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Abstract

The first two years of a child's life are a particularly critical time period for obesity prevention. As such, an increasing amount of research across the world is aimed at understanding factors that impact early childhood obesity and developing interventions that target these factors effectively. With this growing interest, new and interdisciplinary research teams are developing to meet this research need. Due to rapid growth velocity during this phase of the lifespan, typical assessments used in older populations may not be valid or applicable in infants, and investigators need to be aware of the pros and cons of specific methodological strategies. Therefore, this paper provides an overview of methodology available to assess obesity-related factors in the areas of anthropometry and body composition, nutrient intake, and energy expenditure, in infants ages 0-2 years. Gold standard measures for body composition, such as DXA or other imaging techniques are costly, require highly trained personnel, and are limited for research application. Nutrient intake methodology primarily includes surveys and questionnaires completed via parent proxy report. In terms of energy expenditure, methods of calorimetry are expensive and may not differentiate between different activities. Questionnaires or physical activity sensors offer another way of energy expenditure assessment. However, questionnaires have certain recall bias while the sensors require further validation. Overall, in addition to understanding the pros and cons of each assessment tool, researchers should take into consideration the experience of the interdisciplinary team of investigators, as well as the cost and availability of measures at their institution.

Keywords

Infant, obesity, overweight, body composition, nutrition, energy expenditure, physical activity

Introduction

Globally, over 42 million children under the age of 5 are classified as overweight or obese (World Health Organization (WHO), 2016a). The first two years of a child's life are a particularly important time period for obesity prevention, as risk factors during this time, such as rapid increases in body weight, are linked with a greater propensity for overweight and obesity later in childhood (WHO, 2016a; Blake-Lamb et al., 2016; De Kroon et al., 2010; Flores and Lin, 2013). As childhood overweight is associated with an increased risk of obesity in adulthood (Biro and Wien, 2010), overweight infants are not only at an increased risk of remaining overweight but also more likely to suffer from numerous physical (e.g., type 2 diabetes, coronary heart disease) and psychological (e.g., depression, anxiety) comorbidities as well as an overall lower quality of life (Centers for Disease Control and Prevention (CDC), 2015a). As the economic burden of obesity is growing for both individuals and countries as a whole with total direct and indirect costs ranging from \$2-\$73 billion (Dee et al., 2014), it is prudent to begin obesity prevention as early in life as possible. In response to the recognition of the importance of early childhood development, researchers across the world have begun to develop interventions that effectively prevent obesity.

Because obesity is a direct result of a chronically positive energy balance, obesity-related interventions for all age groups are primarily aimed at reducing energy intake and/or increasing energy expenditure (Biro and Wien, 2010). However, assessing energy balance is challenging in children, as one must account for the energy required for the growth process over time. Further, as children are expected to grow and gain weight, it can be particularly complex to differentiate between healthy and pathological weight gain (Butte et al., 2007; Hall et al., 2013). In addition to rapid changes in body size, composition, and metabolism, the first two years provide an additional challenge to assessment because methods typically used in older populations are not suitable for infants due to their small body size, limited communication abilities, and reliance on others for mobility. Therefore, it is imperative to ensure that assessment tools utilized for energy intake, energy expenditure, and related impact on body composition are precise and accurate in this demographic.

Of the few published studies on obesity prevention in children 0-2 years of age, energy intake (e.g., feeding practices) has been the primary intervention focus, and studies have largely targeted infant feeding practices including parent knowledge and attitudes, as well as understanding infant feeding cues (i.e., signs of satiety) (Ciampa et al., 2010; Paul et al., 2011; Wen et al., 2012). These studies have shown the potential to positively impact obesity; however more rigorous studies in a variety of settings and countries are needed to establish a direct link to infant obesity (Ciampa et al., 2010).

While targeting energy intake has been more dominant in the scant literature, few studies have targeted energy expenditure in infants through promotion of physical activity to parents (Campbell et al., 2008; Cardon et al., 2011). Only a handful of researchers have assessed physical activity in infants themselves and how it relates to obesity as well as ideal growth and development (Li et al., 1995; Tennefors et al., 2003; Ulrich and Hauck, 2013). Additionally, the methodology used to assess infants' body composition to better understand the relationship between body composition and healthy behaviors (e.g., physical activity, nutrient intake) varies considerably in this population (Demerath and Fields, 2014).

Given the importance of the early years to obesity prevention, there is a growing demand for interventions that promote a healthy body composition through reducing energy intake and/or increasing energy expenditure by means of physical activity. With this growing demand, there is a critical need for research focusing on body composition, nutrient intake, and energy expenditure in this age group. In addition, interdisciplinary teams (i.e., teams with two or more disciplines/areas of expertise) that are needed to develop a comprehensive approach to obesity require reliable and suitable methods to assess these parameters. While numerous instruments and technologies exist, an overview of measurement tools for obesity-related parameters has not been addressed for this population of interest. Providing a broad overview of assessment tools can help researchers as well as clinicians identify which tools may be appropriate for their study and provide an avenue to inform interdisciplinary partners about the assessment tools available. Therefore, the purpose of this paper is to provide a broad overview of the assessment methods available for use in obesity-related research in full-term infants age 0-2.

