affecting obesity progression, and both are important in understanding the overall development of the infant. The researchers' main findings were that the majority of studies reviewed reported an association between infant size and later obesity and a link between growth rate and later obesity. This review adds strength to the theory that obesity genesis may begin in infancy. However, the study was limited by several factors. The majority of the articles were considered to have medium to high bias because confounding variables were not controlled, or the studies had high attrition rates. In addition, the researchers were unable to perform a meta-analysis because variable definitions for infant size, growth, and obesity differed among studies.

Kinra and colleagues (2005) believe that obesity development is not dependent on one particular critical period, but instead develops progressively throughout the perinatal and postnatal periods. They conducted a historical cohort study of 1,335 children in

Southwest England in which they collected weight data at birth, 6 weeks, and 18 months from computerized health records. Data was then compared to the main outcome measure of BMI at age seven using multiple linear regression modeling. They found a positive association between the z scores for weight at all ages and BMI at age seven. Another model included weight gain and showed that all three weights contributed about the same to the variation in BMI suggesting the progressive nature of obesity development.

Methods for Assessing Body Composition

The composition of the human body has been outlined by Wang, Pierson, and Heymsfield (1992) using five different levels. These include the atomic, molecular, cellular, tissue-system and whole body levels. Each level defines body weight using different components.

The atomic level measures body weight by summing the elements that comprise the body including oxygen, carbon, hydrogen, nitrogen, calcium, phosphorus, sulfur, potassium, sodium, chloride, magnesium and a residual mass of other elements. Measurement of these elements is accomplished either by cadaver studies or whole body counting and neutron activation techniques in vivo. Neutron activation techniques determine the levels of body elements by measuring the “induced gamma activities in humans following exposure to neutrons (Cohn, Shukla, Dombrowski, & Fairchild, 1972, p. 487).” An irradiation facility is required to measure the gamma activity. There are only a few of these facilities world-wide.

Cadaver studies involve dissecting the body into various components including skin, adipose tissue, muscle, bones, organs and viscera (Clarys, Provyn, & Marfell-Jones, 2005). These components are further analyzed to determine their atomic makeup, water content, weight and density. These data provide estimates that can be used for development of more practical techniques for determining human body composition. For example, recent adult human cadaver studies have estimated the water content of adipose tissue, muscle, organs and bone to be 20%, 70%, 80% and 10%, respectively (Clarys et al., 2005). Such data are used when developing equations for body composition techniques.

It is essential that accurate and reliable methods for monitoring growth be available because of the mounting evidence that obesity and associated chronic diseases may originate early in life. It is especially important that body composition methods be available for the pediatric population considering the associations that have been made between adiposity and disease development. Body composition techniques either directly measure or estimate the various components of the body including fat mass, which contains small amounts of water, and fat free mass (FFM) comprised of bone mineral, protein, and water (Ellis, 2001). Several techniques exist for measuring body composition but not all are practical or accurate for use in infants and children.

The ideal body composition method would directly measure all of the components of FFM. In addition, a method that measures the distribution of body fat would be idyllic since studies indicate body fat distribution may be just as crucial as overall body fat in the

prediction of chronic disease (Clarys et al., 2005). It is also important for a body composition method to be fairly non-invasive, especially when assessing children.

Criterion Methods

To assess the accuracy of a body composition technique, one must select a criterion or reference method as a standard for comparison. Multi-compartment models are often used as reference methods because they measure two or more of the components of the FFM (Reilly, 1998). Fat-free mass is composed of bone mineral, protein, and water (Ellis, 2000). It is important to measure the various components of FFM because the overall composition of the FFM varies based on age and gender (Baumgartner, Heymsfield, Lichtman, Wang, & Pierson, 1991; Ellis, 2000; Hewitt, Going, Williams, & Lohman, 1993; Reilly; Wang et al., 1999).

Four-compartment models. A four-compartment (4-C) model that takes into account body water, protein, bone mineral, and fat mass is ideal for measuring body composition (Ellis, 2000; Fields & Goran, 2000; Reilly, 1998; Roemmich, Clark, Weltman, & Rogol, 1997; Sopher et al., 2004). Four-compartment models measure three components of the FFM including body water, protein, and bone mineral. They use a dilution technique to determine total body water (TBW), neutron activation analysis for measuring body protein, and dual-energy x-ray absorptiometry (DXA) for bone mineral content (Ellis, 2000; Sopher et al., 2004). Fat mass can be calculated by subtracting the sum of those values from body weight once TBW, protein, and bone mineral content are known (Ellis, Shypailo, Abrams, & Wong, 2000).

There are two major drawbacks in using the 4-C model. A neutron activation analysis system is required to determine body protein that requires a neutron source and a gamma radiation detector (Cohn et al., 1972). Very few facilities are available worldwide that can perform this technique. Second, it requires moderate amounts of radiation exposure. Thus 4-C models are often an impractical choice for assessing body composition, especially in children (Ellis, 2000; Fields & Goran, 2000; Wang et al., 2003).

Three-compartment models. An alternative to the 4-C model is the three-compartment (3-C) model that combines two of the components of FFM into one category (Ellis, 2000; Reilly, 1998; Roemmich et al., 1997; Withers et al., 1998). Thus, the three compartments considered are fat mass, TBW, and FFM. Two 3-C models have been developed including a water-density model that combines protein and mineral into a solids category and a mineral-density model that combines water and protein into a lean soft tissue category (Roemmich et al.). These methods require measuring TBW and body density, using standardized equations (Roemmich et al.; Withers et al.). Since this eliminates the need to determine protein content separately, it is a more practical approach for researchers.

On the other hand, 3-C models require making certain assumptions about the densities of fat mass and FFM and the hydration of the FFM in order to determine body density (Baumgartner et al., 1991; Hewitt et al., 1993; Wang, Heshka, Wang, Wielopolski, & Heymsfield, 2003). Most experts agree that the density of fat mass is fairly consistent at 0.9 g/cm³. It is also widely accepted that the density of FFM is 1.10

g/cm³, and the hydration of FFM is 73% in human adults (Baumgartner et al.; Wang et al., 1999; Withers et al., 1998). However, the density and hydration of FFM has been shown to vary based on age, gender, and ethnicity (Baumgartner, et al.; Hewitt, et al; Wang, et al., 2003). Therefore, it is especially important to adjust for age and gender differences in the chemical composition of the FFM when using body composition techniques in children. Fomon, Haschke, Ziegler, and Nelson (1982) presented data on the density and hydration of FFM for children ranging in age from 5 months to 10 years. Most experts agree that a 3-C model is an acceptable alternative to a 4-C model as a criterion method when these age and gender specific adjustments are made (Ellis, 2000; Schoeller et al., 2005).

Modified two-compartment models. Two-compartment (2-C) models divide the body into fat mass and FFM (Ellis, 2001). TBW analysis is an example of a 2-C model that has been recognized as a criterion method by several researchers (Reilly, 1998; Schoeller et al., 2005; Yao, Roberts, Ma, Pan, & McCrory, 2002). Fat mass and FFM can be calculated using standard equations if TBW is known (Sheng & Huggins, 1979). This technique is more credible when the age and gender-specific hydration of FFM constants proposed by Fomon et al. (1982) are incorporated into the formula (Reilly, 1998). Reilly referred to this as a modified 2-C model.

A widely used method for measuring TBW is to use a technique that applies dilution principles dating back to the 1800's (Sheng & Huggins, 1979). The dilution principles state that if a known amount of a tracer has time to become evenly distributed throughout the body's fluids, the concentration of the tracer in a sample of body fluid can

be used to determine TBW (Ellis, 2001; Sheng & Huggins). Ellis (2000, p.655) outlined the four assumptions that apply when using dilution techniques including “1) the tracer is distributed only in the exchangeable pool, 2) it is equally distributed within this pool, 3) it is not metabolized during the equilibration time, and 4) tracer equilibration is achieved relatively rapidly.”

Deuterium oxide, a stable isotope of hydrogen, was discovered in 1932 and has been used extensively as a tracer in determining TBW in both human and animal studies (Sheng & Huggins, 1979; Yoa et al., 2002). Moore conducted dissection studies in animals in 1946 that showed TBW estimates using dilution techniques compared well with direct measures of TBW (Sheng & Huggins). Recent reports suggest that dilution techniques are accurate in predicting percent body fat within 1-2% (Bray, DeLany, Volaufova, Harsha, & Champagne, 2002; Ellis, 2000; Schoeller et al., 2005; Wang et al., 1999). However, one must take into account that approximately 4% of the hydrogen tracers exchange with hydrogen molecules outside the aqueous space in order to achieve values within 1-2% (Ellis, 2000; Wang, 1999). Therefore, a mathematical adjustment of 4% must be made in the calculation so that TBW will not be overestimated when using deuterium oxide as the tracer.

Popular Methods for Assessing Body Composition

Some of the most popular methods for assessing body composition include skinfold (SF) measures, bioelectrical impedance analysis (BIA), underwater-weighing (UWW), DXA, and ADP (Ellis, 2000). Research and clinical experience have shown that some of these body composition techniques may not be accurate or practical for use

in children (Ellis, 2000; Fields & Goran, 2000; Pintauro, Nagy, Duthie, & Goran, 1996; Roemmich et al., 1997; Sopher et al., 2004). Following is a description of these techniques along with a critique of their usefulness in pediatric body composition assessment.

Skinfold measures. Skinfold measures (SF), one of the most common methods used to predict body fat, represents a 2-C model. It requires that a trained technician take SF measurements at various sites on the body using calipers that measure subcutaneous fat in millimeters (Ellis, 2001; Reilly, 1998). These values are then plugged into standard equations for body fat prediction. Ellis (2001) reported that over one-hundred SF equations exist for body fat prediction. This method is a popular choice because it is fairly inexpensive, non-invasive, and can easily be administered in field studies when no large equipment is available (Reilly).

However, SF measures have limitations. They are based on the assumption that total body fat can be estimated based on the subcutaneous fat layer. In addition, technical skill is also required to achieve accurate SF measures (Ellis, 2001). Researchers have reported SF measures vary significantly when compared to 3-C and 4-C models (Bray et al., 2002; Roemmich et al., 1997). Roemmich et al. found that the Slaughter equations using tricep, subscapular, and calf SF had a percent body fat total error of approximately 5%. Bray et al. had similar findings showing that the Slaughter equation differed from a criterion method by approximately 4%. Bray et al. also found that an equation by Ellis underestimated percent body fat by 7.31% when compared to a 4-C model. Ellis (2001)

stated that SF measures were most valuable for tracking changes in subcutaneous fat over time rather than to predict body fat.

Bioelectrical impedance analysis. BIA is another 2-C method often used to measure body fat. This technique is based on the principle that the aqueous, electrolyte-rich tissues of the body have good conductance qualities while body fat and bone do not (Ellis, 2000). It requires that a weak electric current pass through the body. This is accomplished by placing two electrodes on the wrist and two on the ankle in order to measure the resistance of flow to the electric current (Ellis, 2000). The typical frequency that is used for most commercial instruments is 50 kHz but measures can be taken at any frequency (Ellis, 2001). This allows one to predict body fat because the drop in voltage between two electrodes is proportionate to fluid volume and, therefore, can be used to predict TBW (Ellis, 2001). Estimates of fat mass and FFM can be calculated using standard equations when TBW is known as explained previously.

Several factors influence the accuracy of the BIA technique. First, assumptions are made when estimating TBW with the BIA technique and these assumptions have limitations. The assumptions are 1) the body is considered a cylindrical conductor, 2) the cylindrical conductor is proportionate to the individual's height, and 3) body impedance is determined based solely on the measured resistance and not the reactance of the voltage signal because reactance is so small it is considered insignificant (Ellis, 2001). However, the body is not a perfect cylinder and the reactance of body impedance is not zero (Ellis, 2000). Second, the use of 50 kHz frequency has been reported as problematic because the fraction of the current that passes through the body at that

frequency is not known (Ellis, 2001). Finally, BIA results are influenced by previous exercise and hydration status (Ellis, 2001).

Roemmich et al. (1997) compared two BIA models to a 4-C model in 10-13 year old children (N = 47). They found that BIA under-predicted body fat by 5-6% using the Houtkooper et al. and Boileau models. Similarly, Bray et al. (2002) compared BIA to a 4-C model in a group of 12 year-old children and found that BIA underestimated body fat as much as 6-12% using the Goran et al. and Suprasongsin et al. models (N = 114). Although these studies suggest accuracy is not optimal, Ellis (2001) reported the precision of BIA to be 1-2%.

BIA continues to be a popular method for body fat assessment despite the limitations and errors reported. It is a painless, inexpensive procedure to perform that only takes a few seconds to administer and the equipment is portable (Ellis, 2000).

Underwater weighing. Underwater weighing (UWW) is an example of a densitometry technique meaning it determines an individual's density and uses that variable to estimate body fat using standardized equations. Densitometry techniques are classified as 2-C models because they divide the body into fat mass and FFM portions only. The equation most commonly used to estimate body fat using density is the Siri equation which was established using Caucasian adult males (Bray et al., 2001). It is based on the assumptions that the density of FFM is 1.1 g/cm³ and the density of fat mass is 0.9 g/cm³ (Ellis, 2000; Ellis, 2001). This assumption for the density of FFM is not accurate for children as discussed previously (Baumgartner, et al., 1991; Hewitt, et al.,

1993; Wang, et al., 2003). Thus, adjustments must be made based on the equations presented by Fomon (1982) when applying this technique to children.

Body mass and body volume must be measured in order to establish body density (Ellis, 2000). Density is calculated by dividing body mass by body volume once these two variables are known (Ellis, 2000). Body mass is quantified by simply weighing an individual on a calibrated scale. However, determination of body volume is more difficult. The UWW technique predicts body volume using water displacement which requires that one be submerged in a tank of water (Ellis, 2000).

The difference in water volume before and after submersion is used to determine body volume. Several limitations must be considered when using the UWW technique. First, its practicality may be limited because of the requirement for individuals to be immersed in water and while under water to exhale as much air as possible out of the lungs (Ellis, 2000; Ellis 2001). This procedure may be difficult for some individuals, and the results will not be accurate if it is not done properly (Ellis, 2000). Second, the residual lung volume that remains must be factored into the equation for predicting volume (Ellis, 2000). This can either be measured using a closed-circuit spirometer system or predicted based on previously obtained values (Ellis, 2000). Errors can result when using both measured and predicted values and these errors can significantly affect the accuracy of the measures obtained. Ellis (2000) reported that an error of 100 milliliters in residual lung volume could produce a 1% error in body fat percentage estimates. Prediction of residual lung volume may produce errors of 300 to 400 milliliters causing a 3-4% error in body fat measures (Ellis, 2000). A third limitation

with the UWW technique is that the equations used to predict body fat use constant values for the density of fat mass and FFM as mentioned above (Bray et al., 2001; Ellis, 2000; Ellis, 2001; Fields & Goran, 2000; Roemmich et al., 1997). Therefore, adjustments must be made in the mathematical formula to account for differences in the FFM or the accuracy of the values obtained will be decreased. Finally, an UWW system requires constant maintenance and it is not portable like the other techniques discussed (Ellis, 2001).

The accuracy of UWW has been reported to range between 1-5% (Bray, 2002; Ellis, 2000; Roemmich et al., 1997). Roemmich et al. compared results of UWW using the Siri equation and the Lohmon age- and gender-adjusted equations to a 4-C model in a group of 47 children ages 10 to 14 years. They found that the Siri equation over-estimated body fat by 5.15% and the Lohmon equations over-estimated by 1.14%. Roemmich et al. also noted that both equations were less accurate in male subjects and children with lower body fat levels. This is consistent with work by Fields and Goran (2000). They found that UWW under-estimated fat mass in children with high body fat and over-estimated fat mass in leaner children when compared to a 4-C model. Bray et al. (2002) reported that UWW using the Siri equation differed from a 4-C model by an average of 3.11% in a group of 12 year-old children (N = 114). A review by Ellis (2000) reported UWW to be accurate within 2-3% and to have precision of 1-2%.

Air-displacement plethysmography. Air-displacement plethysmography (ADP) is a 2-C densitometry technique that applies formulas such as the Siri equation to estimate body fat using a technique similar to UWW (Fields, Goran, & McCrory, 2002). The

ADP method determines body volume by placing an individual in a closed air-filled chamber and measuring how much air they displace once inside the chamber (Ellis, 2000). Procedures for the ADP method are described by Fields et al. (2002) as follows. A calibration phase taking approximately 3 minutes to perform must be conducted prior to the individual entering the chamber. A baseline volume measure is obtained of the empty chamber. Then a second volume measure is taken using a calibration cylinder to ensure the machine is accurately measuring volume. The participant is weighed on the instrument's scales and personal information including weight, height, age and gender are entered into the software. The participant enters the chamber where at least two whole body volume measures are taken, each requiring approximately 50 seconds. A third measure is required if the two measures are not within a tolerance level 0.2%. An optional third step may be done to measure the thoracic gas volume (TGV) of the individual. This is much like the panting maneuver in pulmonary function tests. This step may be skipped and a predicted TGV value may be applied if the individual is unable to complete the procedure.

The concept of air displacement is based on gas laws established by Boyle that state pressure and volume are inversely related at a constant temperature (Fields et al., 2002). Poisson's Law has been applied to the most modern devices for measuring body volume through air displacement because maintenance of a constant temperature is difficult to achieve once inside the chamber. Poisson's Law makes adjustments to Boyle's Law by describing the relationship of pressure and volume under adiabatic conditions. Air inside the chamber behaves adiabatically for the most part because

fluctuations in the air temperature occur once a human body is inside. However, the air trapped in clothing, around skin and hair and in the lungs sustains isothermal conditions. Corrections must be made for these isothermal areas in order to achieve accurate volume measures because isothermal air is compressed 40% more than adiabatic air. This is done by having individuals wear a tight-fitting swimsuit and cap that completely covers the hair. The air in the lungs is accounted for by measuring or predicting TGV as described previously and surface area artifact is calculated in order to adjust for the air around the skin.

The ADP method has some drawbacks. The procedure may not be well-tolerated by some individuals who fear entering a small, confined space and the optional measure of TGV makes it difficult for some to comply. Prediction equations for TGV in adults have been established to circumvent this problem (Bosey-Westphal et al., 2005; Fields, Hull, Cheline, Yao, & Higgins, 2004; Higgins et al., 2006). However, inaccuracies have been reported when applying these prediction equations to children (Bosey-Westphal et al.; Fields et al.; Higgins et al.). Bosey-Westphal et al. found that an error of 2% resulted when age-appropriate adjustments were not made for TGV estimates.

Thus, Fields et al. (2004) developed prediction equations for TGV in children ages 6 to 17 years. First, they developed TGV prediction equations using 224 healthy boys and girls then cross-validated them in another phase of the study using a new cohort of children ($n = 62$). They found that measured versus predicted TGV values were highly correlated ($r = 0.93$, $p < 0.0001$) as were the percent body fat estimates from predicted

versus measured TGV values ($r = 0.99$, $p < 0.0001$). Further, they found no significant differences between the predicted and calculated percent body fat values.

Another limitation with ADP is that the equations used to predict body fat use constant values for density of FFM as was the case with UWW (Ellis, 2000). Therefore, age and gender-specific corrections must be applied to those equations in order to obtain accurate results when using ADP in children (Bosey-Westphal et al., 2005). Bosey-Westphal et al. found that ADP over-estimated percent body fat by 4.8% when age- and gender-specific constants for the density of FFM were not applied to equations.

Another consideration when using ADP in children is the estimation of surface area artifact that was discussed previously. Surface area artifact equations must be adjusted based on an individual's age (Bosey-Westphal et al., 2005). Radley and Fields (2006) stated that an error of 0.297% could be expected if age-appropriate adjustments were not made.

There are some advantages of ADP in spite of these problems. Submersion in water is not required, and the test takes less than five minutes to perform. These factors make ADP a more practical option for assessing body composition in children than UWW (Nunez et al., 1999). Additionally, high accuracy and precision of ADP have been reported in the literature. Ellis (2000) reported the ADP method to be accurate within 2-3% of a criterion method, and repeated measures were stated to be consistent within 1-2% in adults. Similarly, Demerath et al. (2002) found the reliability of ADP to be 1.6% for adults and 1.8% for children ($N = 50$).

Fields and Goran (2000) also assessed the accuracy of the ADP method. Their data on the accuracy and bias of fat mass among DXA, UWW, ADP and TBW with deuterium oxide using a 4-C model as the standard for comparison in 25 children ages 9 to 14 years was of particular interest. Regression analyses were used to measure accuracy of fat mass by comparing a 4-C model to each of the other methods. DXA and TBW had a slope of 0.84 and 0.85, respectively, compared to the 4-C model, both of which were significantly different from the 4-C model. On the other hand, UWW and ADP had slopes of 1.09 and 1.03, respectively, compared to the 4-C model, neither of which was significantly different from the 4-C model. Based on these data, the authors concluded that UWW and ADP were more accurate in measuring fat mass compared to a 4-C model than DXA and TBW. The researchers also examined the potential bias between the various techniques by constructing residual plots. Significant correlations were found between the 4-C model versus DXA ($r = 0.47, P < 0.018$), the 4-C model versus UWW ($r = -0.53, P < 0.006$) and 4-C model versus TBW ($r = 0.61, P < 0.001$). The positive significant correlation found with DXA and TBW indicates these methods underestimated fat mass in leaner subjects and overestimated it in heavier subjects while the negative significant correlation reported for UWW suggests an overestimation in leaner subjects and an underestimation in heavier subjects. ADP was the only technique that showed a non-significant correlation signifying that there was no bias in the technique across the range of fatnesses ($r = -0.34, P = 0.100$).