Most obesity-related methodology research along with a large obesity measures registry has focused on ages 2 and above (National Collaborative on Childhood Obesity Research, 2016). Further, the amount of well-child visits are particularly concentrated in the first two years (American Academy of Pediatrics, nd). Thus, while the infant period typically refers to the first 12 months of life, to fill a gap related to lack of obesity-related assessments, we will refer to the age group of 0-2 years as infants. Each section includes a brief overview of the methodology as well as challenges and limitations associated with each measure. Additionally, each method specifies whether it is appropriate for a laboratory setting, which requires access to laboratory and/or high-end equipment, or field setting, which can be performed in a variety of settings such as the clinic or home, in order to provide researchers initial information on the applicability of each assessment.

Anthropometry and body composition

There is a broad range of methods available for the assessment of body composition, ranging from practical methods suited for in-field measurements to highly sophisticated techniques only applicable in a well-controlled laboratory setting. Further, some methods involve invasive procedures. The choice of a method is largely dependent on the compartments of the body (muscle, fat or bone tissue) to be studied as well as the budget and the availability of the study population. In general, more sophisticated methods such as imaging, provide more accuracy but are more expensive and frequently limited to laboratory settings. Field methods, such as calipers, are more readily used but are associated with up to 10% of interobserver error, making it challenging to use in multi-center studies (de Bruin et al., 1995a). In order to correctly interpret body composition data, it is critical to understand each method's underlying principles as well as its strengths and limitations. It is important to note that due to the relative novelty of intervention research in infants, the use of many of these measures has been minimal. Table 1 provides an overview of these assessment methods for infants.

Anthropometry and body dimensions

Anthropometry refers to the assessment of body measurements. Anthropometric measurements such as weight-for-length have been widely used in population-based studies (Byrne et al., 2016; Ogden et al., 2014; Perrin et al., 2015). In general, anthropometric assessments are relatively easy to administer, but methodological challenges related to whether this is a true

indication of an infants' health exist. Additionally, the accuracy of these measures may change throughout infancy thus making longitudinal studies challenging (Demerath and Fields, 2014).

Weight and length. Typical measurements to assess infants' growth and development include measuring infants' weight and length. These measurements are then plotted on gender-specific weight-for-age, length-for-age, weight-for-length, and/or body-mass-index-for-age growth charts to screen for health related concerns. The WHO growth charts released in 2006 are widely recommended for use as they represent the standards of how children should grow within optimal conditions (de Onis et al., 2012; McCormick et al., 2010). Data for these growth charts were collected in the WHO Multicentre Growth Reference Study from six different countries (Brazil, Ghana, India, Norway, Oman, United States of America) representing optimal nutrition (i.e., breastfeeding) and parental health behaviors (e.g., non-smokers).

Weight-for-length is the most common method used within research to assess infants' growth as well as those who are at-risk for obesity reflected as a weight-for-length percentile at or above the 95th percentile (Ulrich and Hauck, 2013; WHO, 2006). Recent research has begun to suggest the usefulness of also calculating body-mass-index as a measure of adiposity in infants along with potential future risk of obesity (Roy et al., 2015; Slining et al., 2013). For example, Roy and colleagues (2016) found children with a body-mass-index and weight-for-length percentile $\geq 85^{\text{th}}$ percentile at 2 months of age had an increased risk of obesity at age 2 years. A related but not as commonly used calculation is the ponderal index (PI), which is calculated using $\text{weight (g)} \times 100 / (\text{height, cm})^3$. The PI has also been reported to be a useful indicator of growth (Armangil et al., 2011; Holston et al., 2013). The strengths of these measures primarily lie in the ease of measurement requiring little equipment, and the fact that they can be performed in a laboratory or field setting. The challenges with these measures are that these measures alone should not be viewed as a diagnostic tool since they are not a direct measurement of body fat and should be used in conjunction with other measures to properly assess children's health (CDC, 2009).

Body circumferences. Another method of assessing anthropometry is body circumference. To assess body circumference, a tape measure is placed around specific anatomical sites. The most common circumference taken is head circumference. Similar to weight and length, head circumference is plotted on a gender-specific growth chart. Other common circumferences include chest, abdomen, mid-upper arm, mid-thigh, and mid-calf (Daly-Wofe et al., 2015; Demerath and Fields, 2014). While taking body circumference measurements such as the waist circumference is commonplace for adults, this has not been commonly utilized in infants and reference values for abdominal circumference in full-term infants are lacking (Meldere et al., 2015). The WHO has established gender-specific arm circumference-for-age growth standards and similar to collecting weight and length information it is easy to assess; however, the implications and relationship to obesity risk are not well understood (WHO et al., 2016b). This technique can occur in a laboratory or field setting.