Dual-energy x-ray absorptiometry. The main use of DXA is to measure bone mineral content (BMC) either at specific sites of the body, such as the lumbar spine and

femur, or to measure BMC of the whole body (Ellis, 2000). DXA has become the gold standard for the clinical assessment of BMC and is used in diagnosing bone diseases such as osteoporosis and osteopenia (Ellis, 2000). The two primary companies that manufacture DXA equipment are Hologic® and Lunar® (Sopher et al., 2004). Different manufacturers and software upgrades within companies apply slightly different formulas for calculating the various body compartments (Sopher et al.). However, the basic principle that applies for all DXA machines is that BMC is measured by differentiating it from other body tissues. This is accomplished by calculating the difference in the intensity of an x-ray when it enters an object and passes through (Ellis, 2000; Testolin et al., 2000). The reduction in the intensity of the x-ray varies among bone, lean tissue, and fat; therefore, by knowing the density of these materials, bone mineral density (BMD), fat, and lean mass can be calculated (Ellis, 2000; Testolin et al.). DXA has become widely used as a method for assessing body fat because it allows for estimates of lean and fat tissue in addition to the intended measure of BMC. DXA reports values for bone mineral content, lean tissue mass, and fat mass and, therefore, appears to be a 3-C model. However, Testolin et al. (2000) classified it as a 2-C method, and Ellis (2000) explained this by pointing out that the three measures (bone mineral content, lean tissue, and fat mass) are not determined independently. Ellis (2000) explained that DXA uses equations to calculate the mass of soft tissue and the mass of bone. Then, an assumption is made about the lean to fat ratio of the soft tissue in order to establish lean versus fat mass. It is assumed that the fat to lean ratio of soft tissue over bone is the same as the rest of the body's soft tissue. Approximately 50% of the soft tissue in a whole body scan is not

overlying bone. Thus, the fat to lean ratio of the soft tissue mass for a whole body scan is based on a sample of about half of the body.

Several problems have been noted by researchers when applying DXA to body composition analysis. DXA exposes individuals to radiation. A recent article in the *Journal of Clinical Densitometry* (Thomas, Kalkwarf, Buckley, & Heubi, 2005) stated that the effective dose from a total body DXA scan using a Hologic QDR 4500A densitometer was 3.0 μ Sv in 5 year old females and 2.9 μ Sv in 5 year old males. The National Council of Radiation Protection and Measurements (NCRP) recommendations regarding the maximum annual effective dose limit for infrequent exposures to the general public, including children, is 5000 μ Sv per year (Thomas et al.). The NCRP also stated that an annual effective dose of 10 μ Sv is a negligible individual dose (Thomas et al.). The authors stated that performing 50 DXA scans on a child in a year would still be considerably lower than the annual effective dose limit for infrequent exposure. By comparison, the radiation exposure from a whole body DXA scan is considerably less than one day at sea level or a Trans-Atlantic flight (Baylor, 2009). Radiation exposure remains a concern in studies with children despite the fact that the amount of exposure has been reported as minimal.

DXA has been touted as a quick method for assessing body composition because a whole body scan is completed in approximately two to three minutes (Ellis, 2000). However, it requires that the individual remain completely still throughout the scan which can be a challenge for children.

Other potential problems with DXA that may affect accuracy have been cited before. The assumption made about the fat to lean ratio of the soft tissue mass described above is one area of concern (Ellis, 2000; Testolin et al., 2000). Another assumption that DXA applies when calculating mass of various body compartments is that the hydration of the FFM is approximately 73% (Fields & Goran, 2000; Roemmich et al., 1997; Sopher et al., 2004). This is not a correct estimate in children, and some researchers have linked inaccuracies in DXA to this assumption (Baumgartner et al., 1991; Fields & Goran; Hewitt et al., 1993; Roemmich et al.; Sopher et al.; Wang, et al., 2003).

Ellis (2000) stated that the accuracy of DXA for determining fat mass was 5-10% with a precision of 3-4% in adults. Similarly, Roemmich et al. (1997) found that DXA overestimated body fat in children and adolescents by 4.4%. They examined the agreement of DXA with a 4-C model and found only a moderate correlation ($r^2 = 0.713$). The researchers attributed this to the hydration constant of 73.2% used in the software instead of a more child-appropriate value. They recommended that DXA be avoided in validation studies with children until adjustments are made in the software to account for the differences in the hydration of the FFM in this age group.

Fields and Goran (2000) also reported that DXA inaccurately measured fat mass in a group of 9 to 14 year old children ($n = 25$). Regression analysis was used to determine the accuracy of DXA compared to a 4-C method and a significant deviation was observed from the line of identity (slope= 0.84 ± 0.04 , $P < 0.05$). Further, DXA showed bias by under-predicting fat mass in leaner subjects and over-predicting it in heavier subjects. Next, Fields and Goran applied a correction factor presented by

Pintuaro et al. (1996) and found no significant departure from the line of identity nor any bias using DXA versus a 4-C model. It should be noted that these correction factors are only applicable to Lunar DPX-L® DXA machines that were used by Pinturo et al. and Fields and Goran. Therefore, these equations could not be generalized to a Hologic® machine or even a Lunar® machine with different software than was used by those researchers.

DXA was also found to under-estimate percent body fat in subjects with lower body fat levels and over-estimate it in those with higher body fat levels in a large pediatric cohort conducted by Sopher et al. (2004). They compared percent body fat measures using DXA and a 4-C model in 411 children and adolescents ranging in age from 6 to 18 years. Linear regression analyses revealed a fairly strong relationship between the two methods ($r^2 = 0.85$). However, the regression line deviated significantly from the line of identity ($p < 0.0001$). A paired two-tailed t test showed a statistically significant difference between the percent body fat means of DXA versus a 4-C model ($M = 22.73$ v. 21.72 , respectively, $p < 0.0001$). Nonetheless, the authors concluded that DXA could still be applicable in a pediatric clinical setting as long as practitioners were aware of its variations from the criterion method and realized that different DXA systems (Hologics® v. Lunar®) yielded different results.

Schoeller et al. (2005) released data showing that the fan-beam QDR 4500A DXA underestimated fat mass in a cohort of 1,195 adult subjects. The investigators took data from seven previous studies that used the fan-beam QDR 4500A DXA compared to a criterion method. The seven studies used various criterion methods including TBW by

dilution at four sites, densitometry at one, and a 4-C model at the other two sites. Their findings were that DXA estimates of FFM were significantly different from the criterion methods at six of the seven sites with the DXA estimates of FFM being 1.8-4.7 kg larger than estimates from the criterion method ($P<0.05$). The FFM and fat mass data were not available at the seventh site, but they reported a similar trend that DXA estimates of percent body fat were 2% lower in women and 4% lower in men compared to the criterion method. FFM from DXA was regressed on FFM from the criterion methods and the average of the individual slopes was found to be 0.946 indicating that DXA overestimated FFM by 5.4% compared to the criterion methods. The researchers also measured between-laboratory error by circulating a phantom to each center. They found small significant differences in the mass, FM, and bone mineral content values between centers; however, the largest difference for FFM was only 1.4% of the phantom mass which was smaller than the differences observed between the DXA and criterion methods. They concluded that when using a fan-beam QDR 4500A DXA, adjustments should be made for FFM, fat mass and percentage body fat to improve accuracy of results.

The manufacturer of the Hologic® QDR 4500A DXA updated the machine's pediatric software in 2006 using a new algorithm called the Automatic Low Density Whole Body Analysis (T. Kelly, personal communication, June 14, 2006). This new software lowers the bone threshold in smaller subjects (≤ 40 kg) in order to improve bone detection (T. Kelly, personal communication, June 14, 2006). It remains to be seen whether this adjustment will improve DXA's body fat estimates in children.

Summary

The number of children in the U.S. who are overweight and at risk for becoming overweight continues to rise. Overweight children face numerous, negative consequences that impact both their physical and psychological well-being. Many of these conditions are chronic and irreversible. Thus, it is imperative that causes of overweight in childhood are identified so that prevention and treatment strategies can be developed. The literature indicates that environmental factors including diet and physical activity are the main areas of focus.

Several dietary factors were cited in the literature as being positively linked to weight and/or adiposity. These included total fat intake, juice consumption, total Kilocalorie intake, and poor diet quality (AAP, 2001; Aeberli et al., 2007; Manios et al., 2008; Skinner et al., 2003). Other dietary factors reported to have an inverse relationship to weight and/or adiposity included calcium intake, breast feeding during infancy, and polyunsaturated fatty acid intake (Barba et al., 2005; Carruth & Skinner, 2001; Ong et al., 2006; Skinner et al.; Toschke et al., 2007). Physical activity levels were also been explored in the literature. The amount of time children spend doing sedentary activities such as watching television and playing video games were suggested to lead weight gain (Coon et al., 2001; Lumeng et al., 2006; Vandewater et al., 2004). In addition, the home environment was shown to impact a child's physical activity patterns (Spurrier et al., 2008). Researchers also showed that children who have higher physical activity levels are less likely to have health problems such as high lipid levels and hypertension (Pan & Pratt, 2008).

The importance of early identification of abnormal increases in weight and adiposity in children was emphasized in the literature (AAP, 2003). Several researchers cited infancy and early childhood as critical periods for disease development, including obesity (Baird et al., 2005; Kinra et al., 2005; Stettler et al., 2003).

The accuracy of several body composition methods were investigated in children. ADP has only been tested in children ≥ 5 years of age (Demerath et al., 2002; Fields & Goran, 2000; Fields et al., 2004). In addition, the literature indicated that DXA, which is used as a gold standard for measuring body composition by many researchers, may be less accurate in children (Baumgartner et al., 1991; Fields & Goran; Hewitt et al., 1993; Roemmich et al., 1997; Sopher et al., 2004; Wang et al., 2003). Additional research is needed in order to determine which methods are the most accurate and practical for assessing body composition in children.

CHAPTER III

METHODOLOGY

Methods used in this study were approved by the Institutional Review Boards at Texas Woman's University, the University of Arkansas for Medical Sciences (UAMS), and the University of Central Arkansas (see Appendix A). All procedures were carried out at the Arkansas Children's Nutrition Center (ACNC), one of six Human Nutrition Research Centers funded through the Agricultural Research Service of the United States Department of Agriculture. The ACNC is affiliated with Arkansas Children's Hospital, Arkansas Children's Hospital Research Institute and UAMS.

Recruitment

Eighty-two participants were recruited for the study. Eighteen of the participants were recruited from an ongoing study at the ACNC, and the other 64 participants were recruited by distributing information in the form of flyers at local daycare centers, universities and health fairs (see Appendix B). Parents/guardians interested in having their children participate in the study called the ACNC Recruitment Line or sent an e-mail expressing their interest. The researcher conducted a phone screen (see Appendix C) to qualify the child for the study. Inclusion criteria for participation in the study were as follows: 1) child was 3 to 5 years of age, 2) child was full-term at birth (37-<42 weeks gestation), 3) child weighed at least 5.5 pounds at birth, 4) child was fed exclusively either formula or breast milk during infancy for at least the first six months, 5) English

was the primary language of the parent/guardian, and 6) the parent/guardian was ≥ 18 years of age. Children were excluded from the study if they had any of the following conditions: 1) disorders affecting growth such as failure to thrive, hypothyroidism or hyperthyroidism; 2) chronic diseases such as diabetes or cystic fibrosis; 3) disorders of carbohydrate metabolism such as hyperglycemia or hypoglycemia; 4) moderate to severe asthma or other pulmonary disorders requiring regular medication; 5) bone disease; 6) attention-deficit-hyperactivity disorder; or 7) attention-deficit disorders. In addition, children taking medications that could impact bone density and dual energy x-ray absorptiometry (DXA) results, such as corticosteroids, were excluded from the study.

An appointment was scheduled at the ACNC for those children who qualified. Parents/guardians were given verbal instructions at that time for completing the 3-day food records. In addition, parents/guardians were mailed written instructions for keeping the food records along with confirmation of the appointment and directions to the ACNC.

Consent Process

Parents/guardians were given detailed verbal and written explanations of all study procedures prior to data collection. Parents/guardians provided written consent for their child to participate in the study after they had been given time to read the consent form and ask questions (see Appendix D). The confidentiality procedures followed by the ACNC were also described in oral and written form and parents/guardians signed a Health Insurance Portability and Accountability Act (HIPAA) form to verify understanding and agreement with the policy (see Appendix E).

Data Collection

Demographic Data

Contact information was obtained from the parents/guardians including phone numbers and address. Information collected about the child included date of birth, gender, ethnicity, gestational age and birth weight. Additionally, the parent/guardian was queried about complications during pregnancy, the child's nutrition history during infancy, medical history and medication usage, including dietary supplements (see Appendix F).

Dietary Data

Infant feeding. The researcher questioned the parent/guardian to determine the child's primary source of nutrition during the first six months of life. Children were classified as breast fed if they had breast milk without supplemental formula from birth until at least 6 months of age. Children were classified as formula fed if they were on a milk-based infant formula from 8 weeks to at least 6 months of life.

Three-day food records. Three-day food records were chosen as the method for assessing dietary intake. Lanigan et al. (2004) reported that 2 to 5 days of food records were optimal when assessing nutrient intake in young children. This was considered along with the burden of keeping several days of food records and it was determined that three-day food records would produce the most accurate results for the study.

Three-day food records were collected from the parents/guardians of the participants and analyzed using the Nutrition Data System for Research (NDSR) software version 2007, developed by the Nutrition Coordinating Center, University of Minnesota,

Minneapolis, MN. The data were used to measure the associations between intake of total fat, polyunsaturated fat, fruit juice, calcium, Kilocalories, Healthy Eating Index (HEI) scores and percentage of body fat.

The researcher provided the parents/guardians with detailed instructions about keeping food records over the phone when the study visit was scheduled and in writing along with a confirmation letter mailed prior to the study visit. Parents/guardians were instructed to record all foods and beverages consumed for three consecutive days, including one weekend day. Food record forms were provided for the parents/guardians to record the intake with the mailed instructions (see Appendix G). In addition, they were asked to record intake as it occurred to decrease the chance of omitting items and to more accurately approximate portion sizes. The Food Portion Visual™ from Nutrition Consulting Enterprises, Framingham, MA, was sent with the mailed instructions to aid in recording portion sizes (see Appendix H). The parents/guardians returned the food records on the day of the study visit, and the researcher reviewed each item recorded to ensure that correct descriptions and quantities were recorded.

The researcher entered the food records into the NDSR program. The Foods Report and Record Properties Report were printed and attached to the hand-written records. Another trained individual checked the reports against the food records to ensure accuracy. Any errors identified were returned to the researcher and corrections were made to the NDSR database prior to data analysis.

Diet quality. Diet quality is a way of determining the nutritional merit of the diet as a whole instead of focusing on individual nutrients. Some researchers attempt to

assess diet quality by focusing on certain foods or groups of key nutrients. However, the ideal method for assessing diet quality is to use a standardized tool that has been validated. Thus, diet quality was measured in the present study using the HEI, 2005 developed by the United States Department of Agriculture's Center for Nutrition Policy and Promotion. This tool was developed for Americans over the age of two to measure how closely the diet matches the 2005 recommendations of the Dietary Guidelines for Americans and the Food Guide Pyramid. Guenther et al. (2007) measured the content validity, construct validity, and reliability of the HEI and concluded it was a valid and reliable instrument. Refer to Chapter 2 for details on validity and reliability testing.

The HEI provides one score for measuring overall diet quality by taking into account the adequacy components and the moderation components outlined in the Dietary Guidelines for Americans. The adequacy components are assessed by determining how much an individual is consuming from the food groups in the Food Guide Pyramid and from three additional groups, including; 1) whole fruits, 2) dark green and orange vegetables and legumes, and 3) whole grains. Consumption of oils coming from vegetable sources, fish, nuts, and seeds are included in the adequacy component, as well. Optimal scores of 5 are given for all adequacy components except milk, meat, and beans and the oil components and these are given optimal scores of 10.

The moderation component is made up of three parts and addresses the recommendations by the Dietary Guidelines for Americans to consume less saturated fat, sodium and foods or beverages containing solid fats, alcohol and added sugars

(SoFAAS). The saturated fat and sodium components are given maximum scores of 10 while the maximum score for the SoFAAS component is 20.

The HEI is a nutrient density approach for assessing the diet which is reflected in the way the scores are calculated. Scores for the adequacy components are determined based on how much of a particular food group or oil is consumed per 1,000 Kilocalories. The maximum scores of 5 or 10 are given when an individual meets or exceeds the standard for that food group and a score of zero is given if no foods from that component are consumed. Intakes between the standard score and zero are calculated by dividing the amount of food from that group consumed per 1,000 Kilocalories by the standard amount and then multiplying that by the total points possible for that component. For example, the standard for the total fruit group is ≥ 0.8 cup equivalents per 1,000 Kilocalories and the maximum score is 5. An individual who consumed only 0.6 cup equivalents of total fruit per 1,000 Kilocalories would receive a score of 3.75 for that component ($0.6/0.8 \times 5 = 3.75$). See Appendix I for an outline of the standards for each component and the maximum scores.

The moderation components are also assigned maximum scores when the standards are met. In addition, these components have established levels where no points are given when consumption exceeds a specified point. Intake levels falling between the established scores get prorated linearly. In other words, an equal number of points get deducted from the maximum score for each unit below the standard score. The standard for sodium is ≤ 700 mg per 1,000 Kilocalories, and diets meeting this standard are assigned the maximum score of 10. Sodium intakes of 1,100 mg per 1,000 Kilocalories

receive a score of 8 and intakes of $>2,000$ mg per 1,000 Kilocalories receive a score of 0. The standard for the SoFAAS component is $\leq 20\%$ of Kilocalories and diets meeting the standard receive the maximum score of 20. Diets with $\geq 50\%$ of Kilocalories from SoFAAS receive no points. The standard for saturated fat is $\leq 7\%$ of total Kilocalories and the maximum score of 10 is assigned for diets meeting the standard. Intakes of saturated fat at 10% of total Kilocalories receive a score of 8 and intakes $\geq 15\%$ receive a score of 0.

The averaged 3-day food records obtained from the participants were used to calculate the HEI. The food group components were determined by using an NDSR output file that provided the number of servings of foods from various food groups. NDSR provides 166 subgroups of foods and the serving sizes for most foods are based on the 2005 Dietary Guidelines for Americans. The subgroups related to each of the food components in the HEI were identified and summed to determine the total number of servings provided by each group. Then, the number of servings per 1,000 Kilocalories was determined by dividing 1,000 by total Kilocalories consumed and multiplying that by the total number of servings. For example, if the 3-day average for total fruit intake was 2 servings and the child consumed an average of 1,400 Kilocalories, then the number of servings per 1,000 Kilocalories would be 1.43 ($1,000/1,400 \times 2 = 1.43$). The oil component was determined by adding the average intakes of monounsaturated and polyunsaturated fatty acids in grams and converting that to the number of grams per 1,000 Kilocalories as described above. Similarly, the sodium component was calculated by using the average sodium intake and converting that to the number of milligrams per

1,000 Kilocalories. The saturated fat component was determined by looking at the 3-day average of the percentage of Kilocalories coming from saturated and trans-fatty acids. The SoFAAS component was calculated by summing the percentage of Kilocalories coming from saturated fat, trans-fatty acids and added sugars. The total score was calculated by summing the individual component scores. The maximum possible score was 100.

Physical Activity Data

The Netherlands Physical Activity Questionnaire (NPAQ) was used to assess the physical activity levels and amount of screen time of the participants. The NPAQ assesses children's daily activity preferences and typical behaviors that are thought to be associated with physical activity levels in a short questionnaire format. It is designed to be completed by the caregiver.

The NPAQ was chosen for several reasons. First, it was developed for young children less than 10 years of age and has been tested for reliability and validity by Janz et al. (2005). Refer to Chapter 2 for detailed information on reliability and validity testing. Second, it does not require the respondent to quantify duration, frequency and intensity of the child's physical activities. These components of activity are hard for untrained individuals to accurately estimate as well as time-consuming. Third, the burden placed on the parent/guardian and participant was low with the NPAQ. The parent/guardian was able to complete the questionnaire after receiving instructions from the researcher in a few minutes during the study visit. The researcher was concerned that noncompliance might be an issue if young children were asked to wear activity monitors

or if parents/guardians had to spend a lot of time observing and recording activities prior to the study visit.

The questionnaire is divided into two parts. The first part consists of seven statements that describe various activities. The respondent is asked to rate on a scale of one to five the child's preference for those activities. These seven questions make up the composite score for the NPAQ and are a measure of the physical activity level of the child. Scores range from 20 to 100 with the higher scores indicating higher physical activity levels. The second part of the questionnaire asks about the amount of time the child spends watching TV, playing video games, sleeping and the types of physical activities they engage in. See Appendix J for a sample of the NPAQ and scoring system.

Anthropometric Data

Weight and stature measures were obtained for each participant by the researcher. Weight measurements were taken using a Tanita Electronic Scale, model BWB-800. The scales were calibrated weekly using test weights ranging in mass from 2 to 92 kilograms to ensure precision of 0.1 kilogram or better. Children were weighed without shoes, wearing only undergarments and a hospital gown. The participant stood in the middle of the scale's platform and the weight was recorded to the nearest 0.1 kilogram. The child was re-positioned on the scale for a second measure that was recorded to the nearest 0.1 kilogram. The two weights had to agree within a tolerance level of 0.1 kilogram or repeated measures were taken until two weights within this tolerance level were obtained.

Stature measures were taken using a vertical, wall-mounted stadiometer from Perspective Enterprises, model PE-WM-60-76. The child's shoes and any hair ornaments

that interfered with obtaining accurate measures were removed. The child stood against the stadiometer with heels together, legs straight, arms at sides and shoulders relaxed. The child was instructed to look straight ahead and take a deep breath while standing fully erect without altering the position of the heels. The headpiece of the stadiometer was lowered to the crown of the child's head with sufficient pressure to compress the hair. The researcher ensured the child's head was in the Frankfort plane, the anatomical plane that extends from the outer corner of the eye to the opening of the ear. The height measure was recorded to the nearest 0.1 cm. The child was repositioned and a second measure was taken using the same techniques. The two measures had to agree within a tolerance level of 0.5 cm or additional measures were taken until two measures were attained within the tolerance level.

Body Composition Data

The three techniques used to measure body composition of the participants were total body water (TBW) analysis using deuterium oxide (^2H), DXA using Hologic's® QDR 4500 Series, and air displacement plethysmography (ADP) using the Bod Pod®. See Chapter 2 for a review of each method. Total body water was chosen because it is a non-invasive technique cited by several researchers as an acceptable criterion method (Reilly, 1998; Schoeller et al., 2005; Yao, Roberts, Ma, Pan, & McCrory, 2002). Dual-energy x-ray absorptiometry is already being used by many researchers for assessing body fat in children, but its accuracy has been questioned (Fields & Goran, 2000; Roemmich et al., 1997; Sopher et al., 2004). Air-displacement plethysmography has been proposed as a less-invasive, practical approach for measuring body fat in children, but it has not been

tested in children younger than age six (Fields & Goran, 2000; Nunez et al., 1999). Thus, DXA and ADP were chosen because they are emerging techniques in pediatric body composition assessment and their accuracy must be determined for this population.

TBW technique. A 250 milliliter bottle of 99.8% deuterium oxide (^2H) was ordered from Cambridge Isotope Laboratories, Andover, MA. A 10% dilution of ^2H with filtered water was prepared by pouring the 250 ml bottle of ^2H into a 3-liter graduated cylinder. A total of 2,250 ml of filtered water was measured into another container and part of this was used to rinse the ^2H container several times. The rinse water was added to the 3-liter graduated cylinder containing the ^2H . The remainder of the filtered water was added to the ^2H mixture gradually and mixed well. The dilution was divided among smaller containers of 250 to 500 milliliters for easier handling. These containers were dark-colored to minimize light exposure. The containers were labeled with the solution name, dilution and date of preparation and stored in the refrigerator. Ten milliliter aliquots of the dilution and filtered water were prepared for future shipment to Baylor College of Medicine's Gas-Isotope-Ratio Mass Spectrometry Laboratory for analysis.

A baseline urine sample of 3 milliliters was obtained from the child prior to ingestion of the ^2H dose. The baseline urine sample could not be the first void of the day. Urine samples were obtained in a collection device placed over the toilet for all participants, except two who were not toilet trained. Urine was collected for those two individuals by placing the Pediatric Urine Collector from Precision Dynamics Corporation, San Fernando, CA, over the genital area. The time of the urine collection was recorded. The urine sample was divided into two separate vials using a transfer

pipette. One of the vials was shipped to Baylor College of Medicine's Gas-Isotope-Ratio Mass Spectrometry Laboratory for analysis, and the other was retained at the ACNC. Each vial contained 1.5 milliliters of urine. The urine samples were handled carefully to prevent contamination and to ensure that all of the transfer equipment was free of moisture. The samples were stored promptly in a -20 degree Celsius freezer.

Once the baseline urine sample was provided, the children were given a dose of the diluted ^2H in the amount of 1000 milligrams per kilogram of body weight. The ^2H dilution was weighed on an electronic gram scale to the nearest 0.01 gram. The ^2H was administered by mouth in a beverage of the child's choice. The ^2H was mixed into two fluid ounces of the beverage and placed in a cup with a lid to avoid spills. The researcher observed the child consume the beverage, and once it was empty, the container was rinsed with more of the drink, and the child drank the residual. This was repeated twice to ensure all the ^2H was consumed. The researcher noted the time the child finished consuming the beverage.

A second urine sample was collected no sooner than 4 hours and 45 minutes after the beverage containing the ^2H was ingested to ensure adequate isotope enrichment. The time of the collection was recorded. The same procedures used for collection and storage of the baseline urine sample were followed with the post-dose sample.

The samples were sent for analysis in three different shipments to Dr. William Wong at Baylor College of Medicine's Gas-Isotope-Ratio Mass Spectrometry Laboratory in Houston, Texas. Samples were shipped by express courier for overnight delivery with sufficient dry ice to keep them frozen.

The analyzed results provided an estimate of the ^2H space or TBW of the participants. However, one must take into account that approximately 4% of the hydrogen tracers exchange with hydrogen molecules outside the aqueous space (Ellis, 2000; Wang et al., 1999). Therefore, a mathematical adjustment was made (TBW / 1.04) so that TBW was not overestimated. The adjusted TBW was then divided by the age and gender-specific hydration of fat-free mass (FFM) constants proposed by Fomon et al. (1982). These constants were used to improve accuracy for determining FFM. Fat weight was then calculated by subtracting FFM from total body weight. This allowed the researcher to determine percent body fat by dividing fat weight by total body weight and multiplying by 100.

DXA technique. Approval from the Arkansas Department of Health and Human Services and the UAMS Radiation Committee was granted to perform the DXA scans (see Appendixes K and L). Quality control (QC) procedures were performed on the DXA machine daily prior to use. This involved conducting a scan of a spine phantom to ensure the accuracy of the machine. No problems were detected on any of the days DXA scans were obtained on the children in the study.

The children removed all clothing except undergarments and were given hospital gowns to wear for the procedure. All metal objects such as earrings were removed prior to the scan. All scans were performed in the afternoon approximately 1.5 to 2 hours after the child had eaten lunch. No foods or beverages were consumed after lunch prior to the scan. The researcher explained to the child that the table would move and that he/she had to lie completely still throughout the two minute scan. The researcher entered personal

information about the child including age, weight and height into the software and conducted the scan. The parent/guardian and researcher remained with the child throughout the entire procedure. Repeat scans for children who moved were attempted no more than three times. The scans were analyzed using Hologics® Pediatric Whole Body Software, Version 12.3.

ADP technique. Quality control (QC) procedures were performed each day that the ADP machine was used for testing. This involved a 4-step process outlined by the manufacturer, Life Measurement, Incorporated (LMi), Concord, CA. The machine had to be turned on and undergo a warm-up period of at least 30 minutes before beginning the four steps. Part of the modification process for testing young children was to alter the inside of the chamber by inserting a booster seat, so the child was properly placed, and a DVD player in order to occupy the child during the procedure. Therefore, the entire QC procedure was done with the booster seat and DVD player in the chamber. The first step of the QC procedure involved testing the system's hardware once the warm-up phase was complete. The second step was an "auto run" process where the machine took six consecutive volume readings using a 19.993 liter test cylinder. The standard size cylinder used for testing adults is approximately 50 liters but this smaller cylinder was provided by LMi to the ACNC for testing young children. The measures had to average between 19.893 and 20.093 liters with a standard deviation of ≤ 75 ml and this step had to be done within those tolerable limits twice in order to pass this step. The third step tested the system by taking five more volume measures, but in this step the door was opened between each volume measure as it would be when testing a person. The average

volumes and standard deviation for the five measures had to be within the same parameters as in step two. The final step was calibrating the scales which are part of the ADP machine. Two 10 kilogram test weights were placed on the scales twice, and the measure had to be within a 0.01 kilogram tolerance level.

All study participants were tested in the afternoon at least one hour after finishing lunch. No foods or beverages were consumed for the hour prior to testing. The children were asked to void before beginning the procedure because a full bladder can affect results. Each child changed into a tight-fitting Lycra® swimsuit and swim cap provided by the ACNC. The researcher assisted the parent in getting all of the child's hair inside the swim cap. The researcher offered to allow the child a test run in the parent's lap prior to the actual testing to prevent apprehension or fear. The researcher entered the child's name, date of birth, gender and height into the software attached to the ADP machine and then a two-step calibration procedure was conducted. The first part was a volume measure which is typically done with the chamber empty so that a baseline reading can be obtained. However, the booster seat and DVD player were in the chamber during this step since these items needed to be included to tare the machine. The second step was to add the 19.993 liter test cylinder and take a volume measure to ensure accuracy. The child was weighed on the scales attached to the ADP machine at the computer's prompt, and then the calibration cylinder was removed and the child entered the chamber. The child was instructed to remain still and not talk during the 50-second volume measure. At least two volume measures were taken on each child. The door of the chamber was opened between each measure. The two measures had to agree within 0.2% of each other

or the machine would require a third volume measure. The two-point calibration process had to be repeated if two measures were not within the tolerance level of 0.2% after three measures were taken. Thoracic gas volume was predicted instead of measured.

The researcher used the average of the two raw volume measures to determine body density, which was then used in a prediction equation for percent body fat. However, the volume measures had to be adjusted because the values given by the machine were based on adult equations for thoracic gas volume (TGV) and surface area artifact (SAA). In addition, the percent body fat equation used by the manufacturer is Siri's equation that is intended for adults and assumes that density of FFM is 1.1 g/cm². This is not an accurate assumption in children. Thus, several modifications were made for determining body volume and percent body fat to account for the smaller size of the child participants.

The following formula was used to determine an adjusted body volume:

$$\text{Adjusted volume} = \text{raw volume} + 0.4 \times \text{TGV} - \text{SAA} \quad (1)$$

This formula is already used by the manufacturer. However, the values for TGV and SAA were manipulated to arrive at a more accurate value for young children.

TGV prediction equations have been published for children as young as 6 years old (Fields et al., 2004). However, no standardized equations exist for children under age six. Therefore, TGV was predicted based on the following equation cited by Bosy-Westphal et al. (2005), Davis et al. (2007), Higgins et al. (2006) and Wells et al. (2003) where FRC is functional residual capacity and TV is tidal volume:

$$\text{TGV} = (\text{FRC} + 0.5 \text{ TV})/1000 \quad (2)$$

The value for FRC was calculated using the following equations from Polgar and Promadhat (cited by M. Swanstrom, personal communication, April 24, 2008) where H stands for height in centimeters:

$$\text{FRC Males} = 0.00075 \times H^{2.92} \quad (3)$$

$$\text{FRC Females} = 0.00178 \times H^{2.74} \quad (4)$$

The TV value was calculated using the following formula published by Stocks, Sly, Tepper and Morgan (cited by M. Swanstrom, personal communication, April 24, 2008) where age is in years and mass is in kilograms:

$$\text{TV} = (0.4 \times \text{Age} + 9.1) \times \text{Mass} \quad (5)$$

The second adjustment made to the raw body volume was to adjust for SAA of the smaller subjects. Surface area artifact is calculated by multiplying a constant value by body surface area (BSA). The constant (k) value used was -0.00047 as cited by Bosy-Westphal et al. (2005). It should be noted that the Bosy-Westphal et al. publication misprinted this value, and Radley and Fields (2006) made the correction. The Haycock, Schwartz and Wisotsky formula (cited by Bosy-Westphal et al. and Wells et al., 2003) was used for determining body surface area (BSA). The Haycock et al. (1978) formula is as follows where mass is weight in kilograms and H is height in centimeters:

$$BSA = 242.65 \times \text{Mass}^{0.5378} \times H^{0.3964} \quad (6)$$

The BSA and k values were then used to compute SAA as follows:

$$SAA = -0.0000467 \times BSA \quad (7)$$

A recent publication by Bosy-Westphal et al. reported that the Haycock et al. formula improved accuracy in percent body fat calculations in children 5-18 years old as compared to an adult formula. Wells et al. (2003) also compared the Haycock et al. formula to an adult formula and reported that the body volume differences in children ages 5 to 7 years of age were only minimal and did not impact the final percent body fat results.

Body density was calculated once the adjusted body volume was determined using the following formula:

$$\text{Body density} = \text{Mass} / \text{Adjusted Volume} \quad (8)$$

Body density was used along with calculated constant values in the following equation to determine percent body fat where C1 and C2 are constant values 1 and 2 and Db is body density:

$$\text{Percent body fat} = (C1 / Db - C2) \times 100 \quad (9)$$

The constant values were calculated based on formulas cited by Gately et al. (2003) and Wang et al. (2003) using the age and gender-specific density of FFM values published by Fomon et al. (1982) (see Tables 1 and 2).

Table 1

Age and Gender-Specific Values for Density of Fat-Free Mass

Age	Boys Dffm (g/ml)	Girls Dffm (g/ml)
3	1.074	1.071
4	1.076	1.072
5	1.078	1.073

Note: Dffm = density of fat-free mass. Adapted from “Body Composition of Reference Children from Birth to Age 10 Years,” by S.J. Fomon, F. Haschke, E.E. Ziegler, and S.E. Nelson, 1982, *The American Journal of Clinical Nutrition*, 35, p. 1173. Copyright 1982 by the American Society for Clinical Nutrition. Adapted with permission of the author.

Table 2

Age and Gender-Specific Constant Values for Calculating Percent Body Fat

Age	<u>Boys</u>		<u>Girls</u>	
	Constant 1	Constant 2	Constant 1	Constant 2
3	5.582	5.197	5.664	5.289
4	5.529	5.138	5.637	5.258
5	5.476	5.080	5.609	5.228

Note: Constant 1 = Constant 2 X Dffm; Constant 2 = Dfm / (Dffm – Dfm). Dffm = density of fat-free mass. Values for Dffm given in Table 2. Dfm = density of fat mass. Values for Dfm assumed to be 0.9007 for all ages and genders.

Statistical Analyses

All statistical analyses were conducted using SPSS, version 15.0, statistical database software. Descriptive statistics, including frequencies, were used to describe the study participants' characteristics including gender, age, feeding group and anthropometrics. Chi square and Cramer's V statistics were calculated to describe the relationships between gender, age and feeding group. Means, standard deviations and ranges were used to describe the dietary intake, HEI scores, NPAQ scores and percent body fat of the participants.

Deuterium oxide results, information regarding nutrition during infancy, food records, and physical activity data were collected on 78 children. Ten of the children were unable to complete the DXA procedure and four were unable to complete the ADP procedure because they were afraid or could not lie still for the DXA scan. All three body composition methods were conducted with 67 children.

In addition, ADP values from two five-year-old children, one male and one female, were removed from the data set because they produced negative numbers. In both cases, the children had the highest calculated density values for gender. These negative numbers likely resulted from inaccurate volume measurements obtained by the ADP machine that impacted the density calculation. Possible reasons for inaccuracies in volume measures include movement by the child, delayed time with door open between volume measurements, or temperature and humidity changes in the testing room. This left complete data on 65 children.

The missing data were random. Therefore, analyses were conducted with the 65 children providing complete data, and this was compared to data analyzed using a series means replacement with all 78 children. The results did not differ when the series means replacement was applied. Thus, the final analyses were reported based on the series means replacement with a sample size of 78.

Factors that were considered when selecting the appropriate statistical techniques to test the hypotheses included if the data were from a random sample, interval level, and normally distributed. The data were from a random sample and interval level. The Kolmogorov-Smirnov Normality Test was run to determine if the variables were normally distributed. All of the variables met the assumption of normality as indicated by p values of ≥ 0.064 . Therefore, parametric tests were used for statistical analyses.

Pearson's Product Moment Correlation was used to test the relationships between percent body fat and the following independent variables: 1) mean daily intake of total fat grams, 2) mean daily intake of polyunsaturated fatty acid (PUFA) grams, 3) fruit juice in fluid ounces, 4) calcium in milligrams, 5) total daily energy in Kilocalories, 6) HEI scores, and 7) NPAQ scores. A power analysis indicated that a sample size of 65 was necessary to achieve a power level of 0.80 (for a moderate effect size, $r = 0.4$) with $\alpha = 0.01$, assuming a minimum significant correlation coefficient of 0.60. The strength of the relationship between variables was assessed using Cohen's (1992) classifications that r values of 0.1, 0.3 and 0.5 represent small, medium and large effects, respectively.

A repeated measures Multivariate Analysis of Variance (MANOVA) was performed to test hypotheses two, three, four, and five. The procedure investigated

differences in the three methods for measuring body fat (^2H , DXA and ADP) as well as gender and feeding group differences in percent of body fat of the participants. A preliminary power analysis showed that a sample size of 64 was required to test these hypotheses with a power level of 0.80 (moderate $F = .25$) and alpha = 0.05. The within-subjects effects were tested for the following variables: 1) body fat measures; 2) body fat measures and gender; 2) body fat measures and feeding group; and 3) body fat measures, feeding group, and gender. No effect was interpreted as a p value >0.05 . The tests of between-subjects effects were used to determine if statistically significant differences existed in mean percent body fat based on gender and feeding group. The effect size of the within-subjects effects and between-subjects effects was examined using the partial eta squared statistic. Partial eta squared values give an estimate of the size of the differences observed and were interpreted based on Cohen's established values of .01, .06 and .14 for small, moderate and large effects, respectively (Pallant, 2007). Post-hoc comparisons using the Bonferroni test provided pair-wise comparisons for determining which of the body fat measures differed significantly from each other.

CHAPTER IV

RESULTS

Characteristics of Study Participants

This was a prospective, cross-sectional study of 82 healthy children ages 3 to 5 years living in the Central Arkansas area. Similar numbers of children were recruited from each gender, age, and feeding group. Data from three children were eliminated because the laboratory determined the deuterium oxide results were erroneous, and data from one child was eliminated because of the child's refusal to consume the drink containing deuterium. This left a final sample of 78 children to test the hypotheses. Study participants were comprised of children living in the Central Arkansas area.

Thirty-six of the children were male and 42 were female. Thirty-one children were 3 years old, 23 were 4 years old, and 24 were 5 years old. More 3-year-olds were included because eight of them failed to complete the dual energy x-ray absorptiometry (DXA) and air-displacement plethysmography (ADP) procedures. Sixty-four of the children were White (82%), nine were Black (12%), and five were children with other racial backgrounds including two Hispanic children (3%) and three children of mixed racial backgrounds (3%). This closely matches the ethnic distribution for children in Arkansas which is 68% White, 20% Black, 8% Hispanic, and 4% other (The Henry J. Kaiser Family Foundation, 2009). There were 42 breast-fed children and 36 formula-fed

children in the data set. See Tables 3 and 4 for a description of the study participants' characteristics by gender and age.

Table 3

Number of Child Participants according to Ethnicity by Gender and Age (N=78)

Age (years)	Males (n=36)			Females (n=42)		
	White ^a	Black ^b	Other ^c	White ^a	Black ^b	Other ^c
3 (n = 31)	12	2	0	13	2	2
4 (n = 23)	8	1	1	10	2	1
5 (n = 24)	11	1	0	10	1	1
Totals	31	4	1	33	5	4

Note: ^aWhite, Non-Hispanic; ^bBlack, Non-Hispanic; ^cOther – 2 children were Hispanic and 3 children had a mixed racial background.

Table 4

Number of Child Participants according to Infant Feeding by Gender and Age (N = 78)

Age (years)	<u>Males (n=36)</u>		<u>Females (n=42)</u>	
	Breast-fed	Formula-fed	Breast-fed	Formula-fed
3 (n = 31)	7	7	8	9
4 (n = 23)	5	5	8	5
5 (n = 24)	7	5	7	5
Totals	19	17	23	19

Relationships between gender, age, and feeding group were examined. The relationship between gender and age was not significant, $\chi^2(2) = .051, p = .975$, Cramer's $V = .026, p = .975$. There was not a significant relationship between gender and feeding group, $\chi^2(1) = .031, p = .861$, Cramer's $V = .020, p = .861$. Likewise, no significant relationship was found between age and feeding group, $\chi^2(2) = .662, p = .718$, Cramer's $V = .092, p = .718$. Similar numbers of males and females from each age and feeding group were included in the analysis.

Anthropometric and Body Composition Characteristics of Participants

Children with a wide range of body mass index (BMI) for age percentiles and body fat levels were included in the analyses since the goal was to examine dietary factors, physical activity, and the accuracy of DXA and ADP (see Table 5). The mean

weight and height values of all 78 participants were 17.8 kg (SD = 3.2) and 104.5 cm (SD = 7.8), respectively. The mean BMI value for the group was 16.2 kg/m² (SD = 1.4). One child was classified as underweight with a BMI for age less than the fifth percentile. Fifty-eight of the children (74%) were classified as normal weight with BMI for age ranging from 5.0-84.9%. Eleven of the children (14%) were at risk for becoming overweight with BMI for age between 85.0-94.9%, and eight children (10%) were overweight with a BMI for age at or above 95%. See Table 5 for the frequency of children in each BMI category by age and gender.

Table 5

Number of Child Participants per BMI Category by Gender and Age (N= 78)

Age (years)	Males (n=36)				Females (n=42)			
	UW ^a	NW ^b	ARO ^c	OW ^d	UW ^a	NW ^b	ARO ^c	OW ^d
3 (n=31)	0	10	1	3	1	12	3	1
4 (n=23)	0	7	2	1	0	10	2	1
5 (n=24)	0	9	2	1	0	10	1	1
Totals	0	26	5	5	1	32	6	3

^aUW = under-weight with BMI for age <5%; ^bNW = normal weight with BMI for age 5.0-84.9%; ^cARO = at risk for overweight with BMI for age 85.0-94.9%; ^dOW = overweight with BMI for age ≥95%.

Mean weights, heights, and BMI values were similar between boys and girls. The mean weights were 18.1 kg (*SD* = 3.1) and 17.6 kg (*SD* = 3.2) for the boys (*n* = 36) and girls (*n* = 42), respectively. The average height for the boys was 104.6 cm (*SD* = 7.6) and for the girls was 104.3 cm (*SD* = 8.0). The BMI values were 16.2 kg/m² (*SD* = 1.4) and 16.1 kg/m² (*SD* = 1.6) for the boys and girls, respectively. See Table 6 for the means, standard deviations, and ranges of the anthropometric characteristics of the participants by gender and age.