Skinfolds. Skinfold thickness, a measure of subcutaneous fat depots, is generally in good agreement with whole-body fat mass (Schmelzle and Fusch, 2002). Skinfold thickness is measured by applying hand-held calipers at various body sites for a standardized time of three seconds (Demerath and Fields, 2014). When measuring infants, typically the number of sites ranges from 2 to 5 (Demerath and Fields, 2014; Dauncey et al., 1977; Schmelzle and Fusch, 2002). The location of these sites depends on the number of sites selected as well as on the participant's sex, and can include the triceps, biceps, suprailiac, subscapular, and quadricep

sites. There are several equations available for the conversion of skinfold thickness into body fat percentage, depending on the participant's sex, age and race for children, but few have been validated in infants 0-2 years or with diverse populations (Demerath and Fields, 2014). Nevertheless, skinfold assessments as a measure of body composition have been utilized in several recent studies (D'Angelo et al., 2015; Lakshman et al., 2015; McCloskey et al., 2016; West et al., 2013). Further, the WHO reference standards have been deemed the best for reference standards for infants >3 months of age (Miller, 2015). The advantages of this method is it requires only skinfold calipers, is non-invasive and can be used as both a laboratory or field technique. However, the method can cause the infant some discomfort, requires considerable training for the investigator, and for repeated measurements, it is recommended that they are conducted by the same individual as the interobserver error could alter findings (de Bruin et al., 1995a; de Bruin et al., 1995b). Also, estimates of fat mass using anthropometric equations has shown poor agreement with air displacement plethysmography (AD) and Dual-energy X-ray Absorptiometry (DXA or DEXA) in the first year (Cauble et al., 2017; Kulkarni et al., 2014).

Imaging methods

Advances in imaging technology have made it possible to expand body composition assessments to include the analysis of regional or segmental composition. The most commonly used technology is DXA, which has been used historically to assess two-dimensional bone mineral density. However, DXA has the capacity to differentiate fat tissue from lean tissue, and can be applied to assess body composition at the level of the whole-body as well as for regional assessments. As a result, DXA can be employed for assessing body composition utilizing a 3-compartment model (fat mass, bone mineral content, and lean mass). DXA assessment of bone mineral content is further required for a 4-compartment model (fat, mineral, water, and protein), which is currently considered the gold standard for whole-body body composition assessment (Demerath and Fields, 2014; Butte et al., 2000; Fomon et al., 1982). Although DXA involves radiation, overall x-ray exposure is minimal when compared to other procedures involving x-ray methodology and the assessment can be completed without sedation and with limited immobilization (Andres et al., 2011; Binkovitz and Henwood, 2007). DXA has shown to be a feasible method for assessing infants' body composition having been used in several cross-sectional and longitudinal studies (Binkovitz and Henwood, 2007; Butte et al., 2000; Goswami et al., 2016; Henche et al., 2008). Although DXA tends to be the most commonly used method to assess body composition in children, its use in infants has only been minimal (Zanini Rde et al., 2015). DXA scans have to be conducted in a laboratory setting, and require a well-trained or certified staff member to manage or run the DXA (Demerath and Fields, 2014). Further, for the most accurate results infants need to be as immobile as possible which represents a significant challenge.

Other imaging methods that have been used in infant research include computed tomography (CT) and magnetic resonance imaging (MRI), that allows for the assessment of regional body composition body fat distribution, internal fat deposition, and organ size with a high precision (Andres et al., 2011; Fields et al., 2015; Harrington et al., 2002). Both methods (CT & MRI) require sophisticated equipment, are time consuming, and in the case of CT result in significantly greater radiation exposure when compared to DXA. While MRIs do not include radiation exposure, movements by infants can cause issues with the image, and may be best used for infants younger than 6 months (Demerath and Fields, 2014). Both CT and MRI's are primarily conducted in a laboratory setting.

Densitometry

The underlying principle of densitometry is that the density of the body tissues varies depending on its chemical composition. Because hydrostatic is not feasible in infants because it requires extensive participant cooperation, the method of choice to assess densitometry in infants is air displacement plethysmography (ADP) (Ellis, 2000). ADP can be conducted with an infant-specific commercially available device, the PEA POD, which measures the volume of air that is displaced (Cosmed Srl, Rome, Italy) (McLeod et al., 2016; Moore et al., 2016; Ramei et al., 2016; Orlando et al., 2003). A recent systematic review identified the PEA POD as the most frequently used body composition method between the ages 0-2 (Zanini Rde et al., 2015). While additional research on the validity of ADP is needed, initial studies have found acceptable reliability and validity (Demerath and Fields, 2014; Ma et al., 2004; Roggero et al., 2012). Challenges of using this tool are that it does not allow for regional body composition assessment, equipment has a high cost, lack of portability as it can only be conducted in a laboratory setting, and is restricted by infant weight (<8 kg).