Table 6

Anthropometric Characteristics of Children by Gender and Age (N=78)

Age (years)	Males (n=36)		Females (n=42)	
	Mean ± SD	Range	Mean ± SD	Range
	Weight (kg)			
3	15.4 ± 1.7	13.2-18.6	15.1 ± 1.4	13.0-18.4
4	18.7 ± 1.6	16.4-21.6	17.7 ± 1.3	15.2-20.0
5	20.7 ± 3.0	17.5-27.5	20.7 ± 3.5	16.8-28.7
	Height (cm)			
3	97.1 ± 3.8	92.2-105.5	97.7 ± 3.3	92.6-105.5
4	106.3 ± 3.1	101.0-112.3	103.8 ± 3.3	98.5-110.4
5	111.9 ± 5.4	106.0-123.5	113.5 ± 6.3	100.5-123.9
	BMI (kg/m ²)			
3	16.3 ± 1.4	15.0-18.6	15.9 ± 1.4	13.7-18.6
4	16.5 ± 0.8	15.6-18.0	16.5 ± 1.1	14.6-18.5
5	16.4 ± 1.1	14.8-19.1	16.0 ± 2.3	13.9-22.6

Mean percent body fat levels of all participants as measured by deuterium oxide (²H), DXA and ADP were 22.71 (*SD* = 4.54) with a range of 12.94 to 37.48, 27.53 (*SD* = 5.67) with a range of 16.50 to 42.30, and 21.42 (*SD* = 7.83) with a range of 2.49 to 36.64, respectively. Mean percent body fat values were similar among three year olds (*M* = 23.26, *SD* = 4.08), four year olds (*M* = 23.50, *SD* = 3.93), and five year olds (*M* = 21.34,

$SD = 5.37$) as measured by the criterion method, deuterium dilution. Thus, age was not included as a factor in further analyses. However, there was a significant difference between the mean percent body fat levels of boys ($M = 20.39$, $SD = 3.22$) and girls ($M = 24.71$, $SD = 4.59$). Thus, analyses were conducted for male and female participants separately. See Tables 7 and 8 for the mean percent body fat levels of participants by gender, age and feeding group.

Table 7

Percent Body Fat of Males by Age and Feeding Group (n=36)

Age (years)	Breast-Fed (n=19)		Formula Fed (n=17)	
	Mean \pm SD	Range	Mean \pm SD	Range
3 (n=14)				
^a H	21.52 \pm 3.33	18.73-28.10	22.77 \pm 2.49	18.69-25.69
^b DXA	27.26 \pm 2.50	23.80-31.50	26.18 \pm 3.74	19.70-31.70
^c ADP	19.27 \pm 6.01	11.30-30.20	21.74 \pm 10.31	4.55-33.72
4 (n=10)				
^a H	19.98 \pm 1.12	18.38-21.25	20.73 \pm 3.27	18.03-26.22
^b DXA	22.76 \pm 2.92	20.60-27.80	24.50 \pm 3.55	22.00-30.70
^c ADP	18.51 \pm 4.15	12.65-22.21	18.63 \pm 5.84	12.07-24.79
5 (n=12)				
^a H	18.50 \pm 3.45	14.68-24.68	18.18 \pm 3.13	12.94-20.54
^b DXA	23.73 \pm 4.35	18.80-29.40	22.34 \pm 4.02	22.30-25.40
^c ADP	20.57 \pm 3.14	16.44-24.21	17.43 \pm 6.43	8.73-24.62

Note: ^aH = percent body fat measured by deuterium dilution; ^bDXA = percent body fat measured by dual energy x-ray absorptiometry; ^cADP = percent body fat measured by air-displacement plethysmography

Table 8

Percent Body Fat of Females by Age and Feeding Group (n=42)

Age (years)	Breast-Fed (n=23)		Formula Fed (n=19)	
	Mean ± SD	Range	Mean ± SD	Range
3 (n=17)				
^a 2H	24.39 ± 4.36	18.33-30.15	24.00 ± 5.30	17.49-33.50
^b DXA	32.82 ± 5.64	30.60-42.30	30.43 ± 5.98	21.50-38.50
^c ADP	23.70 ± 10.20	5.37-34.11	26.02 ± 8.83	11.32-36.64
4 (n=13)				
^a 2H	25.70 ± 3.40	20.62-28.39	26.94 ± 1.72	24.67-30.62
^b DXA	29.91 ± 2.74	27.00-35.20	32.50 ± 4.53	26.10-36.70
^c ADP	24.21 ± 7.54	8.71-33.59	22.96 ± 5.40	15.28-27.57
5 (n=12)				
^a 2H	23.92 ± 3.72	19.70-30.78	24.27 ± 7.67	15.16-37.48
^b DXA	27.34 ± 6.35	21.20-39.70	26.98 ± 8.65	16.50-40.70
^c ADP	14.21 ± 9.49	2.49-27.28	26.36 ± 3.02	23.95-29.04

Note: ^a2H = percent body fat measured by deuterium dilution; ^bDXA = percent body fat measured by dual energy x-ray absorptiometry; ^cADP = percent body fat measured by air-displacement plethysmography

Dietary Intake of Participants

The mean intake of Kilocalories for all seventy-eight participants was 1,333.2 (*SD* = 323.7). The mean intakes of carbohydrate, protein, and fat grams were 189.0 (*SD* = 55.6), 45.5 (*SD* = 12.2), and 48.0 (*SD* = 16.4), respectively. A mean of 56.0%

Kilocalories came from carbohydrates ($SD = 6.3$), 13.6% from protein ($SD = 2.2$) and 31.9% from fat ($SD = 5.4$). Of the 31.9% Kilocalories from fat, 11.3% came from saturated fat ($SD = 2.5$), 2.8% from trans-fat ($SD = 1.0$), 12.1% from monounsaturated fat ($SD = 2.4$), and 5.7% from polyunsaturated fat (PUFA) ($SD = 1.6$). The mean calcium intake of the participants was 763.6 milligrams ($SD = 318.0$). The mean healthy eating index (HEI) score of the participants was 58.2 ($SD = 9.2$), and the mean juice intake was 7.0 fluid ounces ($SD = 5.1$). See Tables 9 and 10 for the means, standard deviations, and ranges of the study variables by gender. See Table 11 for means, standard deviations, and ranges of additional nutrients by gender.

The components of the HEI were summarized by gender to reveal diet consumption patterns of the male and female participants. Intake was similar for boys and girls for the following components: 1) total vegetables, 2) dark green and orange vegetables and legumes, 3) whole grains, 4) sodium, 5) and solid fats, alcoholic beverages and sugar (SoFAAS). Males and females were well below the optimal scores for all of those components, except sodium. Males had higher scores than females for the components of total fruit, whole fruit, total grains, and saturated fat, indicating they were closer to the goal intake for those components. Females had higher scores for the components of milk and meat and beans, indicating they were closer to the goal intake for those components. See Tables 13, 14, and 15 for a summary of HEI components by gender.

Table 9

Means, Standard Deviations, and Ranges of Food Intake: Kilocalories; Total Fat (g); PUFA (g); Calcium (mg); and Fruit Juice (fl oz) by Gender (N = 78)

Variable	Mean	SD	Range
Kilocalories			
All	1,333.2	323.7	651.8-2,428.3
Males	1,431.7	336.6	750.1-2,428.3
Females	1,248.7	290.2	651.8-1,869.9
Total Fat (g)			
All	47.4	14.1	20.9-86.1
Males	47.8	14.7	20.9-86.1
Females	47.0	13.8	24.2-82.1
^aPUFA (g)			
All	8.5	3.0	3.7-16.8
Males	8.3	2.6	4.3-15.0
Females	8.6	3.3	3.7-16.8
Calcium (mg)			
All	763.7	318.0	166.8-1,554.8
Males	810.3	369.1	166.8-1,524.6
Females	723.7	264.7	284.9-1,554.8
Fruit Juice (fl oz)			
All	7.0	5.1	0-20.3
Males	8.4	4.9	0-20.3
Females	5.8	5.0	0-17.8

Note. ^aPUFA = Polyunsaturated fatty acids

Table 10

Means, Standard Deviations, and Ranges of HEI Scores, NPAQ Scores, ²H, DXA, and ADP by Gender (N = 78)

Variable	Mean	SD	Range
^a HEI score			
All	58.2	9.2	41.9-81.2
Males	59.3	9.3	42.6-80.1
Females	57.3	9.1	41.9-81.2
^b NPAQ score			
All	72.1	12.3	34.3-100.0
Males	75.8	11.0	54.3-100.0
Females	68.8	12.5	34.3-94.3
^c ² H			
All	22.7	4.5	12.9-37.5
Males	20.4	3.2	12.9-28.1
Females	24.7	4.6	15.2-37.5
^d DXA			
All	27.5	5.7	16.5-42.3
Males	24.7	3.8	18.8-31.7
Females	30.0	5.9	16.5-42.3
^e ADP			
All	21.4	7.8	2.5-36.6
Males	19.6	6.2	4.6-33.7
Females	23.0	8.7	2.5-36.6

Note: ^a HEI = Healthy Eating Index; ^b NPAQ = Netherlands Physical Activity Questionnaire; ^c²H = percent body fat measured by deuterium dilution; ^dDXA = percent body fat measured by dual energy x-ray absorptiometry; ^eADP = percent body fat measured by air-displacement plethysmography

Table 11

Means, Standard Deviations, and Ranges of Dietary Fiber (g), Sodium (mg), Iron (mg), Vitamin C (mg), and Vitamin A (mg) by Gender (N = 78)

Variable	Mean	SD	Range
Dietary Fiber (g)			
All	9.5	3.3	3.2-18.0
Males	10.4	3.6	4.7-18.0
Females	8.8	2.9	3.2-15.4
Sodium (mg)			
All	2,135.8	612.1	1,123.9-4,300.1
Males	2,248.9	615.9	1,224.5-3,472.2
Females	2,038.8	599.0	1,123.9-4,300.1
Iron (mg)			
All	15.3	8.5	4.1-33.6
Males	17.2	8.9	4.6-33.6
Females	13.6	7.9	4.1-30.2
Vitamin C (mg)			
All	79.6	60.4	7.7-427.4
Males	86.1	70.6	13.3-427.4
Females	74.1	50.3	7.7-218.5
Vitamin A (IU)			
All	4,309.6	3,208.9	716.4-21,514.5
Males	4,462.2	2,897.6	765.0-13,649.8
Females	4,178.8	3,483.2	716.4-21,514.5

Table 12

Means, Standard Deviations, and Ranges of ^aHEI Adequacy Food Group Components by Gender (N = 78)

Component	Mean	SD	Range
Total Fruit			
All	3.2	1.9	0-5.0
Males	3.9	1.5	0.7-5.0
Females	2.6	2.0	0-5.0
Total Vegetable			
All	1.8	0.9	0.4-5.0
Males	1.8	0.9	0.4-4.0
Females	1.8	0.9	0.4-5.0
Total Grains			
All	4.7	0.7	0.7-5.0
Males	4.8	0.5	2.8-5.0
Females	4.6	0.9	0.7-5.0
Milk			
All	7.9	2.5	0-10.1
Males	7.7	3.0	0-10.1
Females	8.2	2.0	3.4-10.0
Meat and Beans			
All	7.6	2.1	1.9-10.0
Males	7.0	2.3	1.9-10.0
Females	8.0	1.8	2.7-10.0

Note. Maximum points for total fruit, total vegetable, and total grains is 5. Maximum points for milk and meat and beans is 10. ^aHEI = Healthy Eating Index

Table 13

Means, Standard Deviations, and Ranges of Other ^aHEI Adequacy Components by Gender (N = 78)

Component	Mean	SD	Range
Whole Fruit			
All	2.5	2.0	0-5.0
Males	3.2	1.9	0.6-5.0
Females	1.9	1.9	0-5.0
^b DGOVL			
All	0.6	1.0	0-5.0
Males	0.6	0.9	0.4-3.9
Females	0.7	1.1	0-5.0
Whole Grains			
All	1.9	1.8	0-5.0
Males	1.6	1.7	0-5.0
Females	2.2	1.8	0-5.0
Oils			
All	10.0	0.2	8.3-10.0
Males	10.0	0.3	8.3-10.0
Females	10.0	0	10.0-10.0

Note. Maximum points for whole fruits, DGOVL, and whole grains is 5. Maximum points for oils is 10. ^aHEI = Healthy Eating Index; ^bDGOVL = Dark Green and Orange Vegetables and Legumes

Table 14

Means, Standard Deviations, and Ranges of ^aHEI Moderation Components by Gender (N = 78)

Variable	Mean	SD	Range
Saturated Fat			
All	2.1	3.0	0-10.0
Males	3.0	3.5	0-10.0
Females	1.3	2.3	0-9.0
Sodium			
All	4.2	2.3	0-9.2
Males	4.2	2.3	0-9.2
Females	4.1	2.3	0-9.0
^bSoFAAS			
All	11.8	3.9	3.9-20.0
Males	11.7	3.9	4.7-19.6
Females	11.9	3.9	3.9-20.0

Note. Maximum scores for saturated fat and sodium is 10. Maximum score for SoFAAS is 20. ^aHEI = Healthy Eating Index; ^bSoFAAS = Solid Fats, Alcoholic Beverages, and Added Sugars

Relationship between Percent Body Fat and Dietary and Physical Activity Factors

Pearson's Product Moment Correlation Coefficients were calculated to examine the relationships between percent body fat and the following variables in hypothesis one: 1) intake of total fat (g), 2) intake of PUFA (g), 3) fruit juice (fl oz), 4) calcium (mg), 5) total Kilocalories, 6) HEI scores, and 7) Netherland's Physical Activity Questionnaire (NPAQ) scores. Two significant correlations were observed in the male participants. A

significant, positive relationship was found between percent body fat measured by DXA and HEI scores, $r(36) = .332, p < .05$, and a significant, negative relationship was found between percent body fat measured by DXA and NPAQ scores, $r(36) = -.441, p < .01$. These data suggest that percent body fat is higher in males with higher diet quality, and percent body fat is lower in males who are more physically active. The strength of the associations were considered based on Cohen's (1992) effect size estimates that r values of 0.10, 0.30 and 0.50 indicate small, medium and large effects, respectively. The strength of the associations between percent body fat measured by any of the methods and total fat, PUFA, fruit juice, calcium, and total Kilocalories failed to meet the level of a small effect. On the other hand, the associations between percent body fat measured by DXA and HEI scores and NPAQ scores for the male participants had a medium effect.

No significant correlations were found between percent body fat and the variables in hypothesis one for the female participants. However, several variables were approaching significance in the females including: 1) NPAQ scores and percent body fat measured by ^2H , $r(42) = -.246, p = .116$, 2) juice intake and percent body fat measured by DXA and ADP, $r(42) = .246$ and $204, p = .116$ and $.195$, respectively, 3) fat intake and percent body fat measured by ADP, $r(42) = .202, p = .200$, and 4) PUFA intake and percent body fat measured by ADP, $r(42) = .239, p = .128$. Significant results may have been found if more females were included in the analyses.

Other interesting, significant correlations emerged from the analyses. Significant, positive relationships were found in both male and female participants for the following variables: 1) fat intake and PUFA intake, $r(36) = .814, p < .01$ in males, $r(42) = .799, p$

<.01 in females, 2) Kilocalorie intake and fat intake, $r(36) = .843, p <.01$ in males, $r(42) = .868, p <.01$ in females, 3) Kilocalorie intake and PUFA intake, $r(36) = .812, p <.01$ in males, $r(42) = .584, p <.01$ in females, 4) Kilocalorie intake and calcium intake, $r(36) = .621, p <.01$ in males, $r(42) = .460, p <.01$ in females, 5) HEI scores and calcium intake, $r(36) = .457, p <.01$ in males, $r(42) = .399, p <.01$ in females, and 6) percent body fat measured by DXA and ^2H , $r(36) = .702, p <.01$ in males, $r(42) = .800, p <.01$ in females.

Other significant relationships were gender-dependent. A significant, positive relationship between PUFA intake and calcium was found in male participants only, $r(36) = .385, p <.05$, suggesting males who consume more PUFA also consume more calcium. The following significant relationships were found in female but not male participants: 1) juice intake and fat intake, $r(42) = .396, p <.01$, 2) Kilocalorie intake and juice intake, $r(42) = .532, p <.01$, 3) fat intake and HEI scores, $r(42) = -.317, p <.05$, 4) percent body fat measured by ^2H and ADP, $r(42) = .494, p <.01$, and 5) percent body fat measured by ADP and DXA, $r(42) = .520, p <.01$.

Table 15

Correlations between Study Variables by Gender (N = 78)

	^a Fat	^b PUFA	Juice	Calcium	^c Kcals	^d HEI	^e NPAQ	^f 2H	^g DXA
^b PUFA									
Male	.814**								
Female	.799**								
Juice									
Male	-.018	.135							
Female	.396**	.248							
Calcium									
Male	.310	.385*	-.055						
Female	.208	-.042	.201						
^c Kcals									
Male	.843**	.812**	.169	.621**					
Female	.868**	.584**	.532**	.460**					
^d HEI									
Male	-.228	.147	.140	.457**	.049				
Female	-.317*	-.203	-.207	.399**	-.144				
^e NPAQ									
Male	.322	.167	-.163	-.064	.089	-.182			
Female	.283	.278	.133	.071	.244	-.049			
^f 2H									
Male	-.252	-.176	.085	.013	-.218	.213	-.258		
Female	-.041	-.074	.168	-.073	-.114	-.051	-.246		
^g DXA									
Male	-.240	-.148	.296	.150	-.075	.332*	-.441**	.702**	
Female	-.059	-.073	.246	.028	-.089	.069	-.008	.800**	
^h ADP									
Male	-.187	-.052	-.127	.022	-.140	.236	-.176	.227	.266
Female	.202	.239	.204	.009	.133	-.133	.098	.494**	.520**

Note. All correlations coefficients are Pearson's Product Moment Correlations. *p < .05, **p < .01 ^aFat = total fat; ^bPUFA = polyunsaturated fatty acids; ^cKcals = total KiloCalories; ^dHEI = Healthy Eating Index; ^eNPAQ = Netherlands Physical Activity Questionnaire; ^f2H = percent body fat measured by deuterium dilution; ^gDXA = percent body fat measured by dual energy x-ray absorptiometry; ^hADP = percent body fat measured by air-displacement plethysmography.

Comparison of Body Fat Measures in Breast-Fed and Formula-Fed Children

A repeated measures multi-variate analysis of variance was used to test hypotheses two, three, four, and five. The dependent variables were the methods for measuring body fat (^2H , DXA, and ADP) and the independent variables were gender and feeding group during infancy. The following interactions were tested: 1) methods and gender, 2) methods and feeding group, and 3) methods, gender, and feeding group. There were no significant interactions between any of the variables: methods and gender, $F(2, 74) = .720, p = .489$, partial eta squared = .010; methods and feeding group, $F(2, 74) = 1.859, p = .159$, partial eta squared = .025; methods, gender, and feeding group, $F(2, 74) = 2.272, p = .107$, partial eta squared = .030. The main effect for gender was significant, $F(1, 74) = 17.432, p < .0001$, partial eta squared = .191, implying a significant difference in mean percent body fat of the male and female participants.

The main effect of feeding group was not significant, $F(1, 74) = .553, p = .460$, partial eta squared = .007, suggesting no differences in mean percent body fat of participants who were fed breast milk versus formula during infancy. The main effect of methods was significant, $F(2, 148) = 42.892, p < .0001$, partial eta squared = .367. Bonferroni post-hoc comparison tests indicated that mean percent body fat measured by DXA ($M = 27.53, SD = 5.67$) was significantly different from mean percent body fat measured by ^2H ($M = 22.71, SD = 4.54$) and ADP ($M = 21.42, SD = 7.83$), but there was no significant difference in mean percent body fat measured by the ^2H and ADP methods. However, the correlation coefficient between ^2H and ADP was only moderate ($r = .461, p$

<.01). This suggests that while ADP may estimate mean percent body fat well in a group of children, it is not as accurate on an individual basis. In contrast, there was a strong correlation between DXA and ^2H ($r = .823, p < .01$). This demonstrates that DXA correlates well with the criterion method, ^2H , although it overestimates in most cases. See Table 16 for the means and standard deviations of the body fat measures by gender and feeding group and Figures 1 and 2 for a comparison of ^2H to DXA and ADP.

Table 16

Means and Standard Deviations for Body Fat Measures of Breast Fed and Formula Fed Males and Females

	<u>Male</u>				<u>Female</u>			
	<u>Breast Fed</u> (n = 17)		<u>Formula Fed</u> (n = 17)		<u>Breast Fed</u> (n = 22)		<u>Formula Fed</u> (n = 18)	
	<i>Mean</i>	<i>SD</i>	<i>Mean</i>	<i>SD</i>	<i>Mean</i>	<i>SD</i>	<i>Mean</i>	<i>SD</i>
BFADP	19.55 ^{a,x}	4.46	19.55 ^{a,x}	7.90	20.99 ^{a,x}	9.84	25.48 ^{a,x}	6.62
BF2H	20.00 ^{a,x}	3.12	20.82 ^{a,x}	3.36	24.70 ^{b,y}	3.75	24.71 ^{a,y}	5.55
BFDXA	24.78 ^{b,x}	3.77	24.56 ^{b,x}	3.89	30.14 ^{c,y}	5.35	29.79 ^{b,y}	6.68

Note: Measure effect, $F(2, 148) = 42.89, p < .00001$; Gender effect, $F(1, 74) = 17.43, p < .00001$; Feeding Type effect, $F(1, 74) = .553, p = .460$; Measure x Gender effect, $F(2, 148) = .720, p = .489$; Measure x Feeding Type effect, $F(2, 148) = 1.86, p = .159$; Gender x Feeding Type effect, $F(1, 74) = .310, p = .579$; Measure x Gender x Feeding Type effect, $F(2, 148) = 2.27, p = .107$. Column means not sharing a superscript (a, b, or c) are statistically different by Fisher's pairwise comparisons, $p < .05$. Row means not sharing a superscript (x or y) are statistically different by Fisher's pairwise comparisons, $p < .05$.