Bioelectrical impedance

Human tissues differ in their capacity to conduct an electric current. As such, body composition can be assessed by measuring the resistance (and reactance) to allowing a small, insensible electric current. More recent approaches involving the placement of multiple electrodes provide multisegmental information and quasi-regional distribution of body composition. Some studies suggest that bioimpedance analysis could have some advantages in terms of accuracy over skinfold measurements but still might contain substantial error due to infants' changing bodies, fluctuation of body fluids, and lack of appropriate reference values for different ethnicities (Demerath and Fields, 2014; Kyle et al., 2004; Lingwood et al., 2012; Savino et al., 2003). Furthermore, it may not accurately predict fat-free mass in all populations during the first few weeks of life (Tint et al., 2016). One of the major advantages of bioelectrical impedance is that the equipment is noninvasive, safe, cost efficient compared to imagery methods, portable, and the technique is frequently used in laboratory and field settings (Demerath and Fields, 2014; Kyle et al., 2004).

Other less frequently used techniques

Other techniques for the assessment of body composition that have been minimally used in research include dilution/tracer methods such as dilution of ^{18}O or deuterium (^2H) for the assessment of total body water as well as near infrared reactance and ultrasonography, both able to be used to assess body composition at specific sites (Tanabe et al., 2012; Bandara et al., 2015). The use of dilution/tracer methods in infants is challenging as it is time-consuming and dosing/collecting is problematic, but it is a portable approach for field studies (Demerath and Fields, 2014). However, a recent systematic review failed to identify a single study which used dilution methods to assess total body water in children (Zanini Rde et al., 2015). Current literature suggests ultrasonography has shown acceptable accuracy with MRI assessed internal-abdominal adiposity and subcutaneous-abdominal adiposity and is preferred by mothers due to the shorter duration and ability to stay with infants (compared to MRI); however, it has been used minimally in research (Al-Mohaimed et al., 2015).

Nutrient intake

The measurement of nutrient intake has the potential to assess micronutrient deficiencies and malnutrition that may be masked by macronutrient (particularly processed simple carbohydrates) excesses that can lead to obesity in early childhood and late infancy. Assessing the diets and nutrient intake of infants and children presents unique challenges. Dietary intake is difficult to measure, and any single method does not assess dietary exposure perfectly. While a gold standard method may be to weigh and measure an infants' diet, this is not feasible in a population of breastfeeding infants. Additionally, parents or caregivers must be used as a proxy in obtaining information for the child, and nutrient intake in breastfeeding infants presents unique challenges due to measurement difficulties and variation in breast milk composition within and between nursing mothers. Selection of a method is dependent upon the purpose of the study, the precision needed, the population of interest, the period of interest, and the available resources. The limitations of dietary assessment methods should be understood and assessment methods should be selected with caution. Importantly all of these methods could be used in a laboratory or field setting. Table 2 provides an overview of nutrient assessment methods.

Surveys and questionnaires

Many large studies of infant feeding practices have utilized survey instruments conducted either in person with the main caregiver, or mailed to the participants' home (Griffiths et al., 2009; Klag et al., 2015; Fein et al., 2008). These surveys and questionnaires can capture a variety of information, including infant feeding practices such as breast milk feeding practices and duration, formula use, and timing of solid food introduction. There are clear advantages and limitations to this type of methodology. These surveys can be designed to ask specific questions of interest, do not require large amounts of resources to analyze, and may impose the least amount of burden on the study participants. However, the data collected might not provide comprehensive nutritional information, such as absolute values of nutrients, and are subject to self-reporting bias (Ryan and Hay, 2016). Further, definitions of terms within surveys can also be a challenge when deciding whether to assess "breastfeeding." For example, "exclusive breastfeeding" could indicate either feeding an infant human milk alone through breast or bottle, or a combination of human milk and non-human milk items (Ryan and Hay, 2016).

24-hour recalls. With 24-hour dietary recalls, interviewers obtain information on all food and beverage items consumed during a defined 24-hour period. This methodology has been utilized in several large, cross-sectional studies (CDC, 2015b; Dwyer et al., 2003; Dwyer et al., 2003 (2); Hediger et al., 2000; Devaney et al., 2004; Ziegler et al., 2006; Tippett et al., 1999). Recalls are administered with parents face to face, over the telephone, or through an automated computer program with similar results. Depending on the nutrient in question, more than one 24-hour recall is often required to provide information on long-term intake, and recall interviews should be scheduled on various days of the week to account for daily variation in infants' food intake, particularly between weekdays and weekends, as well as seasonal effects on food intake. The 24-hour dietary recall method is labor intensive due to the facts that interviewers must be trained in order to assure that valid information is being collected, and responses must be coded for analysis by software programs. Other challenges include determining breast milk intake, and obtaining intake from multiple respondents who might have cared for the child on a given day. Regardless, parents are generally willing to respond to an interviewer. In addition,

parents are often familiar with their infant's schedule, and the amounts of beverages and jarred baby foods served, thereby improving the estimation of amounts consumed.