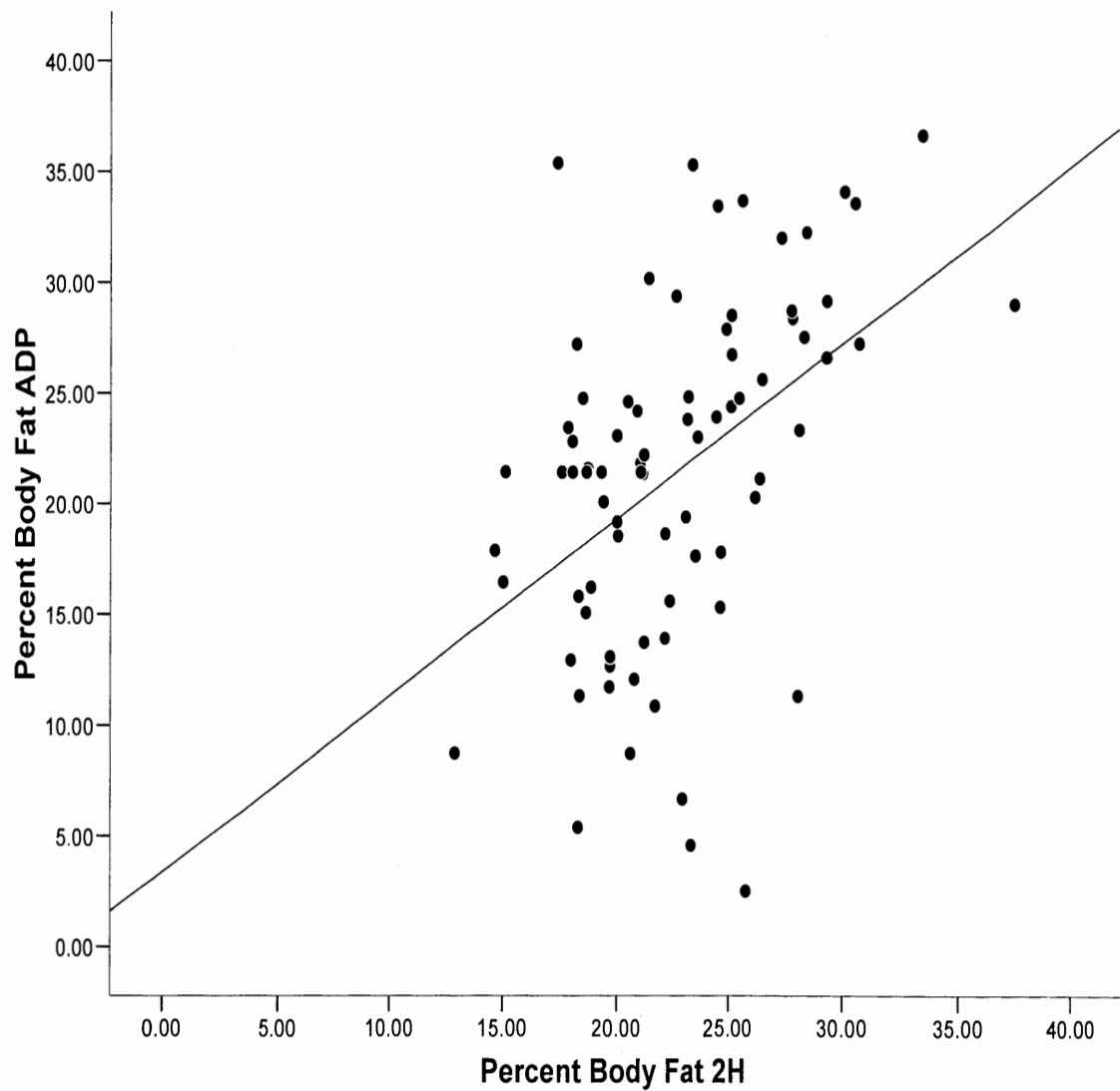


Figure 1. Comparison of percent body fat of participants ($n = 78$) measured by ^2H and ADP, $r(78) = .461$, $p < .01$; ^2H = deuterium oxide; ADP = air displacement plethysmography

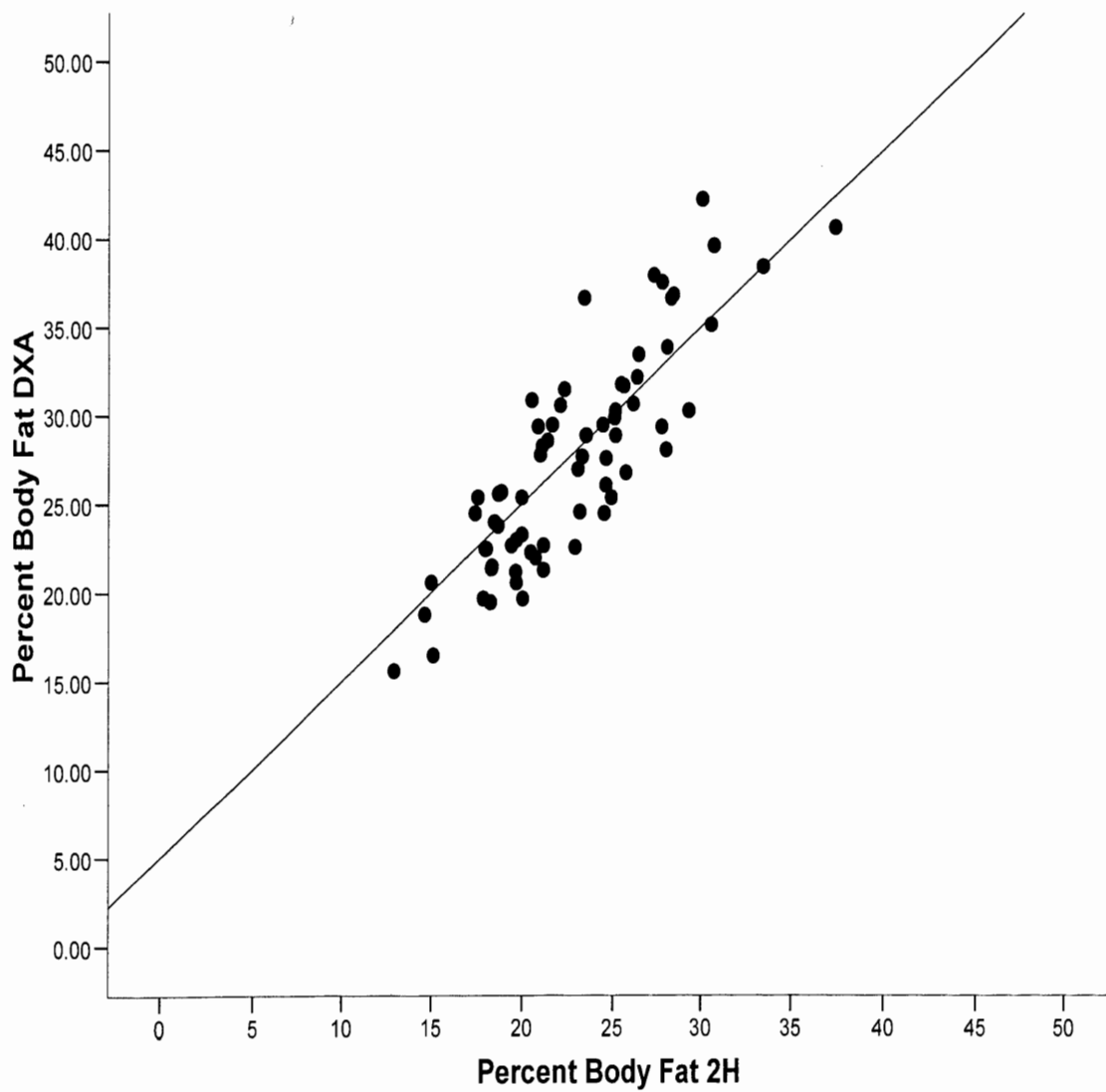


Figure 2. Comparison of percent body fat of participants ($n = 78$) measured by ^2H and DXA, $r(78) = .823$, $p < .01$; ^2H = deuterium oxide; DXA = dual energy x-ray absorptiometry

CHAPTER V

DISCUSSION, CONCLUSIONS, AND RECOMMENDATIONS

This study addressed several topics that have been identified as gaps in the research literature. First, cross-sectional studies investigating the associations between environmental factors and percent body fat were lacking in young children. The present study used percent body fat as an outcomes measure to test relationships of several dietary and physical activity factors. Second, data linking juice intake and other nutrients was deficient in the literature which this study addressed. Third, several studies investigated the association between dairy consumption and body fat in children, but they did not focus on a specific component in dairy foods and looked at a wide age range of children. The present study looked specifically at calcium intake and studied a narrow age range of children. Fourth, studies investigating the impact of diet quality using a standardized tool in children were lacking. The present study used the HEI, a standardized tool, to assess diet quality. Fifth, the present study used an age-appropriate tool to assess physical activity that accounted for various forms of sedentary behaviors as was recommended in the literature. And sixth, this study is the first known to use ADP to assess body composition in children under the age of 5.

Discussion

Anthropometric and Body Composition Characteristics of Participants

The number of children at risk for overweight and overweight in the study was similar to the national average. Twenty-four percent of the children in the study had a body mass index (BMI) for age at the 85th percentile or higher, and 14% had a BMI for age at the 95th percentile or higher. This is similar to national data for 2 to 5 year old children that show 26.2% of children have a BMI for age at or above the 85th percentile, and 13.9% have a BMI for age at or above the 95th percentile (Ogden et al., 2006).

Few studies have reported body composition in children 3 to 5 years old. The majority of studies reviewed used BMI as an outcomes measure, and when body composition was assessed, the most common methods used were bioelectrical impedance analysis (BIA) and skin fold measures. Dual energy x-ray absorptiometry has been used by a few researchers to assess percent body fat in young children. Because the use of the ADP method with children is more recent, no published studies were identified measuring percent body fat with ADP in children under age 5.

DeJongh, Binkley, and Specker (2006) used DXA (Hologic QDR-4500A, software version 8.2) to assess percent body fat in a group of 3 to 5 year old children that included 46 males and 42 females. They found the mean percent body fat of the males and females to be 23.1 ($SD = 3.5$) and 27.8 ($SD = 4.6$), respectively. This is similar to the values obtained in the present study using DXA where males and females had mean percent body fat levels of 24.67 ($SD = 3.77$) and 29.98 ($SD = 5.91$), respectively. On the other hand, earlier studies showed children this age to have lower body fat levels when

measured by DXA (Hologic QDR-2000, software version 5.56), although the sample size was smaller in the studies (Ellis, 1997; Ellis, et al., 1997). The mean percent body fat values reported were 17.6 ($SD = 2.0$) in a group of 12 males and 20.9 ($SD = 4.6$) in a group of 18 females. The higher values obtained in the present study may be the result of an increase in fat mass in children over the past decade. Wells, Coward, Cole, and Davies (2002) compared contemporary children with those from two decades ago and documented a significantly greater fat mass in both males and females. Additionally, the prevalence of children at risk for becoming overweight and being overweight has increased by approximately 4% since the research articles by Ellis and colleagues were published in 1997 (Ogden et al., 2006).

The studies by Dejongh et al. (2006), Ellis (1997), and Ellis et al. (1997) also suggest that gender differences in percent body fat are present in early childhood. The present study had similar findings with males having significantly lower percent body fat levels than females (see Tables 7 and 8). Based on these data, as well as the national average BMI data presented by Ogden et al. (2006), the anthropometric and body composition characteristics of the present study's sample appear similar to that previously reported for 3 to 5 year-old children in the United States.

Dietary Intake of Participants

The participants mean intake of Kilocalories and macronutrient distribution were within the recommended ranges established by the Institute of Medicine's Dietary Reference Intakes (DRI) Macronutrient Report (2002). The recommended range of Kilocalories for children this age is 1,000 to 1,600 Kilocalories, and the recommended

macronutrient distribution for percentage of Kilocalories from carbohydrates, protein and fat are 45-65%, 5-30%, and 25-35%, respectively. However, the participants' intake of saturated fat exceeded the recommendations established by the 2005 Dietary Guidelines for Americans. The recommendation is to limit saturated fat to no more than 10% of total Kilocalories and the participants' average was 11%. Macronutrient intake in this study was similar to what other researchers have reported in children (Aeberli et al., 2007; Carruth & Skinner, 2001).

Mean calcium intake of study participants was >95% of the DRI. The DRI for calcium for children 1 to 3 years of age is 500 mg, and for children 4 to 8 years of age is 800 mg. Average calcium intake for participants in the study was similar among all age groups with 3 year olds consuming an average of 769.3 mg ($SD = 312.9$), 4 year olds 769.9 mg ($SD = 331.9$), and 5 year olds 777.1 mg ($SD = 345.6$). Male participants consumed higher levels of calcium than females ($M = 810.3$, $SD = 369.1$ v. $M = 723.7$, $SD = 264.7$). Children's calcium intake reported by other researchers is similar to that of the present study and indicates that males average higher intakes of calcium than females (Aeberli et al., 2007; Carruth & Skinner, 2001)

Juice consumption of participants in the study exceeded the recommendation by the American Academy of Pediatrics (2001) that children 1 to 6 years old should consume no more than 4-6 fluid ounces of fruit juice per day. O'Connor et al. (2006) reported a similar trend that young children exceeded the recommended intake for juice. Both 100% fruit juice and juice drinks were combined when determining the participants'

intake in the present study because parents were not always sure if the juice their children consumed was 100% fruit juice.

The Healthy Eating Index (HEI) scores of participants in the present study indicate the participants' diet quality needs improvement (see Table 10 for means, standard deviations, and ranges of HEI scores by gender). The Center for Nutrition Policy and Promotion stated that HEI scores higher than 80 indicated good diet quality, scores between 51 and 80 represented diets that needed improvement, and scores less than 51 were indicative of poor diet quality (Guenther et al., 2007).

The highest scoring HEI components were those for total grains and oils with optimal scores for these components being 5 and 10, respectively. Participants averaged scores of 4.7 ($SD = 0.7$) for total grains and 10.0 ($SD = 0.2$) for oils. The components that scored lowest included intake of total vegetables, green and orange vegetables, whole grains, saturated fat, and sodium. All of these components had averages that were less than half the optimal score (see Tables 12, 13, and 14 for means, standard deviations, and ranges of HEI score components by gender). Other researchers have reported similar findings that children do not meet the recommended intakes for whole grains and vegetables and their diets tend to exceed the recommendation for saturated fat (Aeberli et al., 2007; Carruth & Skinner, 2001; Langevin et al., 2007).

Relationship between Percent Body Fat and Environmental Factors

Hypothesis one stated that there would be no significant relationship between the percentage of body fat in 3 to 5 year old children and the following variables: mean daily intake of total fat grams, polyunsaturated fatty acid (PUFA) grams, fruit juice in fluid

ounces, calcium in milligrams, total daily energy in Kilocalories, HEI scores, and Netherlands Physical Activity Questionnaire (NPAQ) scores. The hypothesis failed to be rejected for the following variables: mean daily intake of total fat grams, PUFA grams, fruit juice in fluid ounces, and calcium in milligrams. The hypothesis was rejected for male participants on the variables of HEI scores ($p < .05$) and NPAQ scores ($p < .01$).

The negative relationship between NPAQ scores and percent body fat found in the male participants was expected based on data presented by other researchers that a positive correlation exists between weight and sedentary behavior in children (Lumeng et al., 2006; Vandewater et al., 2004). No significant correlation between NPAQ scores and percent body fat was found in the female participants. However, it should be noted that the association between NPAQ scores and percent body fat measured by ^2H in the female participants was approaching the level of significance, $r(42) = -.246, p = .116$. Thus, it appears that physical activity levels impact percent body fat in males and females.

The significant and positive association between HEI scores and percent body fat observed in the male participants suggests that male children with higher diet quality scores have higher body fat levels. This finding was not expected because other researchers have suggested that children with lower quality diets are more likely to be overweight (Hassapidou et al., 2006; Langevin et al., 2007). It should be noted that very few children in the present study had HEI scores that were classified as good, and most children had scores that needed improvement (see Table 10). Including more children with good diet quality scores may have produced different results. In addition, the HEI

encompasses multiple components of the diet, and Guenther et al. (2007) stated that it was not intended to capture weight management. A low score may reflect under-consumption or over-consumption of food groups and nutrients. For example, an individual who does not consume enough servings from food groups will have a low score and an individual who consumes too much saturated fat and sugar will also have a low score. Thus, multiple variables comprise the HEI score, and the moderate relationship observed may be the result of confounding variables.

There was no significant difference between percent body fat of the participants and total fat grams, PUFA grams, fruit juice, calcium intake, and total Kilocalories. Total fat, PUFA and juice intake have been examined by other researchers who reported similar findings (Aeberli et al., 2007; Hassapidou et al., 2006; Magarey et al., 2001; O'Connor et al., 2006). In contrast, Skinner et al. (2003) found total fat to be a positive predictor and PUFA to be a negative predictor of percent body fat in a prediction model.

Several theories have been proposed regarding calcium's role in body fat regulation. One popular belief is that the release of calcitropic hormones in response to inadequate calcium intake results in increased levels of intracellular calcium, an observation noted for some obese individuals (Parikh & Yanovski, 2003; Pereira et al., 2002; Zemel et al., 2000). Experimental data support this theory. Zemel et al. found that mice on calcium-deficient diets gained more weight, had higher fatty acid synthase activity, and a significant decrease in lipolysis compared to mice receiving adequate calcium diets. They also found mice receiving adequate calcium diets to have higher core body temperatures, suggesting that calcium may influence thermogenesis.

The link between calcium intake and percent body fat in human studies is equivocal. Several researchers have reported no association as was found in the current study (Lappe et al., 2004; O'Connor et al., 2006; Phillips et al., 2003). On the other hand, Carruth and Skinner (2001) found both calcium intake and number of dairy servings to be negatively associated with percent body fat. They followed children over an eight-year period and collected diet information six times. The longitudinal nature of the study may be one of the reasons their findings differed from the present study because they were able to assess calcium intake at several time points. Additionally, calcium intake was high for the participants in the present study with mean intakes >95% of the DRI. The lack of a correlation may be the result of few children having low calcium intakes.

Other significant correlations resulted that were not included in hypothesis one. These correlations indicate that boys and girls who consume more Kilocalories also consume more fat, including PUFA, and calcium. The relationship between HEI scores and calcium suggests that boys and girls with a higher quality diet consume more calcium. The positive associations between juice intake and Kilocalories and juice intake and fat imply that females who consume more juice also consume more Kilocalories and fat. This is an important finding since excess energy intake is a factor leading to weight gain. The negative association between fat intake and HEI scores indicates that females who have higher quality diets consume less fat. In addition, the DXA method for measuring body fat was shown to be highly correlated to the criterion method, ^2H , in both male and female participants. The positive relationships observed in females between

percent body fat measured by ^2H and ADP and percent body fat measured by ADP and DXA show that ADP correlates to ^2H and DXA more strongly in females than in males. See Table 15 for correlations between variables by gender.

Comparison of Body Fat in Breast-Fed and Formula-Fed Children

Hypothesis two failed to be rejected. There was no significant difference found in the percentage of body fat, measured by DXA, ADP or ^2H , of 3 to 5 year old children fed breast milk versus those fed infant formula during the first 6 months of life. Other researchers have explored this topic and the results have been equivocal. Apfelbacher et al. (2008) found that breastfeeding for more than 3 months was negatively related to being overweight. Toshke et al. (2007) also found a negative relationship between fat mass and breastfeeding. However, Burdette et al. (2006), Michels et al. (2007) and Ong et al. (2006) did not find breastfeeding to be related to body fat or BMI.

Several factors make it challenging to design studies that explore a direct relationship between breastfeeding and weight or adiposity later in life. First, many studies, including the present study, have relied on retrospective data from parents to determine infant feeding. Second, different outcome measures have been used among studies making comparisons problematic. And third, many environmental factors impact weight and adiposity over time making it difficult to measure the effect of one single factor. Thus, no conclusive evidence exists regarding the effects of breastfeeding on weight and adiposity later in life.

Comparison of Body Fat Measures

Hypotheses three, four, and five addressed if differences existed in the three methods for measuring body fat. Hypothesis three stated that there would be no significant difference in the percentage of body fat of 3 to 5 year old children as measured using DXA versus measured by the criterion method, deuterium dilution. This hypothesis was rejected. Hypothesis four stated that there would be no significant difference in the percentage of body fat of 3 to 5 year old children as measured using ADP versus measured by the criterion method, deuterium dilution. Hypothesis four failed to be rejected. There was a significant difference in the mean values of all three methods which led to a rejection of hypothesis five that stated there was no significant difference in the percentage of body fat of children 3 to 5 years old as measured using DXA, ADP and deuterium dilution. The difference in the three methods was attributed to DXA which measured mean percent body fat significantly higher than either ^2H or ADP.

Findings from this study show that mean percent body fat values obtained from ADP and ^2H are similar in 3 to 5 year old children, varying by only 1.4%. On the other hand, mean percent body fat measured by DXA differs by approximately 5-6% when compared to mean percent body fat measured by ^2H and ADP. This difference is robust as evidenced by a partial eta squared value of 0.367.

Other researchers have reported similar results when comparing DXA and ADP to criterion methods. Several researchers have stated that DXA is inaccurate compared to a criterion method (Roemmich et al., 1997; Fields & Goran, 2000; Sopher et al., 2004). Specifically, Roemmich et al. reported that DXA overestimated body fat in children and

adolescents by 4.4% when compared to a four-compartment (4-C) model. Dual energy x-ray absorptiometry analysis makes an assumption that the hydration of fat-free mass is 73%. It has been documented that this value is higher in children, and this may explain the inaccuracies reported (Baumgartner et al., 1991; Wang et al., 1999; Withers et al., 1998). No other published studies were identified that used ADP in children this age for comparison. However, Ellis (2000) reported the accuracy of ADP in adults to be within 2-3% of a criterion method.