Food Frequency Questionnaires (FFQ). On a FFQ, respondents are asked to report their usual frequency of consumption of each food for a specific period of time. Some FFQs also inquire about the portion size of each food consumed or specify a standard portion size. In order to assess an infants' dietary consumption through a food-frequency checklist, modifications could be made to include questions about: 1) the frequency that the infants received foods from various food groups (e.g., breast milk, formula, fruits, vegetables, cereals); 2) vitamin, mineral, and herbal supplements given to infants; 3) formula feeding, and 4) breastfeeding.

FFQs have been utilized for the assessment of dietary patterns in children as young as 2 years of age (Lioret et al., 2015). More limited applications in FFQ administered in studies of infants include simply identifying if foods have been introduced to the diet or are consumed at all which is important as the early introduction of foods (at or before 4 months) may increase a child's risk for being overweight (Pearce et al., 2013). Advantages of the FFQ method for assessing the diets of infants are that they are relatively inexpensive; may reflect typical diet when weekly consumption is considered; may be used to screen high or low consumers of different foods; and may be designed to focus on certain foods, such as infant formula or foods commonly consumed only by infants (e.g., jarred baby foods). Limitations of this method are that FFQs do not provide specific nutrient information or estimates of absolute intakes unless serving sizes are considered, they cannot be used to measure milk intake from the breast, and they can be difficult for parents or caregivers to estimate the frequency of consumption of a single food item.

Diet records. Food records or food diaries require the parent or caregiver to weigh, measure, or estimate amounts and record all foods consumed over a specified period of time, usually 3 to 7 consecutive days. Diet records have also been used in the assessment of infant feeding, although this method may be the least frequently utilized (Heinig et al., 1993). Food records or diaries are often used in settings that require estimates of the food and nutrient intake of individual infants, as opposed to groups. The major strength of dietary records is that they do not rely on memory and, because parents or caregivers keep the records, they are relatively inexpensive to administer. Another advantage is that these can be collected electronically making it much easier for researchers to streamline the data. However, diet records have serious limitations: the particular time period may be atypical if the infant or child is sick or teething; dietary intake may be altered due to the record-keeping itself; recording intake might be considered too burdensome and time-consuming by respondents or parents especially for frequent feedings; and these records cannot be used to measure milk intake from the breast. In addition, food records entail the same resources for coding and processing as 24-hour dietary recalls (Ziegler et al., 2006).

Breastfeeding intake

While appropriate growth and development over time can assure of adequate intake to meet metabolic and growth needs, and a count of wet diapers and stools can be used to ensure hydration at a minimum, it can be difficult to accurately assess intake from an individual breastfeeding episode. One objective method for assessing daily milk intake is through deuterium oxide elimination, consisting of orally giving infants deuterium oxide and tracing its elimination from the body. However, this method has had minimal use, is expensive and requires extensive training for analysis (Fjeld et al., 1998). Another method to accomplish this is

with weights done pre and post feeding on an electronic scale to determine milk transfer amounts (Spatz and Edwards, 2016). This approach is recommended as opposed to evaluating time and quality of breastfeeding since the former methods can be subjective and transfer of milk is variable under similar assessments. Due to the challenges of breastfeeding, the measurement of additional variables related to breastfeeding may provide a more complete picture of breastfeeding intake. The use of “on demand” breastfeeding and infant temperament may be a variable of interest as infant temperament has been associated with body composition (Wells et al., 1997). Further, assessing the use of “mixed” feeding practices (formula and breastmilk) as well as the timing and frequency of the introduction of solid foods can provide a more accurate representation of breastfeeding intake. However, future research efforts are needed to improve the accuracy of breastfeeding intake in these contexts.

Energy Expenditure

A limited number of researchers have studied actual total energy expenditure and infant weight. Total energy expenditure is an important variable when examining energy balance. Human energy expenditure can be divided into four primary processes: resting metabolic rate (RMR), growth, the thermic effect of food (TEF), and physical activity. RMR refers to the amount of energy that is required to support all biological functions that are maintained in a resting state and include vital functions such as thermogenesis, organ function, cellular homeostasis, and immune function. RMR is largely determined by the amount of energy that is required by the main metabolic organs. In infants, the brain and the liver contribute to a majority of the whole-body energy expenditure (58-70%) (Butte, 2005; Eila, 1992). Although skeletal muscle makes up about 25% of an infant's body weight, it is metabolically much less active (13 kcal/kg/day) when compared to the brain or inner organs (200-440 kcal/kg/day), so that the overall contribution of skeletal muscle mass is small (6%) (Eila, 1992). Growth is another metabolic process that requires energy for the energy expended during anabolic processes. Additional energy is stored due to the retention of protein and fat (Barnerss, 1993). The proportion of energy deposition vary with age and have been reported to be ~35% at 1 month of age, decreasing to 3% at one year of age (Butte, 2005; Butte, 2015). The TEF relates to the amount of energy that is required for the digestion, absorption, transportation, and utilization of nutrients from the diet. In infants, the TEF is estimated to be approximately 10% of dietary energy intake (Barnerss, 1993). Any form of physical activity requires energy, whether unplanned or planned. The amount of energy expended during physical activity is determined by the weight of the infant and type of activity as well as the duration and relative intensity of the activity.