Caution must be taken when interpreting these results. Multivariate analysis of variance results showed mean ADP values compared well to mean ^2H values (see Table 16). However, the correlation coefficient between the two methods was only moderate ($r = .461, p < .01$). This suggests that while ADP may estimate mean percent body fat well in a group of children, it is not as accurate on an individual basis. In contrast, there was a strong correlation between DXA and ^2H ($r = .823, p < .01$). This demonstrates that DXA correlates well with the criterion method, ^2H , although it overestimates in most cases. See Table 15 for the correlation coefficients between all of the methods by gender and Figures 1 and 2 for comparisons of percent body fat measured by DXA versus ^2H and ADP versus ^2H .

Several factors may have influenced the individual ADP results causing the values to be only moderately correlated to ^2H . First, a booster seat and DVD player were introduced into the test chamber. The additional items present inside the chamber may have decreased the accuracy of some results, although these items were present inside the chamber during the calibration phase of testing and during the daily quality control of the

machine. The heat produced by the DVD player may have affected the behavior of the air inside the chamber during testing. Second, thoracic gas volume (TGV) was predicted, not measured directly in the children. Third, equations were used for predicting surface area artifact and density of fat free mass. These factors led to several adjustments in the raw volume measures and the percent body fat calculations obtained from the machine. The adjustments made were extensively researched and supported in the literature. However, assumptions made in the calculations may have led to inaccuracies. Fourth, the ratio of the volume of the chamber to the volume of the study participants may have been a factor. The ADP machine was designed for adults weighing >40 kg. A smaller calibration cylinder was used, but this may not have been enough to overcome the differences in volume. A smaller test chamber may be necessary for testing young children.

Dual energy x-ray absorptiometry is the gold standard for measuring bone density. Many researchers also use it as a criterion method for soft tissue analysis including lean and fat mass. However, assumptions made by DXA manufacturers regarding soft tissue may be problematic when using it to measure fat mass and lean tissue, especially in young children. It is assumed that the fat to lean ratio of soft tissue over bone is the same as the rest of the body's soft tissue and that the hydration of fat free mass is approximately 73% (Ellis, 2000; Fields & Goran, 2000; Roemmich et al., 1997; Sopher et al., 2004; Testolin et al., 2000). This is not a correct estimate regarding hydration of fat free mass for children which has been shown to be higher than adults (Baumgartner et al., 1991; Hewitt et al., 1993; Wang et al., 2003).

Another issue is that periodic updates are made by DXA manufacturers to improve bone detection. These updates result in new algorithms that not only impact bone mineral values but also the values for soft tissue. Shypailo, Butte, and Ellis (2008) showed that percent body fat differed significantly using Hologic's most recent software upgrade (version 12.1) as compared to the previous software (version 11.2) in children weighing less than 40 kg. Use of the new software produced percent body fat values that were 3-4% higher in 3 to 5 year old children (n = 183). The difference was greater in females than males and in the smaller, younger children. Thus, the assumptions made by DXA manufacturers and the periodic upgrades must be considered when interpreting results and suggest that DXA may not be appropriate for use as a criterion method in children.

Limitations

The study was limited by several factors. The study participants were from a limited geographic region in Central Arkansas. In addition, the cross-sectional nature of the study made it necessary to rely on memory of the parents for some of the data. Parents were asked to recall how their child was fed as an infant and some may not have recollected the information accurately. This could have impacted the results obtained for hypothesis two. In addition, a questionnaire administered one time was used to assess the child's physical activity level instead of directly observing this over time. This should be considered when interpreting the results of hypothesis one related to physical activity.

Reliance on study participants to provide dietary intake data always poses potential problems and may have negatively impacted the results of the study related to

dietary factors and body fat. Three-day food records were used to assess the dietary factors of interest in hypothesis one. Parents recorded the children's intake after receiving written and verbal instructions on properly recording all foods and beverages consumed along with appropriate portion sizes. Intake may have been over-estimated or under-estimated despite the efforts made by the researcher to ensure accuracy. Many of the children also consumed meals away from home without parental supervision. Therefore, intake had to be estimated based on reports of second parties in some cases who may not have recalled all foods consumed and accurate serving sizes.

Total body water analysis using deuterium dilution was chosen as the criterion method in this study instead of a four compartment (4-C) model due to the impracticality of using a 4-C model with children. Total body water is a two-compartment model that puts all components of the fat-free mass (FFM) into one category. Adjustments were made in the calculations for body fat to account for the differences in hydration of FFM for children. Nevertheless, it is a disadvantage in this study that bone and protein were not measured directly.

The ADP method requires that TGV be factored into the equation when estimating body volume. This is important because the air in the lungs is more compressed than most of the air in the plethysmograph and inaccuracies will result if this is not taken into account. However, the procedure to directly measure TGV is difficult to perform. Therefore, TGV values were predicted in this study instead of being directly measured. This is a shortcoming of the study although it is unlikely that the young children in this study would have been able to provide direct measures of TGV.

Conclusions

Overweight in childhood and adolescence is a national health crisis. Prevalence has been on the rise since the 1980's, and without interventions, the problem will continue to escalate. Thus, it is crucial to pinpoint environmental factors related to excess body fat in young children so that strategies can be formulated to reverse this trend. This study contributed to the literature by further exploring several environmental factors previously cited to have potential links to adiposity in children. It added to the evidence that percent body fat is influenced by physical activity levels at a young age. While it has been suggested that breastfeeding may offer protection from obesity (Apfelbacher et al., 2008; Toshke et al., 2007), this notion was not supported in the present study. Numerous health benefits have been well-documented with regard to breastfeeding (AAP, 2005), but there is insufficient evidence at the present time to support a direct link between breastfeeding during infancy and adiposity in childhood.

Infancy and early childhood have emerged as key lifecycle stages in which body composition should be closely monitored. Therefore, the need for practical, accurate methods for assessing body composition in young children is essential. This study assessed the accuracy of DXA and ADP for measuring percent body fat in children. The findings revealed there are advantages and disadvantages to both methods for measuring percent body fat.

Some drawbacks noted for DXA were exposure to radiation and the requirement for the child to lie completely still throughout the scan. Ten children in the present study were not able to complete the DXA test because they could not lie still or they became

afraid when the table moved. In addition, evidence was provided that percent body fat values measured by DXA are overestimated in children by approximately 5-6% as compared to the criterion method, ^2H . This overestimation was fairly consistent as shown by the strong correlation between DXA and ^2H . Therefore, DXA may be useful in a clinical setting to monitor percent body fat of children as long as the absolute value obtained is not crucial.

Air displacement plethysmography does have some advantages over DXA. It does not require that the child be completely still and there is no radiation exposure. The participants in the present study were more likely to complete the ADP test than DXA. Mean percent body fat values obtained from ADP compared well with the criterion method, ^2H . Air displacement plethysmography may be an acceptable method when determining the average percent body fat values of groups of children. However, the moderate correlation between ADP and ^2H shows that ADP needs additional adjustments before it can be used effectively in individual assessments. All of these issues must be considered by researchers and clinicians when choosing a method for assessing body composition in young children.

Recommendations

No correlations were observed between percent body fat and total fat, PUFA, fruit juice, calcium, or total Kilocalories. However, some other interesting correlations emerged from the study warranting further investigation. These include the moderate to strong correlations observed between fat and PUFA, Kilocalories and fat, Kilocalories and PUFA, Kilocalories and calcium, HEI scores and calcium, PUFA and calcium, juice

and fat, juice and Kilocalories, and fat and HEI scores. The interactions of all of these variables are important to understand in order to provide appropriate dietary recommendations to young children.

Future studies examining the relationship between calcium intake and percent body fat should include individuals with a wide range of calcium intakes. This would allow one to see if low calcium diets influence body fat levels differently than moderate and high calcium diets. Participants in the present study had high mean calcium intakes which may be the reason why no relationship was observed.

The accuracy of methods for measuring body fat in children requires further exploration. Dual energy x-ray absorptiometry should continue to be tested against criterion methods as updated software is introduced. Correction factors could be considered in the future for groups in which body fat is consistently over-estimated by DXA. Ways of improving the accuracy of ADP in young children should be explored, as well. A smaller test chamber may produce better individual results. In addition, practical methods for measuring TGW in young children should be investigated. Measured values instead of predicted values may improve accuracy in some individual cases. Both DXA and ADP show promise as methods for measuring body fat in young children.

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APPENDIX A

IRB Approval

**Institutional Review
Board
(Human Research
Advisory
Committee)**

Office of Research
and Sponsored
Programs

4301 West
Markham, #636
Little Rock, AR
72205-7199

501-686-5667
501-686-8359 (fax)

www.uams.edu/orsp



FWA00001119

Date: 11/16/2006

Original Review Date
10/10/2006

Tina Crook
1212 Marshall Street, Mailslot 512-20B
Little Rock, AR 72202

Protocol Title: Accuracy of air-displacement plethysmography and dual-energy x-ray absorptiometry for estimating body fat in children age 2 to 5 years

Original Review
Date: 10/10/2006
Principal
Investigator: Tina
Crook
Protocol Sponsor:

Record: 72850

PRN:

FINAL APPROVAL (Major Revisions Met)

The Institutional Review Board approved your major revisions on 11/14/2006 at the latest convened meeting.

Approval period: 11/14/2006 through 11/14/2007.

Approved Documents:

CV Tina Crook Version: 1 Dated: 10/2/2006
PI Letter to IRB Cover Letter Version: 1 Dated: 10/2/2006
Consent Document Version: 1 Dated: 10/2/2006
HIPAA Authorization Version: 1 Dated: 10/2/2006
CRF Version: 1 Dated: 10/2/2006
Protocol Version: 1 Dated: 10/2/2006
CV, Tina Crook Version: 1 Dated: 10/2/2006
Original Submission Version: 1 Dated: 10/2/2006
DSMP Version: 1 Dated: 10/2/2006
Advertisement Version: 1 Dated: 10/2/2006
PI Response to IRB Referral Version: 1 Dated: 11/1/2006
Consent Form Version 2 Version: 2 Dated: 11/1/2006
UAMS Radiation Committee Approval Letter Version: 1 Dated:
10/12/2006
Modification form in response to Major Revisions Version: 1 Dated:
11/1/2006

Next Review Due: 11/14/2007

The risk to children who enter this study was determined to be Category 1,
as described in 45 CFR 46.404.

Please report to the IRB all changes, adverse reactions/deaths or closure of
the study as soon as possible.

Signed: Jimmie Valentine **for** Jimmie Valentine, IRB Committee Chair
on: 11/16/2006 **at:** 16:37:01

[x] _____

Close this Window

[x]



201 Donaghey Avenue
Conway, Arkansas 72035-0001
Telephone: (501) 450-3451
Fax: (501) 450-5339

May 23, 2007

Ms. Tina Crook
University of Central Arkansas
Department of Family and Consumer Sciences
Conway, AR 72035

RE: IRB Review of Research

Dear Ms. Crook:

The UCA Institutional Review Board accepts the University of Arkansas for Medical Sciences IRB Approval #72850 of your research project, *Relationship of dietary factors and physical activity to body fat in 3-5 year-old children*. Continuing review will not be required by the UCA IRB. For future reference, the UCA IRB number is 07-058.

Please inform the UCA IRB by memo of any changes made to the research project.

Sincerely,

A handwritten signature in cursive script that reads "Jacquie Rainey".

Jacquie Rainey, Dr.P.H.
Chair, UCA IRB



Institutional Review Board
Office of Research and Sponsored Programs
P.O. Box 425619, Denton, TX 76204-5619
940-898-3375 Fax 940-898-3416
e-mail: IRB@twu.edu

December 7, 2006

Ms. Tina A. Crook
#10 Garnet Court
Little Rock, AR 72212

Dear Ms. Crook:

*Re: Accuracy of Air-displacement Plethysmography and Dual-energy X-ray
Absorptiometry for Estimating Body Fat in Children Age 2 to 5 Years*

The above referenced study has been received and reviewed by the Texas Woman's University Institutional Review Board and has been determined to be exempt from further review because it has been reviewed and approved by an IRB at the University of Arkansas for Medical Sciences in Little Rock, Arkansas. This study will be carried out entirely at the Arkansas Children's Nutrition Center in Little Rock, Arkansas, an affiliate of the University of Arkansas for Medical Sciences.

Another review by the TWU IRB is required if your project changes in any way, and the TWU IRB must be notified immediately regarding any adverse events. If you have any questions, feel free to call the TWU Institutional Review Board at the phone number listed above.

Sincerely,

Dr. David Nichols, Chair
Institutional Review Board - Denton

cc. Dr. Chandan Prasad, Department of Nutrition & Food Sciences
Dr. Carolyn Bednar, Department of Nutrition & Food Sciences
Graduate School

Simply the **BEST**

APPENDIX B

Flyer

**Healthy Children Age 2 to 5 Years
Needed for Research Study**
Arkansas Children's Nutrition Center
is conducting a study measuring body
fat three different ways in children.
Healthy children 2 to 5 years of age who
weighed of at least 5.5 pounds at birth
are needed to participate. Study will
require one full day at Arkansas
Children's Nutrition Center for testing.
Monetary compensation will be offered
for the study visit.

**Study conducted by
Tina Crook, M.S., R.D., L.D.**
**For more information, call: 501-364-3309, 866-
423-1311 (toll free)**
or email: ACNCstudies@uams.edu
**1212 Marshall Street, Little Rock, AR
72202**
Sponsored by: USDA

APPENDIX C
Screening Form

**University of Arkansas for Medical Sciences and Arkansas Children's Nutrition Center
Relationship of Dietary Factors and Physical Activity to Body Fat in 3-5 Year Old Children**

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Study Sponsor: USDA

TOD POD® Study Screening Form

Screen Date: Participant ID: T-_____

Contact Date: Race:

Child's Name: Gender:

Date of Birth: Age:

I. PARENT INFORMATION

Mom's Name: Phone 1:

Dad's Name: Phone 2:

Address: Pediatrician:

How did you hear about this study?

II. MOM'S/CHILD'S MEDICAL HISTORY

Gestational age: Birth weight:

What was child's primary source of nutrition during the first year of life (breast milk, milk-based formula, soy-based formula, other)?

Does the child have any medical diagnoses, past or present? If yes, explain.

Does the child take any routine medications? If yes, list and give duration and frequency.

Does the child take any supplements such as vitamins, minerals or herbal supplements? If yes, list and give duration and frequency.

**University of Arkansas for Medical Sciences and Arkansas Children's Nutrition Center
Relationship of Dietary Factors and Physical Activity to Body Fat in 3-5 Year Old Children
Principal Investigator: Tina Crook, M.S., R.D., L.D.**

Study Sponsor: USDA

TOD POD® Study Screening Form

III. SCREENING SUMMARY

Mom's Answer Correct
Answer

Was this a full-term birth? (≥ 37 - < 42 weeks)?

YES

Did the child weigh at least 5.5 pounds at birth?

YES

Does the child have any significant PMH or current diagnosis affecting growth?

NO

Is the child on any medications that could impact growth and development?

NO

Is the child on any supplements that could impact growth and development?

NO

Is the parent/guardian at least 18 years of age?

YES

Is English the primary language of the parent/guardian?

NO

IV. COMMENTS/APPROVAL

Comments: _____

Is child eligible to participate in study?

PI's Signature _____ Date _____

APPENDIX D

Consent Form

Protocol #: 72850:
Relationship of dietary and physical activity to body fat in 3-5 year-old children
Arkansas Children's Nutrition Center Study
Sponsor: USDA

Consent Form

Project Title: Relationship of Dietary Factors and Physical Activity to Body Fat in 3-5 Year-Old Children

Principal Investigator: Tina Crook, M.S., R.D., L.D., Doctoral Candidate
Office Telephone: 501-364-3755
After Hours: 501-690-6464

Co-Investigators: Janet Gilchrist, Ph.D., R.D.
Thomas Badger, Ph.D.
Patrick Casey, M.D.

Study Coordinator: Stacey Smith, R.D.
Office: (501) 364-5323
Pager: (501) 405-3499

Study Location: Arkansas Children's Nutrition Center
1212 Marshall Street
Little Rock, AR 72202

The following information is provided to tell you about the research study and your participation in the study. Please read this form carefully. Feel free to ask any questions you may have about the study and the information below. You will be given a chance to ask questions and your questions will be answered. You will be given a signed copy of this consent form.

Purpose Of The Research

The purpose of this study is to determine how practical and precise different body composition methods are for measuring body fat and lean tissue in young children and to determine if certain dietary factors and physical activity patterns are related to body fat levels in children.

Sixty (60) children who are between the ages of 3 and 5 years will be recruited for the study. The study will be conducted at the Arkansas Children's Nutrition Center. You are being asked to be a part of this study because your child is healthy and is between 3 – 5 years old.

Explanation of Study Procedures

Only one study visit will be required. It will take about seven hours to complete the testing. All testing must be done on the same day. We will ask you about your child's medical history, medications and diet and some demographic information (date of birth, gender, race, ethnicity) about your child. You will be asked to provide food records of everything your child eats and drinks for three days. A postage-paid envelope will be provided for you to return the food records. You will also be asked to fill out a three-

page physical activity questionnaire about your child during the study visit. We will measure your child's weight and height. We will also attempt to collect two urine samples from your child. We will run three tests on your child to measure his/her body fat. The first test will involve your child drinking about ½ cup of juice or milk with about 1 tablespoon of a tasteless liquid called deuterium mixed into it. The drink is harmless and will pass in your child's urine allowing us to measure body fat. The second test will measure body fat by having your child enter the BodPod® machine for about three minutes. The third test is a DXA scan that will require your child lie completely still on a table for about three minutes. You will be with your child throughout all of the testing procedures.

You will not be billed for any part of this study. At the end of the study visit, if all tests are complete, we will give you a \$200 Visa® card. You will be partially compensated if the study is only partially completed.

Potential Risks Associated With Participating In This Study

There are no expected risks to your child for being in this study. If a self-adhesive urine collection bag is used on your child, there is a slight risk of skin irritation. If your child is toilet trained, use of this bag may not be necessary. Deuterium has been used for many years in infants, children and adults without any reported side effects. It does not contain any harmful substances. The BodPod® and DXA machines are very safe research devices. The BodPod® measures weight on a scale and body volume by how much air your child displaces inside the machine's chamber. In past studies, a small number of participants have felt anxious or claustrophobic inside the BodPod®. The BodPod® has a large window the child will be able to see through to minimize these feelings. You will be able to see your child at all times. If you or your child becomes uncomfortable, the test will be stopped. The DXA machine uses radiation like an x-ray, but the levels are approximately ten times lower than would be received from a chest x-ray.

Potential Benefits Associated With Participating In This Study

Your child will receive no direct health benefits for being in the study. At the end of the study, we will provide you with a copy of your child's body composition (% body fat) reports. In the future, other children may benefit from this research, as this study may help determine better ways of measuring body composition in young children.

Alternatives to Participating

You may choose not to have your child participate in the study. Your child's pediatrician would normally monitor his/her growth at office visits by taking weight and height measures. Body fat and lean tissue would not normally be measured at these visits.

Private Disclosure of Participating In This Study

Any personal facts collected during this study will be kept private. The results of this study may be published in a medical journal, but your child will not be identified by name. Representatives of the Institutional Review Board (IRB) at the University of Arkansas for Medical Sciences (UAMS), the Office of Human Research Protections (OHRP), the Arkansas Children's Hospital Research Institute (ACHRI) or the United States Department of Agriculture (USDA) may be given access to research study

records and pertinent health records that may contain your child's name or other identifying facts. Some of our personnel are required by law to report any suspected child abuse and/or neglect to the proper authorities. Our staff members are trained professionals and realize that children often sustain injuries through their normal daily activities and these do not constitute abuse. Please feel free to ask our staff for clarification on these issues.

Payment for Study-Related Injury

The Principal Investigator and this institution have made no provision to reimburse you for the cost of medical care beyond emergency medical treatment or to pay for any lost wages, pain and suffering, hospitalization, or other expenses your child may incur as the result of any such complication, injury or illness.

Questions about This Study

If you have any questions during this study about the research, you should contact the Principal Investigator, Tina Crook, at 501-364-3755 (office) or 501-690-6464 (after hours). You may call the Institutional Review Board (IRB) at (501) 686-5667 regarding a research-related injury, with questions about your rights as a research participant or to discuss any problems or concerns about the research. Also, you may call the IRB number if you are unable to reach the investigator or you wish to speak to someone not directly related to the study.

Right to Refuse Or Withdraw From The Study

Your child's participation in this study is voluntary. You may choose to withdraw permission to continue with this study at any time. If you do, all study related procedures will stop immediately. A decision to refuse to be a part in this study or to withdraw from the study will not affect your right to present and/or future health care.

Reasons You May Not Be Able To Stay In the Study

The people doing this study may remove your child from this study without your consent. Usual reasons for the investigator to remove a child from the study are:

- The child's condition changes in a way that would expose your child to undue risk or make study procedures difficult or impossible to complete.
- The child does not comply with study-related procedures.
- The study is halted for any reason.

Consent

I have read the above statement and have been able to ask questions and express concerns, which have been satisfactorily responded to by the investigator(s)/study staff. I understand the purpose of the study as well as the potential benefits and risks that are involved. I hereby give my informed and free consent for my child to be a participant in this study. I have been given a copy of this consent form.

You have not waived any legal right to which you are otherwise entitled by signing this form.

Signature of Parent/Legal Guardian Date/Time

Signature of Person Obtaining Consent Date/Time

Signature of Witness Date/Time

Statement of the Investigator

Any questions expressed by the subject/parent/guardian have been answered.

Signature of Principal Investigator Date/Time

APPENDIX E

HIPAA Form

Protocol # 72850:

Relationship of dietary factors and physical activity to body fat in 3-5 year-old children

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Study Sponsor: USDA

HIPAA Research Authorization

STUDY TITLE: Accuracy of Air-Displacement Plethysmography and Dual-Energy X-Ray Absorptiometry for Estimating Body Fat in Children 2 to 5 Years Old

PRINCIPAL INVESTIGATOR:

Tina Crook, M.S., R.D., L.D.

USDA/ARS and Arkansas Children's Nutrition Center

1212 Marshall Street, Rm. N-1052

Little Rock, AR 72202

Office Telephone: 501-364-3755

After Hours: 501-690-6464

STUDY SPONSOR: USDA

This form and the research consent form need to be kept together.

We are asking you to take part in the research study described in the consent form. To do this research, we need to collect health information about your child. We will only collect information that is needed for the research. Being part of this research study will create new health information including your child's growth and body composition. For your child to be in this research study, we need your permission to collect, create and share this information.

We may share your child's health information with people at the University of Arkansas for Medical Sciences (UAMS) who help with the research process, such as the Institutional Review Board (IRB) and the Office of Research Compliance (ORC). We may share your child's information with the following researchers outside of UAMS who are supervising Ms. Crook's dissertation work: University of Central Arkansas (UCA) and Texas Women's University (TWU). We may also share your child's information with companies that pay for part of the research, such as the United States Department of Agriculture and Life Measurement, Inc., or their legally authorized representative, or anyone who might purchase those companies at a later date. Additionally, we may need to share your child's health information with people outside of UAMS who make sure we do the research properly, such as the Office of Human Research Protections. We believe that those involved with research understand the importance of keeping your health information private. However, some of the people outside of UAMS may share your health information with someone else. If they do, the same laws that UAMS must obey may not apply to others to protect your child's health information.

Your permission to collect, use and share your child's health information expires at the end of the research study.

If you sign this form, you are giving us permission to create, collect, use and share your child's health information as described in this form. You do not have to sign this form. However, if you decide not to sign this form, you cannot be in the research study. You need to sign this form and the research consent form if you want your child to be in the research study. We cannot do the research if we cannot collect, use and share your child's health information.

If you sign this form but decide later that you no longer want us to collect or share your child's health information, you must send a letter to Tina Crook at the address listed on the first page of this form. The letter needs to be signed by you, should list the "Study Title" listed on this form and should state that you have changed your mind and that you are revoking your "HIPAA Research Authorization" for your child. Your child will not be able to continue in the research study if we cannot collect and share any more health information. We may still use and share any information collected between the time you signed this form and when we received your letter saying you no longer want us to use your information.

If you decide not to sign this form or change your mind later, this will not affect your current or future medical care at UAMS or Arkansas Children's Hospital.

SIGNATURE, DATE, AND IDENTITY OF PERSON SIGNING

The health information about _____
can be collected and used by the researchers and staff for the research study described in this form and the research consent form.

Signature: _____

Print name: _____

Relationship to participant: _____

Date _____ Time _____

The researcher will give you a signed copy of this form.

APPENDIX F
Case Report Form

Protocol # 72850: *Relationship of dietary factors and physical activity to body fat in 3-5 year-old children*

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Arkansas Children's Nutrition Center

Study Sponsor: USDA

TOD POD —Study Visit Form

Participant ID # T - _____ Visit Date: _____

DOB: _____ Age (Year): 3 4 5

Demographic Information	Obtained by (initials) _____
Parents' Names:	Phone numbers (list at least 2):
Address:	Child's Name:
Child's Date of Birth:	Child's Gender (circle one): Male Female
Child's Ethnicity Based on Maternal Lineage (circle one): Caucasian African American Hispanic Other If other, list:	

Nutrition History	Obtained by (initials) _____
Did mom have any complications during pregnancy? (circle one) Yes No If yes, describe:	Child's Gestation Age:
What was the child's primary source of nutrition the first year of life? (circle one) Breast Milk Milk-Based Formula Soy-Based Formula Other If other, list:	Child's Birth Weight:

Medical History Questionnaire	Circle the correct response. Comment, when applicable.
Has your child ever been hospitalized? Obtained by (initials) _____	No Yes If yes, for what and when?
Has your child ever been seen in a growth clinic?	No Yes If yes, for what and when?
Has your child ever been diagnosed with a growth disorder?	No Yes If yes, what was the diagnosis and when?
Has your child ever been diagnosed with failure to thrive?	No Yes If yes, when?

Protocol # 72850: *Relationship of dietary factors and physical activity to body fat in 3-5 year-old children*

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Arkansas Children's Nutrition Center

Study Sponsor: USDA

TOD POD —Study Visit Form

Participant ID # T - _____ Visit Date: _____

DOB: _____ Age (Year): 3 4 5

Has your child ever been diagnosed with ADD or ADHD?	No Yes If yes, when?
Has your child ever been diagnosed with any of the following medical conditions or diseases?	Diabetes Cystic Fibrosis Hypothyroidism Hyperthyroidism Hypoglycemia (low blood sugar) Hyperglycemia (high blood sugar) Asthma Pulmonary Disease Bone Disease
Has your child ever been diagnosed with any medical condition or disease not listed above?	No Yes If yes, what and when?
Does your child have any food allergies?	No Yes If yes, what?
Is your child able to go one hour without eating or drinking?	No Yes

Medication Use Questionnaire		Initials _____
D=Daily F=Frequent (as needed) O=Occasional (monthly or less) I=Infrequent N=Never		
ADD or ADHD Meds	D F O I N	Name, dose
ADD or ADHD Meds	D F O I N	Name, dose
Asthma Meds	D F O I N	Name, dose
Asthma Meds	D F O I N	Name, dose
Corticosteroids	D F O I N	Name, dose
Dietary Supplements	D F O I N	Name, dose
Herbals	D F O I N	Name, dose
Vitamins	D F O I N	Name, dose
Minerals	D F O I N	Name, dose
Other	D F O I N	Name, dose
Other	D F O I N	Name, dose

Protocol # 72850: *Relationship of dietary factors and physical activity to body fat in 3-5 year-old children*

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Arkansas Children's Nutrition Center

Study Sponsor: USDA

TOD POD —Study Visit Form

Participant ID # T - _____ Visit Date: _____

DOB: _____ Age (Year): 3 4 5

Food Records			Initials _____
Given (circle one)	Yes	No	
Returned (circle one)	Yes	No	
Comments:			

Netherlands Physical Activity Questionnaire			Initials _____
Given (circle one)	Yes	No	
Returned (circle one)	Yes	No	
Comments:			

Anthropometric Measures					Initials _____
Weight	(kg)	(kg)	(kg)	Avg. (kg)	%
Stature	(cm)	(cm)	(cm)	Avg. (cm)	%
BMI					%
Comments:					

Protocol # 72850: Relationship of dietary factors and physical activity to body fat in 3-5 year-old children

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Arkansas Children's Nutrition Center

Study Sponsor: USDA

TOD POD —Study Visit Form

Participant ID # T - _____ Visit Date: _____

DOB: _____ Age (Year): 3 4 5

Deuterium Procedure	Initials _____
Dose Calculation: Child's weight _____ (kg) X 100 mg = _____ mg	
Baseline urine collection: Time collected _____ Amount _____ (cc) Time processed _____ Initials _____	
Comments:	
Time beverage was consumed (immediately after baseline urine): _____ Amount of beverage consumed: _____ Beverage type (circle one): milk juice Comments:	
2 nd urine collection (≥ 5 hours later): Time collected _____ Amount _____ (cc) Time processed _____ Initials _____ Comments:	

Tod Pod Procedure	Initials _____		
Record the following from the computer screen after the volume measurements. Important: You will not be able to go back and retrieve this information once you press enter!			
Model _____	Reading 1 _____	Reading 2 _____	Reading 3 _____
Best Mean _____	Weight (kg) _____	Volume _____	Area Artifact _____
LV _____	Body Volume _____	Density _____	Percent BF _____
Fat Weight _____	Lean Weight _____		
Tod Pod completed (circle one)?		Yes	No
Excessive movement (circle one)?		Yes	No
Child cried (circle one)?		Yes	No
Comments:			

Protocol # 72850: *Relationship of dietary factors and physical activity to body fat in 3-5 year-old children*

Principal Investigator: Tina Crook, M.S., R.D., L.D.

Arkansas Children's Nutrition Center

Study Sponsor: USDA

TOD POD —Study Visit Form

Participant ID # T - _____ Visit Date: _____
DOB: _____ Age (Year): 3 4 5

DXA Procedure	Initials _____		
DXA completed (circle one)?	Yes	No	
Report in chart (circle one)?	Yes	No	
Limbs under (circle one)?	Yes	No	
Blanket (circle one)?	Yes	No	
Pacifier (circle one)?	Yes	No	
Urine bag (circle one)?	Yes	No	
Earrings/other metals (circle one)?	Yes	No	
Movement (circle one)?	Yes	No	
Position (circle one):	Prone	Supine	Side
Report ID #: _____			
Percent body Fat: _____%			
Comments:			

Visit Start Time: _____

Visit End Time: _____

Data entry completed by (initials): _____ Date: _____

Data entry verified by (initials): _____ Date: _____

APPENDIX G
Food Record Form

APPENDIX H
Food Portion Visual™

**Food Amounts Booklet
Conversion Guide for Dietary Interviewers**

When the participant shows a picture in the Food Amounts Booklet, convert to NDS-R amounts per the following information.

Squares and Rectangles 1/4 inch grid (pg 1)	Circles (pg 2)		
Each square is 1/4" x 1/4". Select shape, cube (3 dimensions) or rectangle (2 dimensions) per NDS-R. Enter as fractions, e.g., 2/4 wide x 13/4 long x 7/4 high.	Select shape, circle or sphere. Enter diameter in inches (") per the following:		
	A = 1 inch B = 2 inches C = 2 1/2 inches	D = 3 inches E = 4 inches F = 5 inches	G = 6 inches H = 7 inches

Wedges (pg 3)						
Radius (R)	Length of Arc (width of rounded edge)					
Select shape, wedge, enter radius:		A	B	C	D	E
1 = 4" radius (8" D) ¹	1	0.6"	1.1"	2.1"	2.6"	3.9"
2 = 4 1/2" radius (9" D)	2	0.7"	1.3"	2.4"	2.9"	4.4"
3 = 6" radius (12" D)	3	0.9"	1.6"	3.1"	3.9"	5.9"
4 = 8" radius (16" D)	4	1.1"	2.1"	4.1"	5.1"	7.8"
5 = 9" radius (18" D)	5	1.3"	2.4"	4.7"	5.8"	8.9"

For 3D² wedge, also use Thickness (pg 4, height=x/16) or Squares and Rectangles (pg 1, height=x/4)

Thickness (pg 4)	Measuring Spoons (pg 5)	Eating and Serving Spoons (pg 6)	Measuring Cups (pg 7)
Each unit is 1/16" thick. Enter thickness as fraction. E.g.: 1 = 1/16" 2 = 2/16" 3 = 3/16" 18 = 18/16" 40 = 40/16"	<u>Standard measures</u> 1/2 teaspoon (TS) 1 teaspoon 1/2 tablespoon (TB) 1 tablespoon	<u>Teaspoons:</u> Level = 1 TS Heaping = 2 TS <u>Tablespoon:</u> Level = 1 TB Heaping = 2 TB	Standard measures 1/4 cup (CP) 1/3 CP 1/2 CP 1 CP

Glasses (pg 8-9) (Fluid Ounces, FO)			
1 A = 1 1/4 FO	2 A = 2 1/2 FO	3 A = 3 FO	4 A = 8 FO (1 CP)
1 B = 2 1/2 FO	2 B = 5 FO	3 B = 6 FO	4 B = 16 FO (2 CP)
1 C = 3 3/4 FO	2 C = 7 1/2 FO	3 C = 9 FO	4 C = 24 FO (3 CP)
1 D = 5 FO	2 D = 10 FO	3 D = 12 FO	4 D = 32 FO (4 CP)

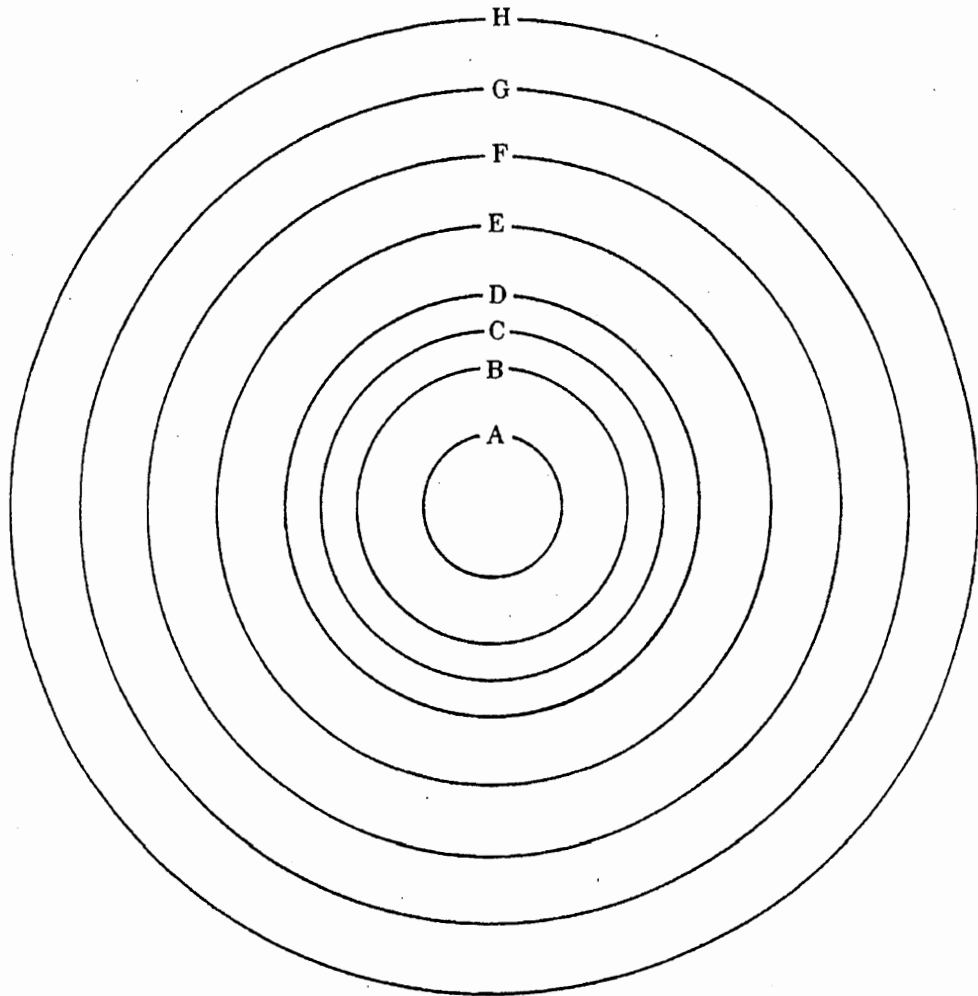
Bowls (pg 10-11)			
1 A = 1/8 CP	2 A = 3/8 CP	3 A = 3/4 CP	4 A = 2 CP
1 B = 1/4 CP	2 B = 3/4 CP	3 B = 1 1/2 CP	4 B = 4 CP
1 C = 3/8 CP	2 C = 1 1/4 CP	3 C = 2 1/4 CP	4 C = 6 CP
1 D = 1/2 CP	2 D = 1 1/2 CP	3 D = 3 CP	4 D = 8 CP

Mounds (pg 12-13)	Meats (pg 14) (Ounces, OZ)	Chicken (pg 15) (Medium, MD)	Fish (pg 16)
1 = 1 CP	All are edible portion	Edible portion	Edible portion
2 = 3/4 CP	1 = 3 OZ	Wing = 1 MD	1 = 2 OZ
3 = 1/2 CP	2 = 3 OZ	Drumstick = 1 MD	2 = 3 OZ
4 = 1/3 CP	3 = 1 1/2 OZ	Thigh = 1 MD	3 = 1/2 OZ
5 = 1/4 CP		Breast = 1 MD	4 = 1 OZ

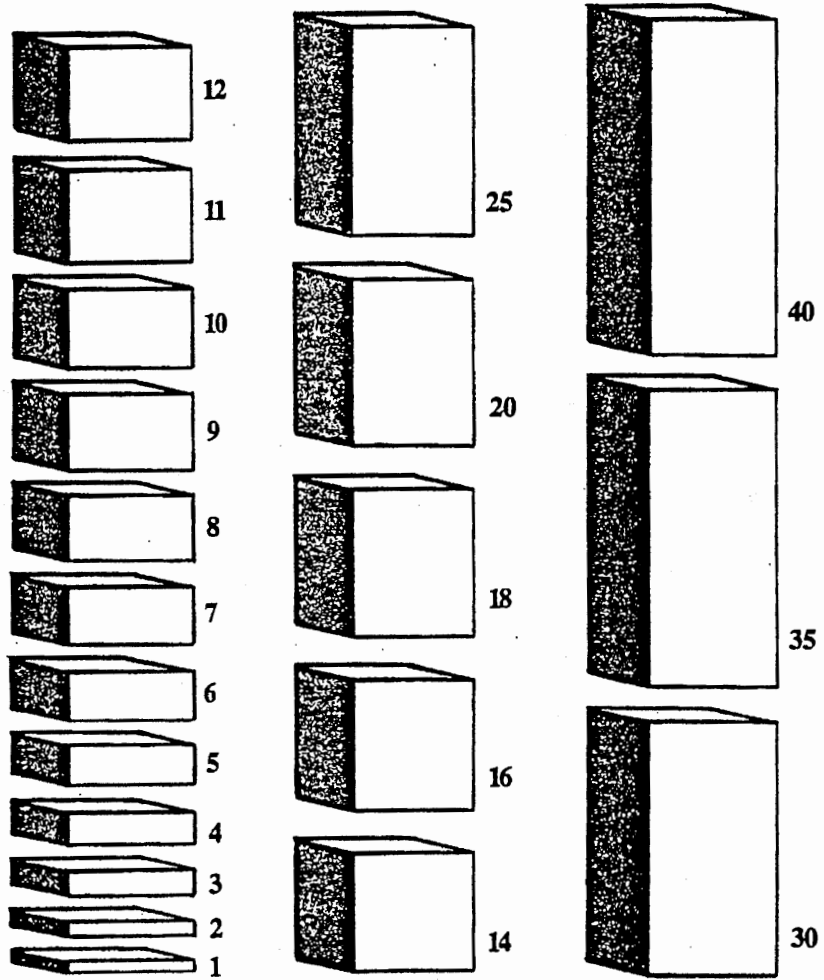
¹ D = Diameter

² 3D = three dimensional

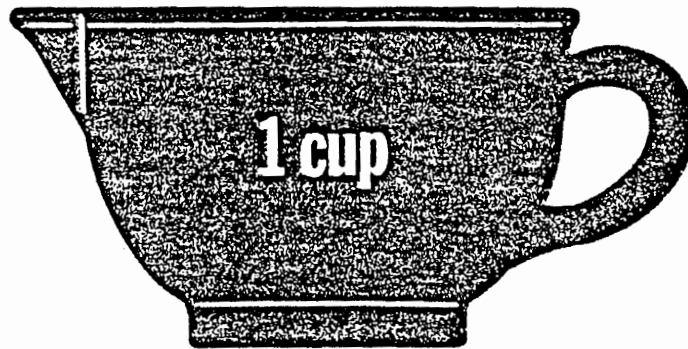
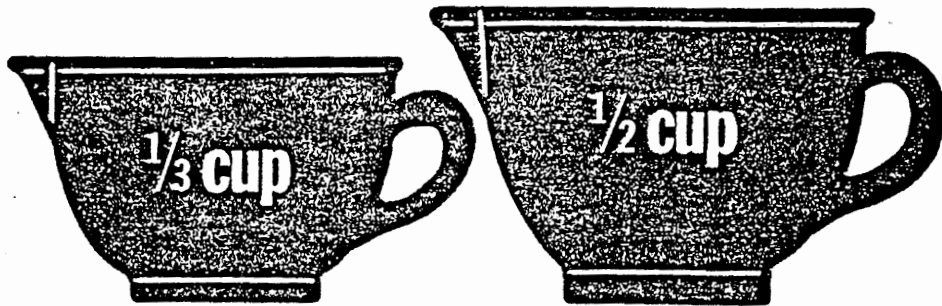
Circles