There are five main areas of assessment related to energy expenditure including calorimetry and physical activity assessment. An overview of these assessment methods is provided in Table 3.

Indirect calorimetry

Because ultimately all energy expended by human metabolism is derived from aerobic processes, energy expenditure is directly related to oxygen consumption and carbon dioxide production, with respiratory gas analysis serving as the gold standard for in-vivo energy expenditure. With infants, respiratory gas can be collected for limited periods using Douglas bags, or more recently using metabolic carts, often referred to as indirect calorimetry (Berger et al., 2014). Specifically with infants, indirect calorimetry can be used to assess RMR under resting conditions, when respiratory gases are collected using a ventilated hood within an

incubator or weaning basket (Keidar et al., 2014). Using the same principle, energy expenditure can also be assessed during non-resting activities using a mouthpiece or facemask, although specific fitting is required to use this technique in infants and requires more testing to determine its efficacy (McGuinness and Childs, 1992). While indirect calorimetry in infants can provide accurate assessments, these methods have primarily been used in studies with pre-mature infants and are mostly limited to high-end research laboratories or clinical settings, thus additional research is needed in this area.

The doubly-labelled water method presents an alternative to utilize an indirect calorimetric approach to assess total energy expenditure in the field. Following the administration of stable isotopes of hydrogen and oxygen (^2H and ^{18}O), carbon dioxide production can be assessed continuously by measuring the exponential loss of ^{18}O , and O_2 consumption and subsequently energy expenditure can be estimated using available literature data (International Dietary Energy Consultancy Group, 1990). While the doubly-labelled water method is considered the method of choice for the assessment of total energy expenditure in the field, it is expensive, requires time-consuming stable isotope analyses, has been used minimally in infants, and does not differentiate between different activities (Guilfooy et al., 2008; Schoeller, 2008).

Physical activity assessment

The assessment of physical activity, the behavior resulting in metabolic energy expenditure, presents a viable alternative to estimate human energy expenditure. Historically, physical activity has been assessed via parent-report, but more recently, highly-sensitive physical activity sensors have been used to objectively assess physical activity. These measures can be implemented in laboratory or field settings.

Questionnaires. Questionnaires relating to infant activity level have primarily been developed for investigating temperament or psychobiological concerns and not for examining activity alone (Gartstein and Rothbart, 2003; Johnson et al., 2016; Worobey, 2014). For example, the Rothbart Infant Behavior Questionnaire includes questions related to activity levels. Other research has focused on specific behaviors such as “tummy time” or time spent in restrictive devices such as strollers or car seats (Moir et al., 2016). Strengths of questionnaires include ease of dissemination in both laboratory and field settings as well as the low-cost of materials. A challenge of questionnaires is the accuracy of the self-report nature of these measures.

Interviews and Focus Groups. Interviews and focus groups have also been utilized to assess parent’s perception of their infant’s physical activity (Jimenez-Cruz et al., 2010; Pesch et al., 2015; Redsell et al., 2010). Often questions about activity were a secondary interest of study to the overall studies goal of examining parent perceptions of weight or infant feeding practices. A strength of this type of measure is that it provides important information related to how parent’s perceptions may influence the promotion of physical activity within the home and can be conducted in field or laboratory settings. However, for all of these measures parents’ perception of an “active” child varies, limited studies indicate correlation with objective measures of activity, and it remains difficult to generalize findings to other populations (Jimenez-Cruz et al., 2010; Tulve et al., 2007).

Direct observation. Direct observation of parent and child interaction is another viable method for assessing infant activity. During observations, trained research personnel observe parents and their infants in their homes or within a laboratory setting either in-person or by video

(Cherney et al., 2003; Conner et al., 2014; Rothbart, 1986). Observational methods have been minimally used within research studies, especially for the purpose of monitoring activity (Worobey, 2014). While observations minimize parent bias in terms of self-report, collecting, coding, and analyzing data is time intensive and costly. This form of assessment could be conducted in either the laboratory or field setting.

Physical activity sensors. A growing area of research in infancy is the use of physical activity sensors to obtain an objective measure of activity within the laboratory and field settings (Moir et al., 2016; Galland et al., 2016; Heale et al., 2015; Jensen et al., 2015). This approach has become extremely popular and user-friendly in recent years. Physical activity sensors can be worn as a watch-like device or sewn within clothing (i.e., socks, wristbands) to make them more comfortable for use with infants. Previous research has found that infants and caregivers can comply with wear protocol and when used in conjunction with an activity diary, is a reliable assessment of infant activity (Tulve et al., 2007). The strength of using these sensors is the objective information they can provide. However, the calibration and validity of physical activity sensors remain the primary challenges for the assessment of energy expenditure in this population. For example, a specific challenge is accounting for parent-child interaction within the data as a parent carrying an infant or the infant being in a swing could incorrectly identify infants as active. While time activity diaries can help to account for some of this information, the exact time of when parents complete the diary (periodically throughout the day vs. end of day) could result in less accurate findings. More research studies are needed to better understand the influence of parent-infant interaction in order for sensors to be more widely used within intervention studies.

Conclusion

The overall purpose of this paper was to provide a broad overview of the assessment tools available for use in obesity-related research in full-term infants. As the interest in infant obesity research expands, the interest of new researchers to the field as well as interdisciplinary teams will continue to grow. Additionally, in order to best address obesity it is imperative to bring together multiple disciplines through interdisciplinary research collaborations that can provide a dynamic and comprehensive perspective. Such collaborations could include pediatricians, pediatric nurses, dietitians, behavioral psychologists, physical activity specialists, and dentists among others. This paper provides a summary of the literature, especially for advanced researchers. For a more in-depth review of related assessments and method references please refer to Tables 1-3.

Body composition, nutrient intake and energy expenditure methods for infants age 0-2 years, have advantages and disadvantages, thus each method should be selected with caution. Researchers should take into account a multitude of factors prior to selecting an assessment method. Researchers must consider the feasibility of implementing the method (laboratory vs. field), along with the cost of the method – both financially and the amount of time an infant/caregiver will be willing to participate. Intuitively researchers should not only consider the accuracy of the measure but also consider the sensitivity to change, especially for longitudinal studies. Further, it is also important to make a measurement decision based on the experience of the research investigators and availability of measures at their institution. Researchers new to the infant assessment field may not have the skill necessary to perform several of the tests, access/funding for expensive equipment (e.g., PEA POD), and/or infants may become tired and irritable with long assessment sessions. As such, decisions on assessment methods need to be carefully considered. Regardless, researchers should seek collaborators skilled in the area who

can at least provide training. Future research should focus on validating existing methods of evaluation of body composition, nutrient intake and energy expenditure in order to determine which ones can be used reliably in studies to reduce obesity levels for children younger than 2 years of age.

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Table 1. Overview of Anthropometry and Body Composition Assessments for Infants

Method	Advantages	Disadvantages
Weight and Length (e.g., weight-for-length, body-mass-index)	<ul style="list-style-type: none"> Minimal equipment needed Applicable in a variety of settings 	<ul style="list-style-type: none"> Not a direct measure of body composition
Body Circumferences	<ul style="list-style-type: none"> Ease of measurement Minimal equipment needed 	<ul style="list-style-type: none"> Not widely used in infants Reference values for infants are lacking
Skinfolds	<ul style="list-style-type: none"> Minimal equipment Noninvasive 	<ul style="list-style-type: none"> Investigator must be trained Limited validation of conversion equations in diverse populations
Dual energy X-ray absorptiometry (DXA)	<ul style="list-style-type: none"> Able to differentiate fat tissue from lean tissue Applicable for regional and whole body assessments 	<ul style="list-style-type: none"> Radiation exposure, although minimal Lack of availability due to high cost Limited use in previous research
Computed tomography (CT)	<ul style="list-style-type: none"> Regional body assessment Accurately assesses body composition, body fat distribution, internal fat deposition and organ size 	<ul style="list-style-type: none"> Expensive Time consuming procedures High radiation exposure
Magnetic Resonance Imaging (MRI)	<ul style="list-style-type: none"> Regional body assessment Accurately assesses body composition, body fat distribution, internal fat deposition and organ size 	<ul style="list-style-type: none"> Expensive Time consuming procedures Inaccuracy among infants <6 months of age
Air displacement plethysmography (ADP)	<ul style="list-style-type: none"> No radiation exposure Commercial device available (PEA POD) Accurate and reliable measure of fat mass and fat free mass 	<ul style="list-style-type: none"> Expensive Lack of portability
Bioelectrical Impedance (BIA)	<ul style="list-style-type: none"> Noninvasive Portable Utilized in clinical settings 	<ul style="list-style-type: none"> Inaccuracy concerns in infants
Dilution (deuterium or ¹⁸ O)	<ul style="list-style-type: none"> Applicable for total body water and hydration assessment 	<ul style="list-style-type: none"> Associated with accuracy errors Not utilized in clinical or applied settings
Ultrasonography	<ul style="list-style-type: none"> Acceptable accuracy Preferred by mothers to MRIs 	<ul style="list-style-type: none"> Limited use in previous research