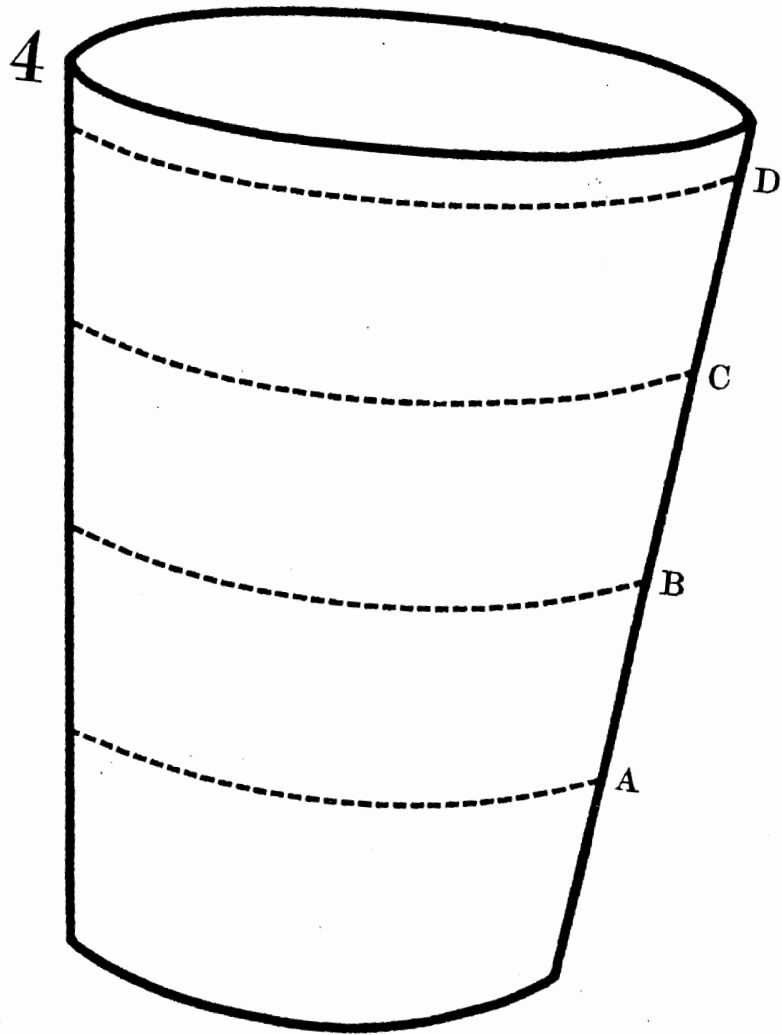
Thickness

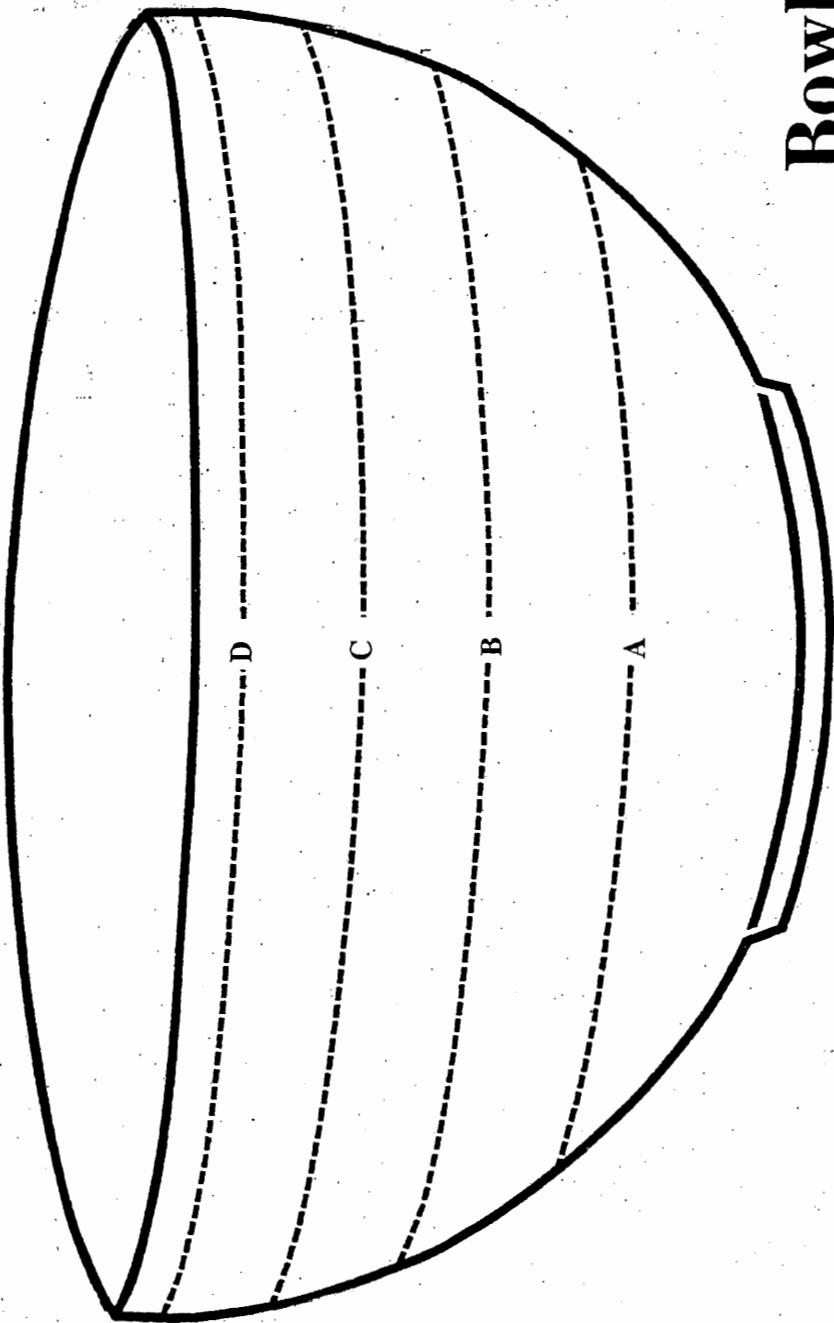


Measuring Cups



Glasses

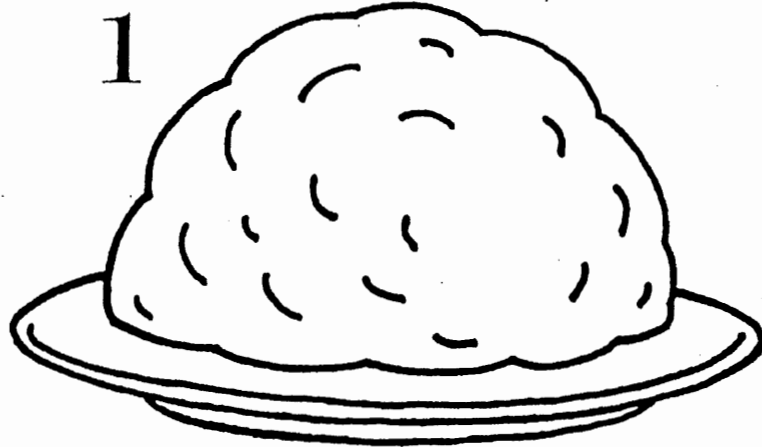




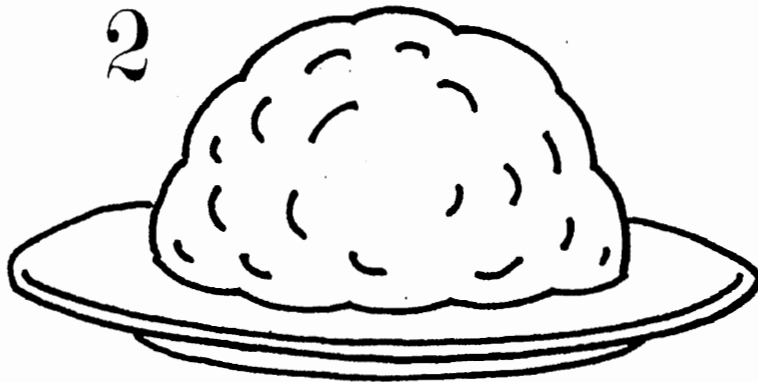
Bowls

Mounds

1

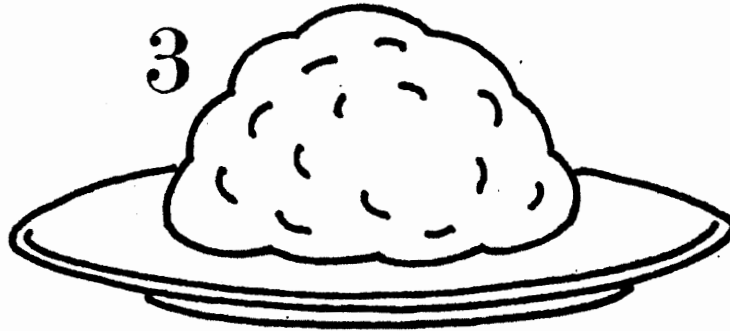


2

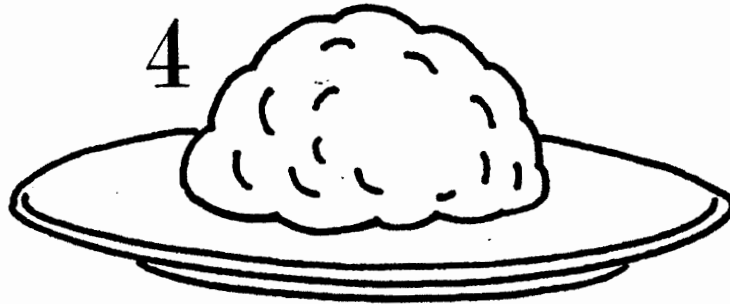


Mounds

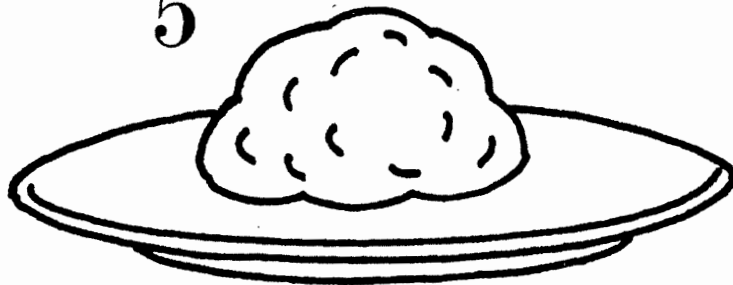
3



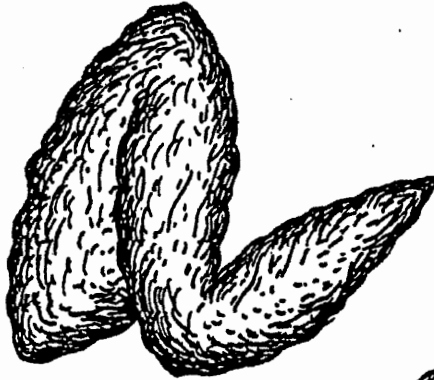
4



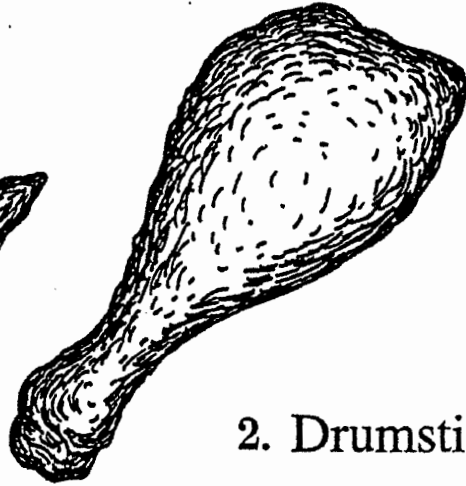
5



Chicken



1. Wing



2. Drumstick



3. Thigh



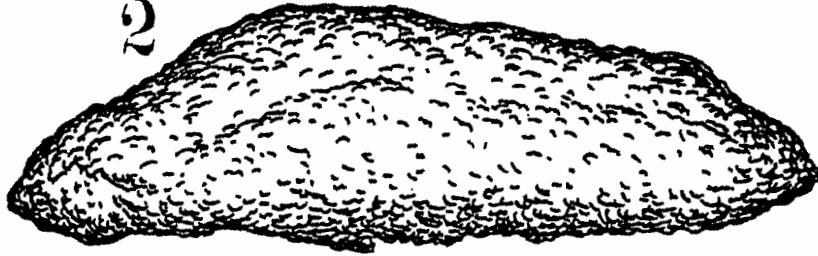
4. Breast

Fish

1



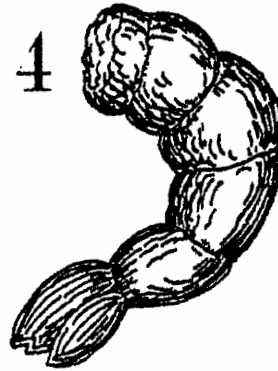
2



3



4



APPENDIX I

The Healthy Eating Index, 2005

Healthy Eating Index—2005

USDA

Center for
Nutrition Policy
and Promotion

3191 Park Center Drive
Alexandria, VA 22302

Voice: 703-305-7600
Fax: 703-305-3300

www.cnpp.usda.gov

CNPP
Fact Sheet No. 1

December 2005
Slightly Revised
June 2008

USDA is an equal
opportunity provider
and employer.

The **HEALTHY EATING INDEX (HEI)** is a measure of diet quality that assesses conformance to Federal dietary guidance. The original HEI was created by the U.S. Department of Agriculture (USDA) in 1995. Release of new Dietary Guidelines for Americans in 2005 motivated a revision of the HEI. The food group standards are based on the recommendations found in MyPyramid (see Britten *et al.*, *Journal of Nutrition Education and Behavior* 38(6S) S78-S92). The standards were created using a density approach, that is, they are expressed as a percent of calories or per 1,000 calories. The components of the HEI-2005 and the scoring standards are shown below.

Healthy Eating Index—2005 components and standards for scoring¹

Component	Maximum points	Standard for maximum score	Standard for minimum score of zero
Total Fruit (includes 100% juice)	5	≥0.8 cup equiv. per 1,000 kcal	No Fruit
Whole Fruit (not juice)	5	≥0.4 cup equiv. per 1,000 kcal	No Whole Fruit
Total Vegetables	5	≥1.1 cup equiv. per 1,000 kcal	No Vegetables
Dark Green and Orange Vegetables and Legumes ²	5	≥0.4 cup equiv. per 1,000 kcal	No Dark Green or Orange Vegetables or Legumes
Total Grains	5	≥3.0 oz equiv. per 1,000 kcal	No Grains
Whole Grains	5	≥1.5 oz equiv. per 1,000 kcal	No Whole Grains
Milk ³	10	≥1.3 cup equiv. per 1,000 kcal	No Milk
Meat and Beans	10	≥2.5 oz equiv. per 1,000 kcal	No Meat or Beans
Oils ⁴	10	≥12 grams per 1,000 kcal	No Oil
Saturated Fat	10	≤7% of energy ⁵	≥15% of energy
Sodium	10	≤5.7 gram per 1,000 kcal ⁶	≥2.0 grams per 1,000 kcal
Calories from Solid Fats, Alcoholic beverages, and Added Sugars (SoFAAS)	20	≤20% of energy	≥50% of energy

¹Intakes between the minimum and maximum levels are scored proportionately, except for Saturated Fat and Sodium (see note 5).

²Legumes counted as vegetables only after Meat and Beans standard is met.

³Includes all milk products, such as fluid milk, yogurt, and cheese, and soy beverages.

⁴Includes nonhydrogenated vegetable oils and oils in fish, nuts, and seeds.

⁵Saturated Fat and Sodium get a score of 8 for the intake levels that reflect the 2005 Dietary Guidelines, - 10% of calories from saturated fat and 1.1 grams of sodium/1,000 kcal, respectively.

Using data from the National Health and Nutrition Examination Survey, 2001-2002, a psychometric evaluation found the HEI-2005 to satisfy several types of validity tests. Reliability analyses suggest that the individual components provide additional insight to that of the summary score. The HEI-2005 is a standardized tool that can be used in nutrition monitoring, interventions, and research. Further details on the development and evaluation of the HEI-2005 and population scores are available at www.cnpp.usda.gov/HealthyEatingIndex.htm

Authors: Patricia M. Guenther,¹ Susan M. Krebs-Smith,² Jill Reedy,² Patricia Britten,¹ Wen-Yu Juan,¹ Mark Liao,¹ Andrea Carlson,¹ Hazel A. Hiza,¹ and P. Peter Basiaotis.¹

¹USDA Center for Nutrition Policy and Promotion and ²National Cancer Institute

APPENDIX J

Netherlands Physical Activity Questionnaire

PHYSICAL ACTIVITY QUESTIONNAIRE

PAGE 1

QID: 02

Subject ID:

Date: 20
 Month Day Year



Instructions: Please circle the number that **best describes your child during the past six months**. For example, if in the past six months, your child preferred to play alone as often as he/she preferred to play with other children, circle the number three for the first question. On the other hand, if he or she almost always preferred playing with other children, rather than alone, circle the number five.

	Almost Always ↙			About Equal ↓			Almost Always ↘	
1. Prefers to play alone	1	2	3	4	5	Prefers to play with other children		
2. Prefers vigorous games (e.g., tag, kickball)	1	2	3	4	5	Prefers quiet games (e.g., board games)		
3. Dislikes playing sports (e.g., soccer, basketball)	1	2	3	4	5	Likes playing sports		
4. Is more introverted (e.g., quiet, reserved)	1	2	3	4	5	Is more extroverted (e.g., outgoing)		
5. Likes to read	1	2	3	4	5	Dislikes to read		
6. Likes to play outside	1	2	3	4	5	Likes to play inside (home/school)		
7. Less physically active compared to other children of same age	1	2	3	4	5	More physically active compared to other children of same age		

Instructions: Please answer the following questions as they relate to your child's **usual daily routine during the past six months**. Estimate the time to the nearest 1/4 hour (15 minutes) per day.

8. On average, how many hours per day does your child spend watching any type of television including video movies?

_____ hours per day

PHYSICAL ACTIVITY QUESTIONNAIRE

PAGE 2

9. On average, how many hours per day does your child spend playing video games (such as Nintendo®) and/or computer games?

_____ hours per day

10. On average, how many hours per night does your child spend sleeping? (Do not include naps.)

_____ hours per night

11. On average, how many hours per day does your child sleep during naps?

_____ hours per day

Please list the **two play- or sport-related physical activities** which your child did most often during the past six months (e.g., kickball, board games, biking, soccer, puzzles, playing on playground equipment, roller blading, swimming, rope jumping):

12. _____

13. _____

14. During the past six months, did your child participate in or take lessons in any of the following **organized sports**? (Check all that apply.)

_____ Swim lessons/swim club _____ Youth soccer _____ Basketball league/camp

_____ T-ball/baseball/softball _____ Gymnastics/tumbling _____ Dance/ballet/jazz/aerobic

_____ Hockey/ice/roller/indoor _____ Tennis/racquetball _____ Track & field/running

_____ Football league/camp _____ Horseback riding _____ Volleyball league/camp

_____ None Others (Please list.) _____

15. When in school, how often does your child participate in physical education (PE)?

_____ daily _____ 2-4 times/week _____ once/week _____ does not participate _____ don't know

16. What arm does your child prefer to throw with?

_____ right _____ left _____ no preference _____ don't know

17. What leg does your child prefer to kick with?

___ right ___ left ___ no preference ___ don't know

Thank you for taking the time to complete this physical activity questionnaire.

Instructions for Scoring

Page one (the seven question likert scale) will be treated two ways: individual questions as markers of activity and a composite score.

The individual questions will be scored exactly as they are so in the end, I would expect a very active child to have a high number for question 6 and a low number for question 2.

To create the composite score do the following: Sum questions 1, 3, 4, 5, and 7. Reverse order 2 and 6 and then sum them with the other 5 questions. I.e., 2 and 6 are worded to avoid response set bias, so that if a parent circles 2 for question 2. We score the response as 4 when creating the composite score. If parent circles 1 for question 6, we score as 5 for composite. The highest summed score (after reversing 2 and 6) would be 35. The equation is: Physical Activity Score = $(\Sigma 7 \text{ Questions} \div 35) \times 100$. For example, a sum of 20 would produce a physical activity score of 57.

Questions on sedentary behavior (TV, video, nap) are treated as individual items.

On the back side of the questionnaire, the Activity 1 and Activity 2 listings of what kids do will eventually be collapsed into categories. By continuing with the current format for a few more rounds (i.e., data entry listing everything) I'll be able to construct meaningful groupings of the individual activities. I anticipate being able to categorize the activities within a few months.

Throwing and kicking may be used for segmented bone analysis (dominant vs non dominant).

APPENDIX K

DXA Approval from Arkansas Department of Health and Human Services



Arkansas Department of Health and Human Services



Division of Health

Paul K. Halverson, DrPH, Director

P.O. Box 1437, Slot H-30

Little Rock, AR 72203-1437

• 501-661-2173

• TDD: 1-800-234-4390

January 3, 2006

Mr. Todd Frost, BS, MA, RSO
ACH, Radiology Dept.
Dr. Janet Gilchrist, Ph.D, RD
Nutrition Center
Little Rock, AR 72202

Registration Number: **HS-0146**
Date of Inspection: December 14, 2005
Inspectors: Rick Kelley and
Wayne Wright

Dear Mr. Frost and Dr. Gilchrist:

This document is in reference to your "Health Screening" study program at the Nutrition Center of ACH. We inspected your equipment, documentation, and procedures for the Hologic Bone Density machine used in the body composition research.

Please mention your "Registration Number", listed above, in any communication with this agency. After reviewing the report of the inspection conducted by the above inspector on the date shown, we are pleased to advise you that your radiation protection program appears to be in compliance with the applicable Rules and Regulations for Control of Sources of Ionizing Radiation with regard to records reviewed and procedures observed.

Thank you for your cooperation and assistance during the inspection.

If we can be of further assistance with your radiation safety program, please call me at (501) 661-2378. Please address any correspondence to me at Mail Slot #30.

Sincerely,

Rick Kelley, Program Leader
X-ray Section
Division of Radiation Control
And Emergency Management

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APPENDIX L

DXA Approval from UAMS Radiation Safety Committee

OCCUPATIONAL HEALTH AND SAFETY

4301 West Markham, #617
Little Rock, AR 72205-7199

501-686-5536
501-296-1339 (fax)

www.uams.edu/safety

Carol J. Price, M.S.
Director/RSO



Date: October 12, 2006

To: Tina Crook, M.S., R.D., L.D., Doctoral Candidate
Arkansas children's Nutrition Center

From: Carol Price, RSO *CP*
Occupational Health & Safety

RE: Protocol: Accuracy of Air-Displacement Plethysmography and Dual-Energy X-Ray
Absorptiometry for Estimating Body Fat in Children 2 to 5 Years Old

The Radiation Safety Office has reviewed your protocol "Accuracy of Air-Displacement Plethysmography and Dual-Energy X-Ray Absorptiometry for Estimating Body Fat in Children 2 to 5 Years Old". The Radiation Safety Committee grants approval for the above named study.

If you have questions concerning this approval, please contact the Safety Office at 686-5299.