For additional in-depth review of related assessment methods please see the following studies:

- E. W. Demerath and D. A. Fields, "Body composition assessment in the infant," *American Journal of Human Biology*, vol. 26, no. 3, pp. 291-304, 2014.
- K. J. Ellis "Evaluation of body composition in neonates and infants," *Seminars in Fetal and Neonatal Medicine*, vol. 12, pp. 87-91, 2007.
- S. Heymsfield, T. Lohman, Z. M. Wang, and S. Going (eds). *Human Body Composition 2nd Edition*, Human Kinetics, Champaign, IL, 2005.

Table 2. Overview of Nutrient Intake Assessment for Infants

Method	Advantages	Disadvantages
Nutrient Intake		
Surveys & Questionnaires (in general)	<ul style="list-style-type: none"> • Ability to ask specific questions • Not time intensive • Minimal participant burden 	<ul style="list-style-type: none"> • Data may not be comprehensive • Limited data collection availability • Self-report bias
24-hour recalls	<ul style="list-style-type: none"> • Willing participation • Estimation accuracy when utilizing parents for recall 	<ul style="list-style-type: none"> • Labor intensive for researchers • Researchers must be trained • Need for analysis software
Food Frequency Questionnaires (FFQ)	<ul style="list-style-type: none"> • Inexpensive • Provides information on food being consumed • Can be designed to focus on specific foods 	<ul style="list-style-type: none"> • No specific nutrient information or absolute intake estimates provided • Unable to measure milk intake from breast • Caregiver estimation inaccuracies
Diet Records	<ul style="list-style-type: none"> • Non-reliance on memory • Inexpensive • Ability to focus on specific foods 	<ul style="list-style-type: none"> • Burdensome for participant and researcher • Participant motivation and high literacy required • Potential for atypical time periods • Unable to measure milk intake from breast
Breastfeeding Intake	<ul style="list-style-type: none"> • Minimal equipment required (i.e., scale) 	<ul style="list-style-type: none"> • Analysis software required • Accuracy • Variation in energy density of breastmilk
<p>For additional in-depth review of related assessment methods please see the following studies:</p> <ul style="list-style-type: none"> • V. Kipnis, D. Midthune, L. Freedman, S. Bingham, N. E. Day, E. Riboli, P. Ferrari, and R. J. Carroll. "Bias in dietary-report instruments and its implications for nutritional epidemiology," <i>Public Health Nutrition</i>, vol. 5, no. 6A, pp. 915-23, 2002. • S.A. Bingham. "A dietary assessment of individuals, accuracy, new techniques, and recommendations" <i>Nutritional Abstracts and Reviews</i>, vol.57, pp. 705-42, 1987. • A. S. Ryan and W.W. Hay. "Challenges of nutrition research: A commentary," <i>Nutrition Journal</i>, vol. 15, pp. 42, 2015. 		

Table 3. Overview of Energy Expenditure Assessment for Infants

Method	Advantages	Disadvantages
Energy Expenditure		
Indirect Calorimetry	<ul style="list-style-type: none"> • Comprehensive assessment • Accuracy 	<ul style="list-style-type: none"> • Only available for high end research applications • Expensive
Doubly-labelled Water	<ul style="list-style-type: none"> • Safe for infants • Accurate 	<ul style="list-style-type: none"> • Expensive • Time-consuming • Does not differentiate between activities
Physical Activity Assessments Questionnaires	<ul style="list-style-type: none"> • Provides parents perceptions • Easy to administer • Inexpensive 	<ul style="list-style-type: none"> • Subjective findings • Difficult to generalize to populations
Direct Observations	<ul style="list-style-type: none"> • Minimizes parent bias 	<ul style="list-style-type: none"> • Expensive data collection and analysis • Time consuming
Physical Activity Sensors	<ul style="list-style-type: none"> • Objective measure 	<ul style="list-style-type: none"> • Calibration and validity of sensors has not fully tested • Expensive • Confounding variables (e.g., carrying child) • Energy costs of activities for infants is largely unknown

For additional in-depth review of related assessment methods please see the following studies:

- E. Jéquier, K. Acheson, and Y. Schutz. "Assessment of energy expenditure and fuel utilization in man," *Annual Review of Nutrition*, vol. 7, pp. 187-208, 1987.
- J. Worobey, "Physical activity in infancy: developmental aspects, measurement, and importance," *The American Journal of Clinical Nutrition*, vol. 99, no. 3, pp. 729S-733S, 2014.
- D. A. Ulrich and J. L. Hauck, "Programming Physical Activity in Young Infants At-Risk for Early Onset of Obesity," *Kinesiology Review*, vol. 2013, no. 2, pp. 221-232, 2013.